Evaluation of mini-cuttings as a propagation system for *Eucalyptus* hybrids©

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Abstract

Clonal asexual propagation by cuttings is an efficient technique for capturing genetic gain in forestry. However, selected clones (selected for growth, wood properties and stem form) often prove to be difficult to root, thereby limiting the rate of deployment for further field testing and subsequent commercialization. This constraint will also delay the time taken for new clones to be identified. It is thus imperative that a propagation system runs efficiently and economically to realize genetic gain. It is widely hypothesized that rooting ability of clones is under genetic control. Although true for some clones, this study showed that the sand bed mini-hedge system resulted in improved rooting percentages through rejuvenation, better nutrition and improved climatic control of hedges. Additional benefits of this system included a more robust root system, faster growth and improved plant quality of mini-cuttings, which are favourable traits to reduce transplant stress when planted in-field.

INTRODUCTION

According to Stape et al. (2001), de Assis et al. (2004), and Titon et al. (2006) the following observations can be made regarding clonal asexual propagation on Eucalyptus:

- Clonal propagation is an efficient technique to capture genetic gain.
- The inability to root is often a constraint to the deployment of some clones.
- Three factors are crucial in the rooting success of *Eucalyptus*:
  - Condition of the mother plant
  - Rooting environment conditions
  - Genetic disposition

CONVENTIONAL MACRO-CUTTING AND MINI-CUTTING PROPAGATION

Macro-cutting propagation

1. Conventional vegetative propagation using macro-cuttings in the open (Figure 1).

- Hedges in the ground, widely-spaced (clone bank)
- Semi-lignified coppice harvested
- Cuttings set (8 to 10 cm)

2. Limitations of conventional vegetative propagation approach.

- Controlling hedge nutrition
- Climatic extremes
- Maintaining juvenility

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Figure 1. Conventional vegetative propagation using macro-cuttings from hedges grown in the open.

Mini-cuttings

1. Characteristics of mini-cutting.

- Mini-hedges in sand beds under cover (Figure 2)
- Stock plants are closely-spaced
- Herbaceous coppice harvested
- Daily irrigation and nutrient supply
- Smaller cuttings (4 to 7 cm)

2. Expected outcomes of the mini-cutting approach.

- Good hedge nutrition results in better rooting
- Hedges sheltered from climatic extremes
- Cuttings retain their juvenility

Figure 2. Mini hedges in sand beds under cover.

AIM AND OBJECTIVES

- To measure hedge productivity between the macro-and mini-hedge methods
- To compare rooting from mini-hedges with macro-hedges
- To compare plant quality and field survival

MATERIALS AND METHODS

Planting procedure

- Six clones [SG (*E. smithii* × *E. grandis*), NG (*E. nitens* × *E. grandis*), GU (*E. grandis* × *E. urophylla*)] spanning three taxa planted into sand beds (Figure 3).
  - o Temperate hybrids (alternative to *E. nitens*)
  - o Sub-tropical (alternative to *E. grandis*)
- A layer of stone was first placed in the bed followed by washed, sieved river sand.
- Hedges were planted at approximately 10×15 cm and irrigated using drippers.

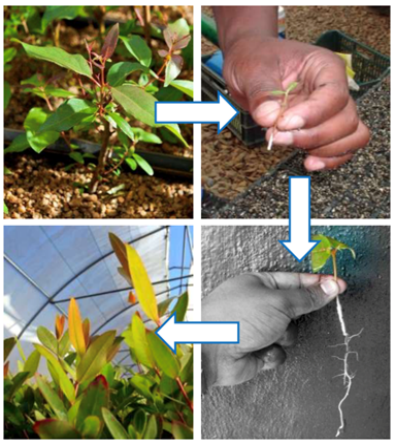

Figure 3. Planting procedure.

Trial analysis

The trial was designed and analysed according to the following model:

$Y_{ijk} = u + taxa_i + propagation\ system_j + (taxa * propagation\ system)_{ij} + \sum_{ijk}$

where:

- y = parameter of interest (productivity, rooting, plant quality, field survival)
- μ = overall mean
- taxai = fixed taxa effect (n = 3)
- propagation systemj = fixed propagation effect (macro or mini)
- Taxa * propagation system = factor interaction
- Σ = random error associated with the i^{th} taxon, the j^{th} propagation system and the k^{th} plant

Data were collected over a period of 3 years.

RESULTS

Hedge productivity of the clone GU (*E. grandis* × *E. urophylla*)

Number of cuttings per hedge per harvest, number of hedges m^{-2}, and number of cuttings m^{-2} for clone GU (*E. grandis* × *E. urophylla*) are shown in Figures 4 and 5. Measuring hedges per square meter and number of cuttings per square meter is based on macro-hedge spacing = 0.6×0.8 m: 2 × hedges m^{-2} and 24 × cuttings m^{-2} and mini-hedge spacing = 0.10×0.15 m: 66 × hedges m^{-2} and 264 × cuttings m^{-2}. Mini hedges offer an 11-fold increase in cuttings m^{-2}.

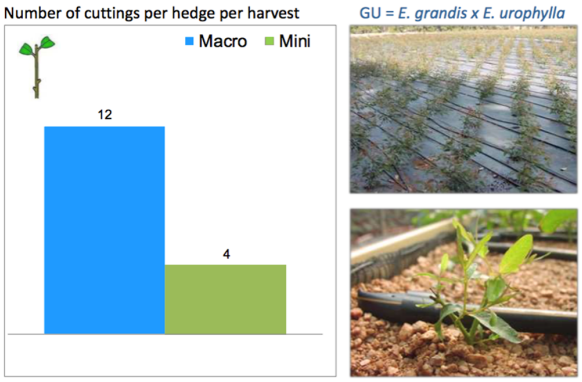

Figure 4. Number of cuttings per hedge per harvest, macro vs. mini cuttings for GU (*E. grandis* × *E. urophylla*).

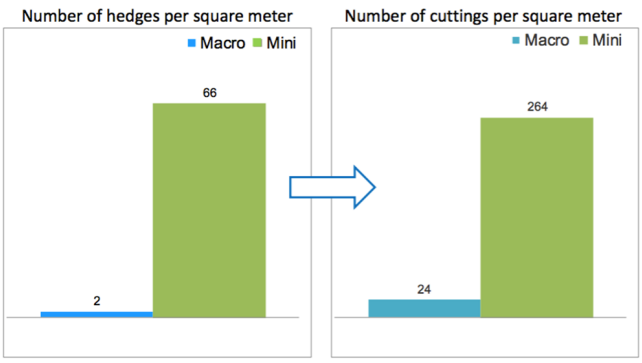

Figure 5. Number of hedges and number of cuttings m^{-2}.

Percent rooting results for the three clones SG (*E. smithii* × *E. grandis*), NG (*E. nitens* × *E. grandis*), GU (*E. grandis* × *E. urophylla*)

Percent rooting was significant for clones NG and GU (Figure 6).

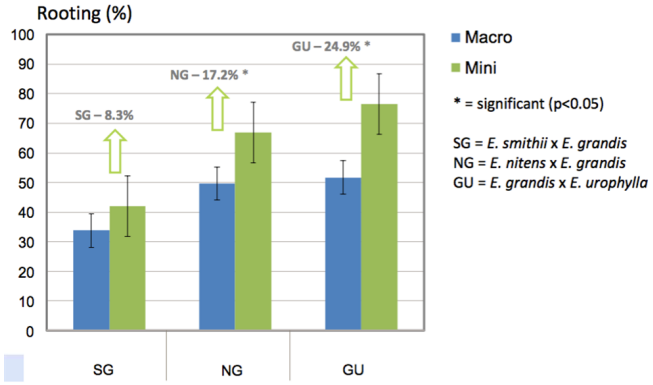

Figure 6. Rooting results for the three clones: SG, NG, and GU.

Root quality at 6 weeks of the clone GU (*E. grandis* × *E. urophylla*)

Macro-cutting vs mini-cutting (Figure 7).

- Cumulative root length (mm): Macro = 20, Mini = 246
- Root dry mass (mg): Macro ≈ 0, Mini = 55
- Shoot dry mass (g): Macro = 0.75, Mini = 1.00

Figure 7. Macro cutting vs. mini cutting.

Plant quality at 12 weeks for the three clones SG (*E. smithii* × *E. grandis*), NG (*E. nitens* × *E. grandis*), GU (*E. grandis* × *E. urophylla*)

New shoot height (cm) for macro and mini clones showed the greatest gains for clone GU and only clone SG was not significant (Figure 8). Dry mass (g) is shown in Figure 9 with root dry mass on the left two columns for each clone and the right two columns for shoot dry mass for each clone.

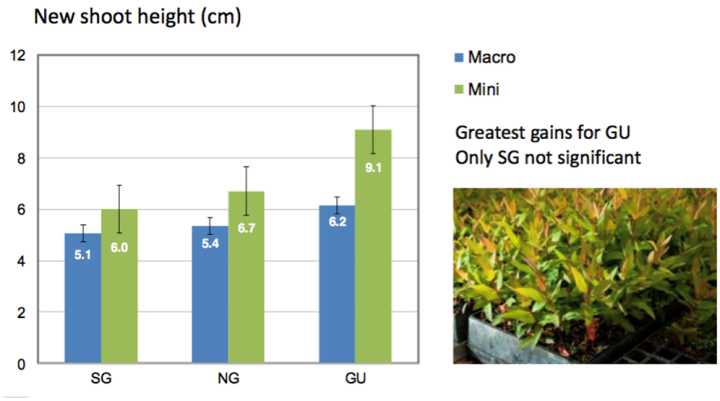

Figure 8. New shoot height (cm) for macro and mini clones.

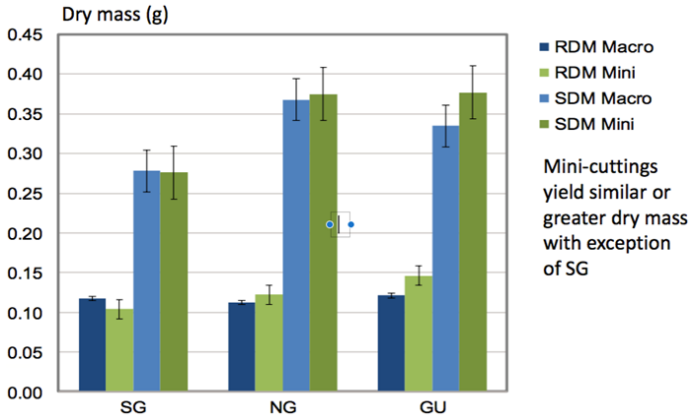

Figure 9. Dry mass (g) for the three clones.

CONCLUSION

Mini-cuttings offer many benefits:

- More juvenile, herbaceous cuttings.
- Improved control over hedge environment.
- Better productivity per square meter allows for intensive management over a small area.
- The superior rooting success results in better nursery efficiencies.
- Higher quality root systems.
- Increased rooting speed contributes to optimizing nursery capacity.
- Better plant quality results in better initial field performance.

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Current nursery industry issues in Australia©

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THE BIG PICTURE

- The Australian economy is being driven by mining boom time carry over, population /immigration growth and government spending (mostly not sustainable).
- Retail plant volume growth is through Bunnings and Masters chain stores and underpinned by strong housing development.
- Smaller retailers surviving by e-commerce and diversification of products and services.
- Endemic plant demand strong due to swing back to natives and environmental impacts and offsets.

AUSTRALIAN NURSERY ISSUES

- Contract growing terms being documented by nurseries working in conjunction with each other.
- A contraction in the number of production nurseries as owners retire or businesses close or fail.
- Some remaining nurseries expanding to pick up closure volumes.
- Nursery *Phytophthora* control is a growing issue with potential threats from new dieback species and demands for testing by clients.
- Restrictions on propagation of threatened flora remaining an issue with attempts to lift ongoing.
- Increased diversification: landscapers and revegetation contractors becoming growers and visa versa.
- Rental of plants and plant displays showing growth potential.

AUSTRALIAN CONSERVATION AND ENVIRONMENTAL ISSUES

- Climate variability influencing policy and practice.
- Miners under pressure to restore damaged areas. Retrospective funds established.
- New top level protocols being established for native seed, nursery hygiene and environmental restoration, driven by industry groups, Revegetation Industry Association of Western Australia Inc (RIAWA) and Society for Ecological Restoration Australasia Inc (SERA).
- Fire-fuel load management pervading all development assessments and decisions.
- Serious concerns over *Phytophthora* spp. and spread of myrtle rust.

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Lets' not re-invent the wheel: simple tools for a tree nursery©

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INTRODUCTION

My talk is about some simple tools or solutions to everyday jobs that we use with our business. These are used in combination with other techniques to grow our crops and are in themselves not complicated. They are easy to make, and may save yourself a bit of time, or ease the task by making it a little easier on yourself and your body.

Marie and I operate our small nursery, The Tree Farm, here in the Waimea Plains in Nelson. We grow mainly deciduous trees and shrubs and a small amount of natives mainly in open ground seed beds. The total area we use is less than a hectare, and our production numbers are small. The range of items we grow is mainly 1- or 2-year-old trees or shrubs, through to topiary and budded or grafted lines.

The aspects that I will discuss are in the broad categories of:

- A tool frame for working over raised seedbeds.
- Transplanting equipment – a simple tool for transplanting plants into plugs.
- Weed control – soil solarization – a simple technique for weed control prior to planting.

Tool frame

We grow 95% our crops in the open ground, and the daily routine involves a lot of bending or kneeling to tend to our crops at ground level. The plants are grown in raised seed beds and we take a lot of care to not stand or compact the seed bed in any way during the process of growing any crop.

As an apprentice, Eric Appleton the founder of Appleton's Tree Nursery in Nelson, explained to me on my 1st week of work that "You need to treat a seedbed the same way as your wedding bed… You keep your boots off them!!" After being a little bemused at this statement, I really like the message behind it. First being, that we have an underlying respect for the soil that we grow our crops in. And secondly, we also have respect for the plants that we are growing. With this in mind, when we had the opportunity to develop our own business, I have been able to make a simple tool frame for ourselves that has made life easier.

Figure 1. Tool frame with height adjustable seat.

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The tool frame has made daily tasks such as transplanting, weeding, and thinning a breeze. In terms of transplanting seedlings or plug grown plants we are able to transplant 300 to 400 plants per hour, which for our scale of business is appropriate.

Figure 2. Thinning seedlings using the tool frame.

Now I realise this is at the low end of the technology scale, but still if you need to transplant, sow, thin, weed, and you need to physically get your body low to the ground – or in our case, just over the top of the seed bed, then this makes life a lot easier. If you have had a previous career shearing or crutching sheep then no problem – you can probably get around like a bent fish hook all day, no trouble at all. But if you are like me – 185 cm or 6 ft 1 in. in height, and you have a day or two or three transplanting, thinning or weeding in front of you, then this tool frame means getting dressed the following morning is a breeze. You don't need to lasso your socks on the next day!!

The tool frame is made from recycled 26-in. bicycle wheels and pipe steel. It is height adjustable from 150 mm above the seedbed to up to 400 mm above the seed bed. We can simply hook bins on it to hold transplants or weeds, and the frame is easily pushed up and down the seed row when sitting on the seat, which is a plank of timber.

TRANSPLANTING EQUIPMENT – BARE ROOTED PLANTS TO PLUGS

Open ground production means we broadcast seeds on a seed bed. After germination if thinning is needed, we have the potential to save thinned seedlings and transplant these into plug cells. These can be grown on and lined out for alternative uses for ourselves. Plug technology is well known and used in a range of ways in the industry; this simple template allows us to get the seedlings transplanted into a cell easily, without the hassle of other cells filling with potting mix.

Figure 3. Plug transplanter with BCC 81 Plug Tray.

The Plug transplanter is placed on the tray of cells. Each adjoining cell is presented as an empty cell by means of a sliding shutter. The seedling is placed inside the cell and potting mix is filled around the seedling, while the transplanter keeps the potting mix from filling the other surrounding cells. Once this is finished, slide the shutter to the next cell and repeat.

Figure 4. Planting cuttings.

As a row of cells is completed, pull the transplanter back on row of cells and repeat. The cells we use are BCC81s or Lannen 63s, but you could make this to fit any cell tray.

Figure 5. Side view of transplanter.

The plug transplanter is made out of light tin, 6-mm steel rod (which the sliding panel slides along) a pair of tin snips and a pair of pliers, drill, and a vice or clamps to bend tin with. It is easily made in ½ an hour or so and easily used by left or right handed people. With the use of this tool, we are able to transplant 1500 to 2000 seedlings to plug trays per person each day, which again for our level of production is acceptable.

SOIL SOLARIZATION – SOLAR POWERED WEED CONTROL

The land we lease is quite a weedy block of land, previously used to grow vegetables and pasture for livestock. We add 2 m^3 of composted bark each season to each of the seed beds which are 50 m long by 1 m wide. The compost is in turn rotary hoed into the seedbed as it is formed prior to seed sowing and this application helps greatly with soil structure and makes for a more friable soil longer term. As a rule of thumb, when we are sowing our spring crops we generally have a period of 7 to 10 days after forming the seedbed to the first

germination of weed seeds. Weeds that we need to control include fathen, amaranthus, dock, mallow, clover, and dandelion. These in turn can match the germination of seed crops that we have planted. Crops such as *Robinia pseudoacacia* or *Pyrus callerayana* can easily germinate in conjunction with the weed seed crop, and then the dilemma is what course of action to take, to rescue the potential seedling tree crop. Mechanical weeding can be destructive to the seedlings, contact herbicides can be the same, and hand weeding although it can be thorough – does take time.

We have experimented with soil solarization for a couple of seasons, and it has proved to be a very good tool to have in our spring sowing tool box as the results have been impressive.

The seedbeds are formed as we would normally; they are then irrigated to field capacity. The seedbeds are covered with a light weight clear plastic and weighted down every few meters or so, to stop it blowing away.

Figure 6. Clear plastic on seedbeds.

The resulting temperature gain underneath the plastic rises very quickly to 50 to 60°C in our early summer days. With the plastic on the seedbed for 10 days or so, we remove, and sow our tree crops into the seed bed. The results to date have been very good, and although we still need to go and hand weed through our tree crop to take out the occasional weed, 90% of the weed crop is fried off by the heat generated underneath the plastic.

Figure 7. Left row: untreated seedbed; middle seedbed: germinating *Robinia pseudoacacia* with minor weed germination.

Now this is nothing new in terms of technique. The University of California has done a lot of research work in the past with this and my understanding is they are looking at it again with regard to strawberry production. The research recommends keeping the plastic cover on for 6 weeks. Our 10-day cycle would seem to be too short in theory, but the practice has given us good results to date. Fathen, amaranthus, dock, and mallow weeds were almost completely taken out by the 10-day roasting, Clover did still germinate in lesser amounts, and dandelion seed blew in from neighbouring fields during the summer, and germinated in the alleyways.

Figure 8. *Robinia pseudoacacia* at the end of growing season.

The resulting tree crops required only minimal amounts of hand weeding. It is a very simple and cheap technique to use and we can capture and utilise the sun's rays for the price of reusable plastic – that is money for jam!! We can roll out the plastic with a simple cloth unwinder, and wind the plastic up again with our cloth winder, that we use for our artificial crop covers.

SUMMARY

These tools haven't been expensive or complicated to make or use. As one of my previous employers used to say – "There is always a simple solution to all problems we face," and another former boss was fond of saying – "Let's not re-invent the wheel, somebody already has!!"

I hope some of these simple solutions that we use are able to solve some or your own production problems.

Semi-selective herbicide use in nursery weed control©

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SEMI-SELECTIVE HERBICIDE USE

Definition

The use of non-selective knockdowns at ultra-low concentrations to control weeds and to avoid off-target damage in bushland and nursery situations. This presentation is our introduction of this concept to nursery weed control.

Background

A considerable body of science in the use of semi-selective herbicide use has been developed by scientists and practitioners in Western Australia to combat particular environmental weeds in quality bushland.

The intention has been to find effective weed controls using herbicides without off-target damage. This work over many years has led to the development of very successful techniques which may have application to nursery weed control.

Products

The following are some of the knockdown herbicides that are currently being used in semi selective mode with Western Australia bushland; these are permitted for off label uses in Western Australia.

- Metsulphuron (Brush Off®) Du Pont
- Triasulfuron (Logran®) Syngenta
- Clopyralid (Lontrel®) Dow Agrosciences
- Halosulfuron (Sempra®) Nufarm
- Haloxyfop (Verdict™) Dow Agrosciences

New Zealand studies

I could find only one reference to herbicide use in semi-selective mode – Metsulphuron for use on Onehunga weed (*Soliva sessilis*) control on golf courses (Massey University, 2014).

New Zealand herbicide brand name match:

- Metsulphuron: Associate® 600 WDG (Nufarm), Agpro Meturon® (Agpro), Eradicate 600 (Ravensdown), Escort® (E. I. du Pont de Nemours), Matrix™ (Orion Crop Protection), Mustang® (Orion Crop Protection).
- Triasulfuron: Titan (Genfarm).
- Clopyralid: Versatill™ (Dow Agro Sciences).
- Halosulfuron: EnviroMax® (Nufarm).
- Haloxyfop: Hurricane® (Orion Crop Protection), Ignite™ (Zelam).

Overview of trials

- Determine if control could be achieved without off target damage.
- Which chemical would provide best overall results and which was best for particular weeds.
- If mortality was not achieved, was it possible to prevent weed-seed set.

Preparation and application

The following is a guide for nursery application:

- Accurate measurements by weight critical

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- Use clean filtered water
- Granular herbicides – use warm water to aid dissolution
- Waiting period for watering will apply
- Avoid spraying on warm days
- Mix in 20-L volume and dispense to smaller units
- Apply to strong plants
- Apply once, avoid double spray
- Target weeds as best possible

Trial outline

- Various application rates and mixtures were trialled on individual plants, including combinations of two herbicides given their compatibility.
- Nine species of Perth natives chosen for weed treatment
- Settled on the following:
    - Triasulphuron at rate 12 g 20 L^{-1}
    - Metsulphuron at rate 6 g 20 L^{-1}
    - A 50:50 (v/v) combo of above

Weed species targeted for control

Table 1. Weeds targeted.

Scientific name	Common name
Cardamine hirsuta	Flick weed
Chamaesyce species	Asthma weed, cats hair
Gnaphalium species	Cudweed
Oxalis species	Wood sorrels
Sagina procumbens	Pearlwort
Marchantii polymorpha	Liverwort
bryophyte	Mosses

Results for Logran

- Effects in place within 1 to 2 days for cudweed and flick weed.
- Cud weed species were heavily affected; within a week most wilted off.
- Stunted and discolouration of *Oxalis* species; weeds left in an inferior state, roots and stems still in place with leaves wilted off.
- Liverworts and sponge-like moss displayed changes by the 2nd week and treatment appeared to be effective.
- No abnormal changes in grass-like moss (pearlwort).
- Successfully achieved aims; no off-target impact.

Table 2. Effect of Logran after 1 month.

Weed	Impact
Flick weed	Decayed/rotted off/eradicated
Asthma weed	Stunted growth, yellowing of leaves
Cudweed	1 to 2 days; strong signs of wilt, decayed
Wood sorrels	Stunted growth, yellowing of leaves
Liverwort	Eradicated
Moss	Stunted growth
Pearlwort	No effect, seed set of pearlwort not affected

Results for Metsulfuron

- Changes took 2 to 3 weeks to be observed.
- Successful on flick weed and cudweed species; most wilted off completely by the end of the month.
- Similar to the effects of Logran on *Oxalis* species; roots and stems still in place.
- Successfully achieved aims.

Table 3. Effect of Metsulfuron after 1 month.

Weed	Impact
Flick weed	Stunted growth, strong signs of wilt
Asthma weed	Stunted growth, signs of rot
Cudweed	Eradicated
Wood sorrels	Stunted growth, yellowing of leaves

Results for Metsulfuron and Logran mix

- Effects take up to 3 to 4 weeks; slow to act compared to other trials.
- Cudweed did not wilt off completely within a month compared to other trials.
- Good against flick weed species; by the end of the month most had wilted off completely.
- Effective against *Oxalis* species; able to produce adverse effects on infestations.
- Possibility that Logran and Metsulfuron are working against each other.
- Aims achieved but not best option.

Table 4. Effect of Metsulfuron and Logran mix after 1 month.

Weeds	Impact
Flick weed	Stunted growth, strong signs of wilt
Asthma weed	Stunted growth, yellowing of leaves
Cudweed	Stunted growth
Wood sorrels	Stunted growth, yellowing of leaves
Pearlwort	No effect
Moss	Stunted growth

Summary of results

- Earlier stages of trials are positive.
- Trials show that Logran and Metsulfuron act better on certain weeds.
- Same mode of action, different active constituents; affect different weed species at different rates.
- Ongoing trials: Liverwort regrowth, time it takes for new weed growth after application.
- More trials to be done with different Group B Herbicide products.
- Repeat current trials for conclusive evidence.

Potential with caution

- Encouraging results.
- Impacts on succulents/herbs may be adverse.
- May be more relevant to natives and strong ornamentals.
- Suggest small scale trials with very low concentrations, and then upscale to achieve weed morbidity and assess off-target impact.

LET'S STAY IN TOUCH

- We will proceed with more trials and report via IPPS and to New Zealand.
- It's an interesting exercise/variety for staff.

- Let us know of any results from New Zealand.

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New Zealand natives for hedging and screening©

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INTRODUCTION

It has become very obvious that many of the selections of hedge plants introduced to New Zealand have become environmental disasters. Examples of this include gorse (*Ulex europaeus*), privet (*Ligustrum* spp.), *Acmena* spp. (lillypilly), and *Berberis* spp. to name a few.

Others such as *Buxus* spp. have a dreadful smell and are susceptible to rust while various species and cultivars of conifers are susceptible to fungal diseases resulting in large areas of die back. It appears that this is spread by hedge trimmers while poor pruning methods are also to blame.

NEW ZEALAND NATIVES

With this in mind, we have been planting New Zealand native trees and shrubs in various situations to get some idea on how frost and wind hardy they are, how tolerant are they to drought and wet conditions and what is their ultimate size without pruning is.

New Zealand has an amazing array of shrubby trees that fit the bill including *Pittosporum*, *Muehlenbeckia*, *Lophomyrtus*, *Coprosma*, *Myrsine*, *Melicytus*, *Corokia*, and *Olearia*. These are all readily available and these have been used over a number of years for wind breaks, but there are other species that have been very rarely planted for garden hedges and ornamental situations. Many of these that could be used are perhaps slower to grow for some in a nursery situation but are ideal in garden situations as they require little or no pruning and training. A number of these species that have been tried are of divaricating or filiramulate form.

Pittosporum

Starting with *Pittosporum*, there are about seven species we have tried. The taller ones include *P. obcordatum* which grows to 3 m high and is a mass of bronze twigs. A great windbreak for most soils and will tolerate wet situations. *Pittosporum turnerii* also grows to 3 m and has very narrow, silvery growth and grows well in shady positions.

Smaller growing species include *P. anomalum* which grows to 1 m high with tight dark brown growth and pale cream flowers. *Pittosporum crassicaule* reaches 50 cm high and is very slow growing with tight growth and black flowers. *Pittosporum rigidum* grows 1 m high with narrow dark twigs. All these *Pittosporum* have perfumed flowers at night! All grow from cuttings or seeds which can take some time to germinate.

Melicytus

Melicytus, which can look like *Buxus* species, can be trimmed into small hedges or topiaries such as turkeys or elephants as the mood takes. Forms of *M. obovatus* have been very good some reach 1.5 m down to 50 cm in height. *Melicytus crassifolius* is an excellent tiny hedge and has several forms, also some hybrids that are well worth trying. It provides food for lizards, moths, etc. All *Melicytus* are very long lived.

Myrsine divaricata will take damp soils and freezing if South Island forms are used. It is also tolerant of salt winds and is a slow growing tangled twiggy shrub, but will grow into a larger tree over time. *Myrsine divaricata* 'Poor Knights', now known as *M. aqualonia*, is also great for coastal and drier windy sites. A must mention is the related *Elingamita johnsonii* which makes a great hedge for salt windy costal conditions. It is a small shrubby tree but must have good drainage and no frost.

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Coprosma

There are a huge number of *Coprosma* suitable for coastal windy conditions. Some may need trimming to keep them under control but the narrow, taller growing species form a windbreak to 3-4 m high. *Coprosma virescens* is possibly the best with orange or pink stemmed twigs, doesn't require much trimming and great fruit for the birds. *Coprosma rhamnoides* has great potential also with its many forms and leaf colours and will also tolerate shade and a range of sites.

Muehlenbeckia

Muehlenbeckia complexa will soon cover any space, but with training and trimming it can make any old fence into a work of art. *Muehlenbeckia astonii* grows up to 2 m high and forms cloud shape hedges. *Metrosideros perforata* inter planted with *Metrosideros carminea* will make a nice flowering hedge with not much trimming or can be planted to cover a wall so it looks like a hedge.

Ozothamnus

Ozothamnus species grow to 1 to 2 m high forming a bushy shrub that will grow in the most dreadfully windy, salty, and sandy sites. They have a range of grey or golden leaf forms with masses of tiny daisy flowers for native bees.

Daisy family

Most of the daisy family are tricky in a nursery situation as many get root rots in summer due to over watering, but are fine when planted out and provide plenty of flowers over their silvery foliage in many cases. One for a wetter site is *Olearia solandri* which also has an amazing perfume as do several members of the *Olearia* family.

Tree species

There are several tree species that are worth a mention, but note these are not for all sites due to the size they can grow over time. Many can be trained and trimmed quite happily if this is the desired look. Examples include *Carpodetus serratus*, *Corynocarpus laevigatus*, *Griselinia*, *Hoheria angustifolia* or *H. sexstylosa*, *Kunzea*, *Leptospermum*, *Libocedrus*, *Metrosideros excelsa* or *M. robusta*, *Planchonella costata*, *Podocarpus totara,* and *Streblus*. Just remember what's on the label is not necessarily always correct in terms of information.

Naming and trading for cultivars©

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INTRODUCTION

Commercial horticulture and agriculture is reliant on the production of new cultivars. In order that these cultivars, and products from them, can be effectively traded their accurate identification and naming in the market place is important. This is particularly important if the cultivar is subject to or associated with intellectual property such as Plant Variety Rights (PVR) or Plant Breeders Rights (PBR).

NAMING FOR BOTANY AND SCIENCE

The naming of cultivars (nomenclature) consists of two components, the first being the botanical or scientific name and the second is the naming of the cultivar itself. Both of these components have respective sets of rules (codes) governing their correct usage. Botanical names follow a binomial (two name) system of nomenclature which provides the genus and species. There can be ranks below the level of species including subspecies, botanical variety, and form, and many ranks above genus, such as family. Collectively these ranks constitute a classification. The binomial system of botanical nomenclature began with Carolus Linnaeus in the mid-18th century and today is overseen by the International Code of Nomenclature for algae, fungi, and plants (ICN), formerly the International Code of Botanical Nomenclature (ICBN). The ICN is periodically reviewed via meetings of the International Botanical Congress held every few years (ICN, 2012).

Some plant groups have been subject to numerous name changes by botanists, often as a result of molecular studies, in an attempt to more accurately reflect true taxonomic relationships. These changes are required to be formally published under the rules of ICN, before a taxonomic change can be accepted. For the practical commercial user this can be frustrating and challenging. With respect to cultivars, the absence of stability in some botanical names creates problems for aspects of legislation, administration, and database management (Taxonomy of Cultivated Plants, 1999). Botanical name changes can impact on the checking for suitability of cultivar names for PVR protection; for example where there are two cultivars legitimately with the same name in different genera, then the two genera are recircumscribed into a single genus. Previously the same cultivar name could be used in each genus but now there are two cultivars, illegitimately with the same name in the same genus. Relatively recently a prominent genus level change has been made for the tomato. They were previously classified as *Lycopersicon lycopersicum* (L.) Karst. ex Farwell (and also *Lycopersicon esculentum* Mill.), but following reclassification the botanical name for tomato is now *Solanum lycopersicum* L. var. *lycopersicum.* This name change affects about 7,500 cultivars (PLUTO, 2014).

For many cultivars the botanical name consists solely of the genus with no species name stated. This situation is acceptable in some circumstances such as having uncertain or unknown species information, or a complex breeding history for that cultivar (Taxonomy of Cultivated Plants, 1999). For example, it is unusual for many modern rose cultivars to be assigned a species due to a long and complex history of breeding that has involved crossing several species (Mordern Roses XI, 2000).

In most cases the breeder or introducer of a new cultivar does not have any choice regarding the botanical name. It is pre-determined by current usage; the breeder or introducer is only responsible for checking to ensure that the correct name is used. In some cases, where botanical reclassification has occurred, there may be a need for a decision to be made to continue with the former treatment or change to the new one. Several years ago the

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former genus *Michelia* was merged into *Magnolia*, and you will now find cultivars in the market under both *Michelia* and *Magnolia.* The use of two names in commerce for a single genus exists even without recircumscribed botanical names, for example, the name *Bacopa* is commonly used to sell cultivars belonging to *Sutera cordata*. *Bacopa* is an entirely different genus of aquatic plants and there is no botanical or morphological connection between the two. At some point, an error was made when naming the first cultivars of *Sutera cordata.*

The International Union for the Protection of New Varieties of Plants (UPOV) uses the terminology "denomination class" to describe the botanical name component of a cultivar name (Explanatory Notes on Variety Denominations under the UPOV Convention, 2012). This provides a direct link with cultivated plant taxonomy, which requires that a cultivar name is unique and cannot be repeated in that genus or denomination class. Although a denomination class is usually equivalent to a genus the terminology of denomination class is used because there are exceptions, allowing closely related genera to be usefully grouped within a single denomination class. A number of grass genera are grouped together this way in a single denomination class. For example, it would be confusing to have a brown-top cultivar with the same cultivar name as a fescue cultivar as both could be sold together as a turf seed mixture. Another example is the single denomination class for *Petunia* and *Calibrachoa*, taking into account the botanical connection and the commercial use of cultivars from both genera. The UPOV website contains the full list of denomination classes which comprise of more than one genus.

THE CULTIVAR NAME

Following consideration of botanical (scientific) names is cultivar names. The word "cultivar" is a contraction of "cultivated variety" and is used to make the distinction from a formal botanical variety (ICNCP, 2004). Plant variety protection and UPOV use the word "variety" in the sense of cultivar, not in the botanical sense. The 1991 UPOV Convention defines a variety as a plant grouping within a single botanical taxon of the lowest known rank. The names of cultivars can also be referred to as cultivar epithets or variety denominations (UPOV, 2006; ICNCP, 2009).

The system for naming cultivars is overseen by the International Code of Nomenclature for Cultivated Plants (ICNCP) often shortened to the Cultivated Plant Code or even more simply, the Code (ICNCP, 2004, 2009). The ICNCP is periodically reviewed by the International Union of Biological Sciences Commission for the Nomenclature of Cultivated Plants, with the latest review carried out in 2013. The Code provides a stable and simple system for the naming of cultivars using a list of Articles containing detailed provisions divided into rules. The Code aims to provide a consistent set of rules that are applied internationally. Cultivars protected under plant variety protection are subject to the UPOV Recommendations on Variety Denominations and coexist with ICNCP, but go further in several key areas than the Code. For example, it is common practice for a cultivar to be protected in a number of countries or territories and Recommendation 5 states that a cultivar should have the same denomination in all places where plant variety protection has been applied for (Explanatory Notes on Variety Denominations under the UPOV Convention, 2012). This highlights the importance of the same cultivar name or variety denomination being used for that cultivar in all parts of the globe. Associated with the principle of a single global variety denomination, the denomination must be unique to that cultivar, universally applicable and used while under protection and after protection when free in the public domain.

A single, universal cultivar denomination must be able to clearly differentiate that cultivar from others and should not mislead or cause confusion regarding characteristics or identity of the cultivar, or the origin or identity of the breeder. The combination of the use of ICNCP and the UPOV Recommendations create a level of global certainty and consistency regarding cultivar identification.

The responsibility for the selection of a cultivar name or denomination begins with the breeder or introducer. ICNCP is utilised for the voluntary international cultivar registration system and UPOV Recommendations and ICNCP are used in the formal approval process for

protected varieties. It is important to recognise that it is not the role of any official or voluntary authority to select a suitable name, only to approve or reject a name selected by the breeder. For the numerous cultivars not subject to any intellectual property or voluntary registration, the breeder or introducer has the greater individual responsibility to select a legitimate name that follows the rules. It should be noted that ICNCP has no rule enforcement provisions and numerous illegitimate cultivar names are known to exist.

The cultivar name or variety denomination is intended to be the only reliable and consistent means of identifying a cultivar worldwide, but for many there also exist one or more commercial synonyms associated with and used to sell the cultivar which in some cases may become a de facto or be seen as alternative cultivar names. It should also be noted that under the rules of priority in the ICNCP, the earliest validly published cultivar name should take priority and any other names are technically illegitimate.

COMMERCIAL SYNONYMS OF CULTIVARS

Commercial synonyms broadly cover all fancy names, selling names, brands and trade designations, as well as registered and common law trade marks. This description for commercial synonyms could also be used to describe trade marks. Registered trade marks are subject to a formal registration system and must conform to provisions under that law. Commercial synonyms have no legally defined status but there may be common law Rights attached, which may be recognised. The use of a commercial synonym may not in itself be enough to provide any Right to exclusive usage.

Commercial synonyms are used to sell cultivars and are an important plant marketing tool. Many plant variety protection schemes recognise this by unofficially holding such information in databases and permitting the association of a commercial synonym with a variety denomination to sell a protected variety, providing that the denomination is always used and clearly recognisable (Trade Marks and Variety Names, 2014). National authorities tend to have regulations which require use of the denomination on plant labels in particular but in a broader sense the awareness and knowledge of individual cultivar names or variety denominations in many genera are increasingly only known for official purposes and to relatively few in industry or the public.

The use of commercial synonyms has increased in recent years. To an extent this is understandable when a breeder attempts to satisfy the requirements of plant variety protection (or other official registration) and also the demands of marketing and selling plants of that cultivar. The cultivar may be commercialised in many countries and a name may be successful in one market but a complete flop in another. Add in the complexity of different languages, translation and cultural interpretation, and choosing a cultivar name that meets ICNCP, UPOV Variety Denomination Recommendations and is also a market winner is challenging. A good name goes a long way to sell plants and that is the primary objective of plant producers.

The wider acceptance and use of this alternative name approach across many genera has led to what some have described as nonsense variety denomination and cultivar names, such as alpha numeric combinations, very different from names of 20 years ago. For example, *Calibrachoa* 'KLEC02073', *Agapanthus* 'CORAG02BL', Japanese Plum 'Suplumthirtytwo' and *Cordyline* 'Jel01'. The alternative name approach can lead to problems in correctly recognising cultivars, in particular when the cultivar name or variety denomination name is not used as it should be. Rose breeders were one of the first groups to promote and develop code-like denominations, partly to avoid name duplication in the registration or variety protection process and to address the problem of different roses being sold with the same name (Mordern Roses XI, 2000). This approach is now entrenched across the horticulture industry with the commercial synonym used to sell plants and the formal cultivar name or Variety Denomination used only for identification and official purposes. Accurate identification involves the ability to separate and recognise cultivars, and the similarity of some code-like denominations questions whether this is actually achieved. Variety denominations such as 'DBB03', 'DCNCO', 'Gruetib01', and 'Gruetib02' are acceptable under UPOV and the Code, but whether they allow for easy recognition and identification is

another question. In many instances, breeders and variety owners themselves do not have familiarity with, or routinely use or recognise, variety denominations for their own cultivars. The alternate name approach can be workable providing the commercial synonym is used together with the cultivar name, but having effectively more than one single global name for each cultivar is not compatible with the ICNCP or the UPOV Variety Denomination Recommendations prescribing or recommending clear and consistent identification.

Consideration should also be given to the nature and usage of the commercial synonym itself, with no official or international code guidance available for the breeder or producer. From a marketing point of view the long term use of the synonym may be desirable and over time could be associated with several cultivars from the same breeder or introducer.

A successful synonym may become closely associated with a single cultivar, with the synonym itself clearly identifying a specific cultivated variety. In such a case, the synonym itself has effectively become the cultivar name. An example is the lavender variety 'James Compton' which is widely known by the synonym Fairy Wings. This may limit the possibility of using the synonym to sell other cultivars from the same breeder and may also rule out the possibility of the synonym being accepted as a registered trade mark. To avoid such a situation, trade mark registration of the synonym should be considered early and care taken regarding how the synonym is used.

The commercial synonym name itself should not have been previously used by the breeder or anyone else as a cultivar name or variety denomination for a different variety. The use of an existing cultivar name as a commercial synonym to sell a different cultivar may be viewed as misleading and create an element of confusion as to the identity of both cultivars involved.

Along with the use of commercial synonyms as a whole, trade marks have become more common in the market place to sell cultivars. Trade marks are an important business tool and are used by a business to identify goods and distinguish them from those of others. The main function of a trade mark is to identify the origin of goods, and with respect to sale of cultivars, the breeder or producer. However, trade mark use in the sale of plant varieties often identifies the cultivar itself rather than the breeder or producer. Going further, it could be said that some trade marks are used as substitute names for cultivars and clearly identify that cultivar. This situation raises questions regarding correct use and possible validity of the trade mark. It is not uncommon for rose trade marks to effectively be used as the name for the cultivar and many rose growers and buyers would have no idea that the commonly used name is a trade mark and that the rose also has a cultivar name or variety denomination (Gioia, 1995).

The commercial use of synonyms associated with a variety name will continue to be practiced but would be improved by creators of synonyms giving greater consideration to whether a synonym is advantageous at all, and to the short, medium and longer term usage implications for the synonym itself and on the cultivar name or variety denomination. Care should be taken to ensure that the commercial synonym does not become a second cultivar identifier or clearly describe the cultivar. PVR is for a fixed term and if the exclusive use of the synonym is anticipated beyond the term of PVR protection then a trade mark application for the synonym should be made early in the life of the cultivar. There is a risk in applying for a trade mark of the commercial synonym at the end of the PVR period because it is possible that your commercial synonym could be viewed as a descriptor for the variety which will preclude it from becoming a trade mark.

Any use of synonyms should be included in a business's marketing plan and, as with any other business practice, be documented and subject to objectives and goals. The role of the synonym is to sell and market plant cultivars not to specifically identify them, which is the function of the cultivar name or Variety Denomination.

INFORMATION SOURCES

Lists of cultivar names for particular genera, such as those maintained by voluntary registration systems, can be found in published books or checklists and on the web but in

most cases there is a need to know where to look. These cultivar lists are available for relatively few genera. For a broader approach the UPOV Plant Variety Database (PLUTO) is available on the UPOV website and consists of all cultivated varieties protected in most of the UPOV member states (PLUTO, 2014). It is possible to search by genus and denomination and both in combination. You can check a possible cultivar name by entering that name and retrieving an exact or similar match. In addition the database contains the names of cultivated varieties subject to any national official variety registration systems, a common practice in some countries for the marketing of vegetable and agricultural varieties.

Checklist for naming a new cultivar

1) Confirm the genus or, if possible, both genus and species. Check for any botanical revisions that will affect the cultivar.
2) Select a suitable cultivar name or variety denomination which clearly identifies the cultivar. Consider if PVR protection or voluntary cultivar registration will occur and whether the cultivar is likely to be globally marketed and protected in the longer term.
3) Will the cultivar be sold in association with another name or trade mark? How will the other name be used and for what period? Has the other name been used anywhere else for any purpose? Could the synonym become generic and a de facto second cultivar name?
4) Use the cultivar name or variety denomination to identify the cultivar and include it on labels, product lists and catalogues.

Checklist for applying for a trade mark:

1) Is the proposed trade mark name distinctive? Will it identify your goods from those of other traders? A trade mark cannot describe your goods.
2) Is it a recognised Variety Denomination or cultivar name in New Zealand? Your trade mark cannot be a variety denomination for a current or expired protected variety.
3) Is your trade mark a recognised cultivar name elsewhere? This could also prevent your trade mark from being registered as the overseas cultivar could be known in New Zealand. It could also lead to market confusion regarding the true identity of your cultivar.
4) Is it the same or similar to other trade marks? Your trade mark cannot be the same or similar to someone else's trade mark on the same or similar goods/services.

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IPPS Japan exchange 2014©

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Excited, amazed and lucky were just a few good descriptive terms I could have used to describe how I felt when I was informed I was the fortunate recipient of the IPPS exchange scholarship to visit Japan. So on Wednesday 17th September, I boarded my flight, leaving Nelson and my job at Waimea Nurseries behind, while I went off into the big wide world to a country I had no comprehension of.

After arriving in Miyazaki after a series of pleasant flights, I was greeted by Mr. Takuya Tetsumura who took me to my host family. Miyazaki is located in southern Japan in the island of Kyushu it has a warm wet climate and facilitates a diverse range of horticultural activities including mango growing, tea production, and market gardening. All of which are grown on a relatively small scale compared with New Zealand production.

My hosts were the Kusano family of Aya Engei, a nursery producing flowers. The nursery produces pot plants, bulbs, seeds, and cut flowers for the main market. The nursery is well set up with its own tissue culture lab, facilities to store and dry bulbs and seeds and many greenhouses. Various flowers are crossed and offspring that have flowers with favourable characteristics are then reproduced asexually through the tissue culture lab and flowers, seed, and bulbs are sold. This gives Aya Engei a competitive advantage and makes it one of the top flower nurseries in Japan.

As well as spending some time working in the business, I also got the opportunity to explore Miyazaki. Some key highlights included Aya Castle, the Aya biosphere reserve, and visiting the sacred shrines. Along the way we also called into several horticultural related organisations and firms. Aya town's organic centre, a local mango grower, and the Miyazaki Agricultural Research Institute were of notable interest. But without a doubt one of my favourite days in Miyazaki was the day we climbed Mt. Takachiho, about 1-h inland. The mountain is 1574 m high and to get to it, we had to sidle a volcano!

After a week it was time to move on. I travelled north via car and plane, witnessing a volcano eruption along the way near Kagoshima. After arriving at Nagoya city which is approximately a 5-h drive from Tokyo, my host, Mr. Uchida, took me out of the city to his nursery.

Tumugi (Mr. Uchida's nursery) focuses on producing high quality strawberries and figs for the local market. The operation pays careful attention to its environmental impact and hopes to become organic in the future. It produces around 2000 kgs of both strawberries and figs, seconds and excess are processed onsite into high quality value added products such as jams and cakes.

Again mixed with work, I was lucky enough to see some of the great attractions of the area. The 60-m-high Buddha at the Todai JI Temple and Kinkaku (the golden temple) certainly highlighted the differences in culture between New Zealand and Japan and were a real eye opener.

After a week, I proceeded to my final destination for the trip, Kanagawa, which is close to Tokyo. Here the IPPS Japan region conference commenced, and I got the opportunity to meet some of the key people involved in plant propagation in Japan and therefore made some valuable contacts. I also got the opportunity to share with them an insight into my life in Nelson and the work I do at Waimea Nurseries.

I'd like to thank both New Zealand and Japan IPPS members for their generous contributions to my trip. It's been an opportunity of a lifetime and one I'll never forget.

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Buying quality nursery stock – a consumer perspective©

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INTRODUCTION

It goes without saying that trees can be produced at variable levels of quality and it is one of the challenges for consumers in today's marketplace to be able to pick the winners. Unfortunately, many consumers don't know what a good quality tree is and that is part of the problem. They rely on the market to produce good quality trees for them but this requires producers to know what a good quality tree is.

Figure 1. These two trees would have cost the buyer the same amount of money. The one on the left has good form and good trunk taper, but the one on the right has poor structure and only lasted 10 years.

As trees grow, little problems can develop into really big problems. Figure 2 shows what happens when root-bound stock gets planted in field. Poor quality tree stock like this is unlikely to grow beyond 10-20 years.

Figure 2. This is what happens after a few years when root-bound trees are planted out into the landscape. Top right image credit, Dr. Ed Gilman, University of Florida. Used with permission.

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If consumers make good choices when buying trees they will be making a good investment; their trees will live longer, perform well, provide a very good environmental contribution, be generally low maintenance, and generally be safer trees. Unfortunately, finding trees to fit this bill can be challenge as a lot much of the tree stock I see produced in New Zealand is simply not fit for purpose.

So when a consumer goes to a local nursery to look for a tree, or browses through a catalogue, there are many factors to consider when purchasing container-grown trees. Things we will consider here are root collar location, problems caused by root depth, and issues with branch structure.

FACTORS TO CONSIDER WHEN PURCHASING CONTAINER-GROWN TREES

Correctly locating the root collar

First, let's just define what a root collar is and where it should be located in the grow bag. The root collar should be above the soil line and the uppermost root needs to be within 50 mm of the top of the container or bag. Unfortunately, the majority of root collars get buried during production as trees are bagged on to larger grades. Buried root collars encourage roots to grow up into the media on top of the main root system and invariably girdle the main stem.

As a consequence of this, when trees are planted out into the landscape they perform poorly because they struggle with soil aeration. They do all right in the nursery because the nursery is really a holiday resort for trees – they're being fed and watered and get adequate oxygen. But as soon as they are planted out they struggle when their roots are situated below grade. Small girdling roots can also develop to become a serious problem.

What is a high quality root ball?

A high quality root ball is where the root collar is above grade and the point where the topmost root emerges from the trunk is within 50 mm of the soil surface such as those shown in Figure 3. In contrast, a poor quality root ball is where the root collar is buried, either by being planted too deep in the container or excess soil is placed on top as shown in Figure 4.

When trees are container-grown, containers are often topped-up with extra soil or medium as the trees settle lower. As trees are potted-on into bigger containers, this cycle can be repeated. This results in roots growing up into the medium above the root system and encircling the stem.

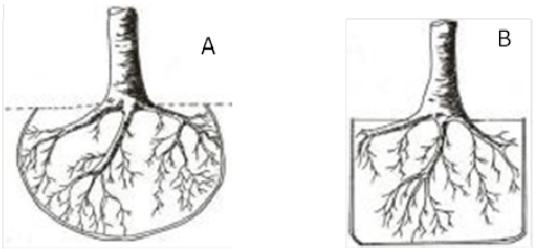

Figure 3. What a high quality rootball should look like with the top most root emerging no more than 50 mm from the soil surface. From BS 8545: 2014 Trees: from nursery to independence in the landscape – Recommendations. Drawings courtesy of Keith Sacre.

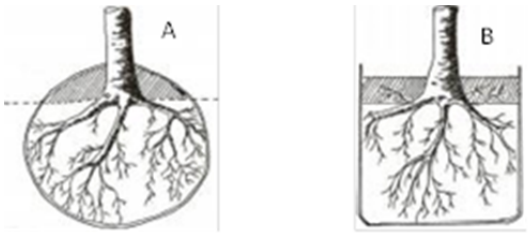

Figure 4. Poor quality root balls where the top most root and the root flare are buried inside the root ball.

Basically, trees such as that shown in Figure 5 have been placed in the bottom of the bags, then medium put on top of them two or three times in the course of their production so they ended up nearly in the bottom 25% of the bags. Trees like this should not be planted.

Figure 5. A very poor quality tree where the root collar and the original uppermost root were found to be in the lower 50% of the bag.

The problems that we see with trees on sale in the marketplace like those in Figures 5 and 6 could easily be avoided with education and can simply be addressed during production as they can't be undone once established. In discussions I've had with some nurseries I'm surprised that many don't seem to even know about these issues. I learnt about these problems more than 20 years ago and it is unfortunate to see how widespread they still are.

Figure 6. Newly potted stock in a wholesale nursery. The white line indicates where the root collar is – in the bottom half of the container.

Root defects have a significant impact on landscape performance and tree stability. Basically, a tree needs to have really good structural support to take the wind load it will

receive to its canopy as it grows. Trees with poor root structure are not going to be able to produce good, radiating, stabilising roots because they had well-established girdling or circling roots at the time of planting.

Trees with root systems like that in Figure 7 should not be planted. This tree had a price tag on it of $NZ 225 and it's really quite a defective tree. Unfortunately, in the line of trees where it was situated, there were lots of gaps where people had been buying these trees and they will have been planted out in the landscape.

Figure 7. An example of a poor quality tree with roots encircling the bag which was so tight, the stitching was pulling apart. Lichen on the stem indicates also the tree had been in the nursery for several years.

I have seen the results of much of this type of plant material over the past 20 or so years. People ask me to come along and tell them why their tree has died or why it keeps falling over. Why do we need to keep staking our trees? Why do we need to keep staking our trees? In most instances I find the problem appears to relate to defective root systems created during production. So, right from the start these trees were set up for failure.

Root defects are one of the leading causes of early tree mortality. Without a really good, well-developed root system a tree just can't perform in the landscape. Inferior trees will be lucky to reach 10 to 20 years of age when they should last 80 to 100 years or more. Clearly when plant material like this is sold it is just not fit for purpose.

Plants people may ask, "oh, when you buy a tree like this, can't you just butterfly the root ball or cut the roots on the outside and all will be well?" But root ball defects can occur at all stages of production and defects on main roots close to the trunk are difficult to correct. In Figure 8 below, you can see the different bag sizes used as the tree was bagged up.

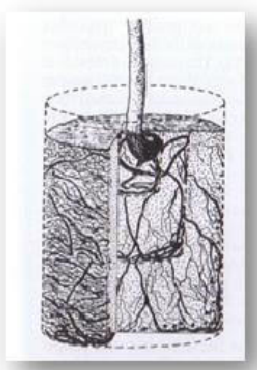

Figure 8. Diagram showing how roots have grown as a tree has been bagged up into different sizes. From: Harris et al., 2004.

You can deal with the ones on the outside of the bag by cutting these roots but many

root defects aren't very visible to the purchaser when they're buying their trees. Trees that have been recently bagged up will look ok but unfortunately can have hidden defects well within the root ball that may have started at very small grades and lead to premature death (Figure 7).

It is a real challenge to produce container-grown trees without root defects. People all over the world have put a lot of effort into trying to overcome these challenges. Many different bags, containers and methods have been tried. There are copper-impregnated bags which are designed to prune roots when they reach the bag sides; there are containers with corrugated sides to stop circling roots and others designed to air-prune roots. So it's obviously a recognised problem.

Figure 9. This tree got to 25 years old and then died. It is not hard to see why.

Branch and stem structure

Consumers come along and pick through the stock shown in Figure 10, get the best of the worst and take them away. The ones that are left sit there and sit there, the price gets dropped, so they then go out the door as well and will end up failing in the landscape. This stuff is really just complete rubbish.

Figure 10. Examples of poor quality trees found in a big store retail nursery.

Trees of lesser quality have two or more leaders. Ideally a good tree should have one central stem extending to the top of the canopy and this sort of thing is the easiest to get right in a nursery. All you need is for someone to go out and do a bit of pruning (Figures 11 and 12).

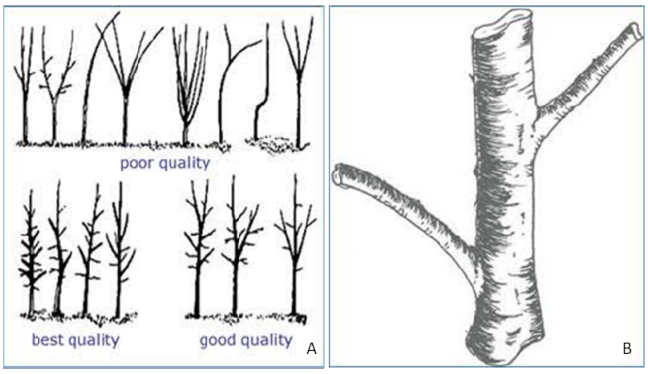

Figure 11. Trees of poor and good quality (A); Drawings used with permission from Dr. Ed Gilman, University of Florida. Branches need to be about 50% of the diameter of the main stem, avoiding co-dominant stems of similar diameter (B); From BS 8545:2014 Trees: from nursery to independence in the landscape – Recommendations. Drawings courtesy of Keith Sacre.

Figure 12. Examples of good and poor quality trees and how they should look. Used with permission from Dr. Ed Gilman, University of Florida.

Good quality trees need to have main branches that are not touching each other or the main trunk. Branches on large trees should be about 450 mm apart and have nice radial spacing while the main branches on smaller trees should be about 150 mm apart. Ideally, branches need to be about 50% of the diameter of the main stem, avoiding co-dominant stems such as that illustrated in Figures 12 and 13.

Figure 13. An example of a poor quality tree with co-dominant stems.

Co-dominant stems

Many poor-quality trees look like the example in Figure 10 illustrating a tree with co-dominant stems, major branches are touching, there are v-shaped crotches, lots of included bark, so all in all a very weak branch structure. It is really difficult to correct this. In fact this tree was heavily pruned after it was purchased and that's what could be made out of it after it was pruned. Basically, the wind did the deed and the tree failed in the end. These branch unions just failed so the tree had to be removed.

Stem taper

Trees need to receive a wind load to enable them to start developing good taper and become wind-firm. Staking trees needs to be very well managed in order to produce trees with good taper. Trees will not be self-supporting (develop good taper) if staked throughout production.

Trees with the same stem diameter at the soil line as they do further up the stem will have real difficulty in performing in the wind when they are planted out in the landscape. Trees with well-developed taper are what consumers should be looking for when selecting trees. Figure 14 illustrates good and poor stem taper.

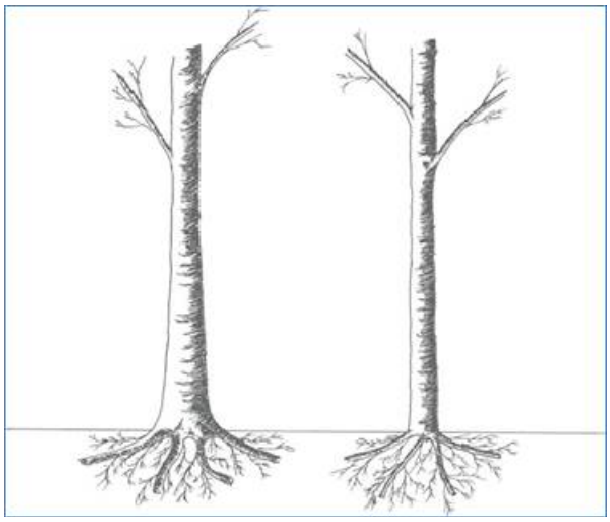

Figure 14. Tree to the left has well-developed stem taper; the one on the right has poorly-developed stem taper. From BS 8545:2014 Trees: from nursery to independence in the landscape – Recommendations. Drawings courtesy of Keith Sacre.

Pin oaks are a classic example you often see – big floppy things, very narrow-stemmed at ground level and very, very tall because they've been staked or held against a wire system or grown too close together.

It's very difficult to hold these trees up in the landscape. You have to continue to stake them, further inhibiting the development of taper as movement is restricted.

Often a staked tree in the landscape gets fatter where it flexes above the stake when it really needs to build a good taper at ground level. Ideally, trees should have good height-to-stem girth ratio such as that illustrated in Figure 15.

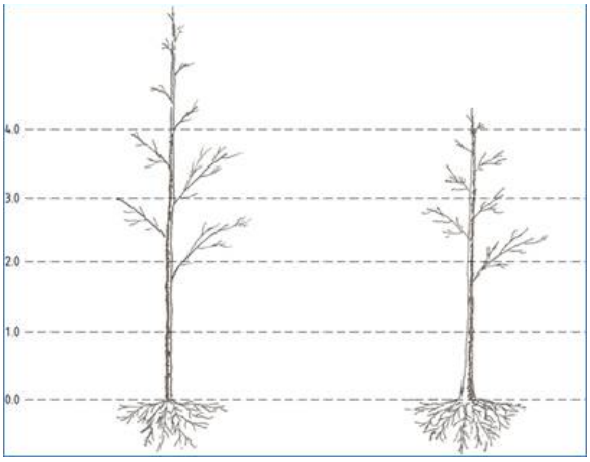

Figure 15. Tree to the left has poor height to stem girth ratio; the one on the right has good height to stem girth ratio. From BS 8545: 2014 Trees: from nursery to independence in the landscape — Recommendations. Drawings courtesy of Keith Sacre.

Clearly the tree shown in Figure 16 has had structural issues from an early stage that could have been corrected in the nursery or have been avoided by the consumer. Small poor quality trees can become big expensive mistakes. It now presents a considerable challenge for anyone managing it.

Figure 16. Small, poor quality trees become big expensive mistakes.

And where trees also have poor root systems such as that shown in Figure 17 – well, we've had lots of wind events in recent years and these storms provide a good test and indicator of the quality of trees in our landscapes.

Figure 17. A common sight after wind events

Of course there are the big trees that fail for various other reasons but there are hundreds and hundreds of small trees falling over as well. On close inspection, many are seen to have bad root systems. The problem is they had well established circling or kinked roots when placed into the landscape and they've just got no ability to produce any stabilising roots and take those peak loads when storm events come along.

So maybe the time has come for nursery standards in New Zealand to address some of these issues and to offer some confidence to consumers? I know Australia has been working hard over quite a few years to get theirs going and I think it's in the final draft stage at the moment. The British have got a fantastic document called BS 8545 Trees: from Nursery to Independence in the Landscape. It's quite an outstanding document.

For consumers to be able to grow good trees in the landscape, they need a supply of good trees. It would obviously help if consumers knew what a good tree is but unfortunately they don't so they will continue to keep buying what the nursery industry serves up to them. There are some good people out there trying to do a good job and produce good trees, but unfortunately they are a minority.

At the moment the market appears to be very producer-driven in New Zealand. The stuff just gets served up and it goes out the door.

Maybe if consumers knew more about what a good quality tree is, the pressure could be put on producers to deliver. Either that or the industry has got to start doing something to direct change.

Obviously, we need more nurseries to know what a good quality tree is and obviously there's a bit of education needed there. No one really sets out to produce a poor quality tree but there appears to be a fundamental lack of understanding of how trees grow.

In conversations I've had with growers, they've not been aware that the root flare should be at the top of the bag, and they didn't understand that when it wasn't, that this was actually a serious problem. So I don't know what sort of message that sends.

Why growers of tree material wouldn't know this sort of information is surprising when there is so much published literature on the subject of nursery quality. Maybe it's an education thing or that the market needs to speak louder.

The bottom line is that poor quality trees, especially those that have intact circling roots, are a total waste of time and there's no place for them in the landscape. They just become a liability and a bad illustration of what a decent tree should be. Trees are getting a

really bad knock for falling over in the wind and a lot of it is because they're flawed.

Trees are incredibly robust structures but unfortunately if they've got flaws then they don't perform as they should.

Retailers are consumers too. They need to know what a good tree is because at the end of the day what they're doing with a tree they stock is putting their brand on it and selling it. So they're the ones that will get the flack if it fails. Retailers need to pay a bit of attention to this issue as well and demand higher quality trees and support the growers producing them.

And I think the New Zealand nursery industry really does need to set some standards. I know you've got a lot of other challenges at the moment but this is one that I think you could put alongside the FMS (Farm Management System) program that you're adopting.

Given that Australia has done a lot of the hard work on a Nursery Standard already, it wouldn't be very difficult for New Zealand growers to take some lead from this and other international standards.

How it should be done

And just to prove that it can be done – the guys at Trees Impact are a very impressive crowd in Australia, that are producing the most amazing tree stock as shown in Figure 18.

Figure 18. Some of the impressive trees grown at Trees Impact, New South Wales, Australia. Image courtesy of Trees Impact.

How they can produce container-grown trees with root systems like the one shown is pretty astounding. Every single one of those roots shown goes right back to the main stem. There are no girdling or circling roots in that root ball and that's proof that it can be done.

If there is anyone in New Zealand producing trees like this, then there are a tonne of consumers out there that would really like to know about you.

Literature cited

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Food for thought – nurseries into the future©

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INTRODUCTION

I don't even think I'm particularly qualified to speak on the future. In fact I don't know any more or probably less for that matter, than some of the folk who make a partial living talking about these things. However, like everyone I do have a perspective though; — that of a small producer.

DISCUSSION

We know that there is of course a consequence to predicting future paths and that is, it is to some extent self fulfilling. It shapes the future just as the choices we make shape the future. So if an expert says this is how plants will be sold in the future it is very easy for the rest of us to decide we need to embrace the future and as such we create an impetus in that direction.

One technique those advising on the future use is to tell a parallel anecdote from another industry that you don't know much about, to illustrate their point. Of course they draw selectively and as such, I to, am going to draw on a selectively chosen anecdote.

The Northampton shoe industry

Since the 16th century the British shoe industry has been almost exclusively centred around Northampton. By the 1970s they were in serious decline. They manufactured by traditional methods, often in old factories that hadn't been updated since the 1930s. They made the best shoes in the world but they were expensive. They were warned. Every industry expert explained that shoes were coming in from Asia that looked almost the same and sold for a quarter of the price. The industry was doomed. The problem is that they were largely family owned enterprises with long traditions and about as flexible as a steel girder so they just kept doing what they did; making beautiful, expensive well made shoes. By 1997, production had dropped by 60% in the Northampton footwear industry. Most of their craftsmen gradually retired and weren't replaced.

Since 1997 staffing levels have increased by 60%. Northampton again employs 6000 people in the shoe making industry almost exclusively producing expensive shoes by traditional methods often in the same old factories. These are brands like Trickers, Grenson, and Crockett & Jones.

These are shoes that sell for $400 upwards.

However they are facing problems again.

Ivor Tilley from Grenson said; "A lack of demand isn't the problem, the main challenge, the real challenge is a shortage of skilled labour."

Another manufacturer that has brought back its apprenticeship scheme says we are getting good applicants. Young people are starting to realise we are artisans not factory workers. The only problem is it takes so long to train them.

All they have done is produced quality, stuck to their guns, and waited for the wheel to turn.

Like all parallel anecdotes it is only partially relevant. The advent of the Internet has helped sell shoes worldwide but it must be noted that a lot of the increased demand comes domestically. Still if someone said in 1990 "don't worry it's all going to turn soon and you won't be able to keep up with demand" they would have been laughed out of Northampton. The staffing parallels are there too for us and we won't attract good people if what we offer them is outdoor factory work. You are kidding yourself if you don't think that's what a lot of jobs in horticulture have become.

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The point is that the future is not something that moves down a predictable path. It is not only, not going to be quite as we expect but it can also take totally unexpected directions.

As much as we try and predict it, it will elude us and the future will affect each of us uniquely. There isn't a one size fits all explanation and so I can only provide you with my perspective on it and of course my perspective is created by my particular exposure to the nursery industry and so I best start with my story.

My story

I began working for Rod and Julia Tallis in 1980. They were not just enthusiastic but also thoroughly descent human beings and we were not just given our jobs to do and then paid but were taken several times a year to other nurseries, garden centres, and gardens. So we came to realize that we were part of something that led to something substantial and beautiful.

I found this profoundly important when I was starting out: "That what we produced went on to make the world more beautiful and thus it was important and because it was important it was important that I did it as well as I could and learnt everything I could about it."

I stayed at Overland Nursery for a total of 9 years (6 of those as production manager) and then started Heritage Plants in conjunction with Bob Dunn who owned Heritage Gardens at East Maitland. I ran Heritage Plants independently whilst Bob owned and ran the retail and so whilst growing plants for the retail was perhaps our main focus it wasn't certainly our only focus. We needed other customers.

Bob decided to retire and sell his business and so 6 years ago I purchased 40 acres at nearby Paterson and relocated Heritage Plants.

I suppose if I do have any insightful perspective it is gained from being a wholesaler who worked for 20 years every day with a retailer that continued to grow and develop. It was a great opportunity to experiment to see what the public (or at least our public) chose to buy.

My business

I was lucky because Heritage Gardens has always concentrated on plants not peripherals and not landscape supplies.

Maitland is also an interesting place to do this because at 60,000 people it is a pretty good representation of a cross section of society and a wide range of people shop at Heritage Gardens. The best customers as you know are loyal, spend well and are a pleasure to deal with and they become the core of your business. Your instinct usually tells you with in 5 min. of meeting someone whether or not they belong in that category. They are certainly not limited to those with deep pockets. Indeed, if I had to put on a label I would probably describe them as discerning.

Quite a few years back I listened to a very successful marketer give a talk. He ran a large office and had run advertising campaigns across lots of categories. His firm did some of their own market research and purchased the rest from 3rd party providers. During his talk he used the same words several times "our research shows that the majority of people…"

I grabbed an opportunity to speak with him later and asked him what constituted a majority. He said it was interesting but the majority is almost always in the low to high 70% range regardless of what you are marketing. I asked him what the other 20% to 30% want and he said it often seems they want the opposite.

The interesting thing here is that I think the sustaining customers of Heritage Gardens come largely from the smaller category and it is one of the reasons nurseries need to be a different thing to other retailers. There is an enormous pressure to homogenise everything. There is a belief that if it works there it will work here and that the majority are always who we need to reach. However I think plants and gardens provide one of the escapes from the sameness. So in away a nursery needs to be the antithesis of the modern shopping experience. I don't think this is well understood.

I think the Industry Association has done a wonderful job representing our industry to

government and an equally wonderful job at providing technical support and knowledge. They have represented and understood the sectors of the industry that are primarily commodity sellers but haven't quite managed to understand what a gardener or a potential gardener is seeking. I have a point to illustrate this, but that said I still believe our industry body represents us well as a whole industry.

I think it can be realistically stated that Heritage Gardens and Parkers at Turramurra are two of the best and most successful garden centres in NSW. They are both very plant centred and have entered Garden Centre competitions. After reading the feedback provided both managers had something interesting to say:

Bob from Heritage Gardens said, "They just don't understand I sell plants not signs."

Rohan from Parkers said, "If I made the changes they suggest I might win the competition but my customers would never come here."

They both realise that most of their customers do not want the big, gaudy box experience. They want something akin to the opposite. Call it charm if you like and charm is a very hard thing to contrive.

Another thing that quite overwhelmed me at Heritage Gardens was the amount of young people who had just bought a house or started a family and now really wanted a garden to make a home. One thing we noticed over 20 years is that if they have initial success they keep gardening. They may try again if they fail initially but they won't try three times.

I think potential gardeners are the most neglected group out of every group that buys plants and there are a lot of them. I regularly question the people who own gardens that open to the public and they constantly reaffirm that a high proportion of people who come through their gates are relatively young. I realise gardeners won't buy the majority of plants sold but they tend to make regular, loyal customers. They are potentially a large part of the future if the horticulture industry can manage to stop losing them.

The way most plants are sold probably discourages potential gardeners. The people selling them have too little knowledge and the proportion of unsuitable plants sold and planted probably outweighs the correctly selected ones

It is hard to think like someone else. Thinking back to when I was deciding what to grow at Overland, I knew mother's day was busy. So we would specifically grow some 140-mm flowering lines like *Dianthus* to make the most of this little spike in autumn sales. I grew the *Dianthus* because they sold and so it seemed to me that I was doing the right thing.

When I first arrived at Heritage Gardens we would stock up on 140- and 20-mm chrysanthemums for mother's day and sell most of them. The public bought them somewhat reluctantly because we didn't offer a better alternative. What it took us a while to realise is that people don't want to spend $20 on their mother. They want to spend at least $60 but they don't want to get three plants either and they don't want something big. They want something small and beautiful – not a small plant in a ceramic pot.

If I could produce the hydrangeas I produce in November in a squat 200-mm pot for Mother's Day garden centres would sell thousands at $50 or $60. At least until someone worked out that they could sell tens of thousands if they halved the price. Then they would proceed to work three times as hard for less money.

The public would feel cheated because those hydrangeas are cheap and everywhere, but at least the grower could say I grew 30,000 hydrangeas last year. I think it is called a lose-lose situation. Too much comes back to mine's bigger than yours as opposed to what makes good sense I'm afraid.

Of course not all plants are sold as gifts; in fact it is only a tiny percentage. To use an awkward analogy, and if influencers can use appalling analogies, so can I. We are like the homebuilding industry. If the *Lomandra* 'Tanika' cells are like the nails in a house someone still supplies the Caesarstone bench top.

If a nursery grows *Dianella* tubes then continuity of supply and price are the most critical things to affect their sales. They are commodity sellers. If a nursery sells 140-mm, seasonal-flowering plants to chain stores it is a combination of price and quality that affect them but if a nursery grows large Japanese cloud trees it will primarily be the quality alone

that decides their success.

People are changing and they will change more if they are guided. My grandmother would never buy a hydrangea because you could strike it yourself. My mother would only buy a small cheap hydrangea not because she couldn't afford the better one but because she was conditioned. At Heritage Gardens several years ago we tried selling well grown 140-mm hydrangeas with flower for $12 and 200 mm plants for $30. People under 50 years almost invariably took the larger one whilst those over 60 years took the smaller one.

Examine a nursery that grows large trees. Lots of trees go into commercial jobs and the process involves a landscaper quoting meaning price becomes critical. When you think of how long a tree remains, its importance and its potential to create damage the quality of the tree and choice of tree should be the main concern. That can be hard to convince people of when they are planting hundreds. However if someone has just bought or built a house, even if they are financially stretched it is not hard to convince them of the value of the better tree. It is not hard to convince them because it is the truth and younger people are even further removed from my grandmother's; "but I could take a cutting" than I am.

I mentioned the Caesarstone bench tops earlier because I am installing a new kitchen. The man I bought the kitchen through said that he thinks laminex tops are almost as good as stone. Seeing as how they are a third of the price you can afford to replace them if needed. He said that despite this two thirds of his customers choose stone because they have been convinced it is worth the extra couple of thousand dollars. Hence it is not hard to convince someone to pay $100 more for the right large tree, and you are doing them a favour.

Older people often say that is because they don't know the value of money but it is not just that. They are more likely to be open to influence than previous generations and less likely to be captured by the values of their parents and while we are constantly told they crave information, whether they realise it or not, what they really want is knowledge. Knowledge and information are two different things but convincing people to buy wisely so they gain the right end result is a lot nobler and a lot more professional than flogging them something just so we get a sale.

The thing that we are short on and the thing we need and the thing that will become valuable in the future will be horticultural knowledge. We are bulging at the seams with people who know about and are interested in production efficiencies and logistics but too few people who can guide an end user who is struggling to find the knowledge they need.

I think that for wholesalers the current mix of large and small nurseries could stay in about the current ratio. However when the current crop of small nursery owners retire quite a few nurseries will close. This is not because they are unprofitable (although some are) but because there won't be enough people prepared nor skilled nor with the resources to take them on. So there will be a shake down by natural attrition and that will leave large voids and of course these voids need filling.

I think that changes are coming, larger wholesale nurseries will have even more market share but it won't be because they are doing things better or are more profitable.

The void left will be filled initially by big producers but perhaps eventually by smaller ones. As the market demands specialist products of a high quality someone will start to produce them again. Just like Northampton's footwear makers, skilled labour is going to be our challenge and I think we need to look at how we remedy this differently. I don't think school leavers are the answer but that's another talk for another time.

The losers from all this will be everyone: The big wholesalers (because they will lose range of supply), retailers because they won't find what their customers want, and most of all the public. This will exert a further downward pressure on the profile of gardening. Gardening is an instinct in a lot of people and no matter how small our blocks or balconies become people will want to create their own manipulations of nature.

RETAIL WILL BE VERY DIFFERENT

The value of real estate is going to force many good nurseries out. Once again it is not that they aren't profitable it is just that the land they sit on in Sydney and other large centres including Newcastle is too valuable. You need a lot of land for a nursery and car park.

Councils with their usual foresight won't treat nurseries differently and so new ones won't be able to set up easily.

The lazy assumption is that more business will end up at the chains. It could initially but it won't eventually. I think you will find that new innovative retailing will spring up. Gardening is such a universal passion that it needs to be able to be serviced much more effectively than large "sell it all stores" can do and entrepreneurs will see opportunities. I think these are likely to involve the satellite store – small shops in the city that are serviced by a large retail centre on the peripheral area of the city, and the pop up store. A business which moves into a vacant shop for 4 months on a short term lease, sells high impact product and then closes. They may combine satellite stores with florists or stores selling other beautiful things and there is good potential there for someone who produces high-end, high-impact product.

Gardening will be driven further underground and more and more true gardeners will buy plants online and have them delivered by post not because they want to but there won't be an available option for them to get what they want.

Is there a future for small nurseries?

Smaller nurseries will still face the same problem they do now; growing plants is cheap, selling them expensive. They don't have the efficiency of scale that larger players do but there are ways around this.

I can think of an example of a small nursery that producers a lot of plants at cheap prices and rides on the back of the large landscape supplying nurseries. Old pots, no labels, little machinery, hand written invoices and no delivering but when the big places run out they have the numbers and so they can pick up. For this to work the nursery needs to be located in the right place. Their prices can be low because their costs are kept down and they only grow plants with a reasonable shelf life. So it is not always about keeping prices up. It sounds simple but profit is simply about the relationship between cost and sale price. It is not about one independent over the other.

Our response and situation is quite different

We have changed what we grow a lot over the years. We started growing trees and shrubs but have ended up growing hydrangeas and flowering plants. I try to focus on plants that will perform in the garden, both in the types we select and how we grow them. I do this for the gardening public. For myself I try to grow what will make the most money for the least effort.

It's funny how conditioned we are because when I was thinking of ways to express that sentiment I thought that is the truth of it. This makes us sound lazy and perhaps mercenary in our attitude and we are anything but this. At least by doing this we can make a reasonable living for working reasonably hard. So we grow the best plants we can and hold the line on price.

I have put most of my energy into working out how to grow the plants I choose to grow, better. I have never thought about marketing. We don't do a sale run and we don't chase new customers. We only invest in basic technologies.

We've always worked on the premise that if I think I can sell 1000 of something we grow 700 and remind those who miss out to be quicker next time. Almost everything we grow has a relatively short sale window. We don't try to sell volume outside of spring and accept that the market is seasonal. So we only deliver fortnightly outside of spring and keep the nursery two thirds empty over summer.

We have a van which is cheaper to run than a truck and more comfortable to drive. We don't grow plants over 40 cm tall because they take up too much shelf space. I explain all this to my customers, if they ask and they agree because they want us to be there for next spring. We produce a lot fewer plants than the first example and we sell at a higher price but we are very conscious of our costs.

CONCLUSION

The examples of I have chosen have very different approaches to solving the problem of remaining profitable. There is no one, single, correct way.

There is a great future for ornamental horticulture but we are going to have to overcome a few difficulties in the next few year. As long as the operator doesn't confuse growing more without considering his selling costs there is a glowing future for the small player as well.

Revegetating farmlands in northern New South Wales: problems and solutions©

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INTRODUCTION

Why is revegetation important?

Since European settlement, land clearing for agriculture has led to the widespread destruction, modification, and fragmentation of Australia's native vegetation (Atyeo and Thackway, 2009; Bennett and Saunders, 2010; Yates and Hobbs, 2000). These changes have occurred so quickly that plant and animal communities have had little time to adapt, leaving them vulnerable to extinction (Bauer and Goldney, 2000). Consequently, Australia rates high on the extinct species list, particularly for mammal extinctions, which over the past 200 years has been higher than any other continent (Cardillo and Bromham, 2001; Hobbs and Mooney, 1998; McConnon, 2015). One way of addressing our declining biodiversity is by revegetating farmlands previously cleared for agriculture (England et al., 2013; Vesk and Dorrough, 2006). Studies have shown that revegetation, which is structurally and floristically diverse, is important for conserving biodiversity because it provides nesting, perching and shelter for birds, microhabitats for seedling establishment, and sources of food and shelter for fauna (Collard et al., 2013; Munro et al., 2009). Effective conservation in rural environments also requires interconnected networks of native vegetation that together, have the capacity to support large populations of native flora and fauna (Bennett and MacNally, 2004). In this respect, revegetation can function as stepping stones or continuous corridors to allow movement between subpopulations, thus maximising the persistence of a species and minimising inbreeding depression (Bennett and Saunders, 2010; Hilty et al., 2006).

However, the idea of integrating natural resource management with agriculture to achieve a more sustainable landscape has been met with some resistance. Specifically, landholders are concerned that the two are incompatible, particularly in terms of the constraints that native vegetation places upon agricultural productivity and land management flexibility (Schirmer and Bull, 2014). On the upside, traditional beliefs are waning amidst the increasing realisation that the agricultural industry is directly dependent on native vegetation for a range of vital ecosystem services (Fischer et al., 2006). These services include the provision of clean water, healthy soils and important crop pollinators, the regulation of pests and diseases, and the mitigation of salinity and soil erosion (Fischer et al., 2006; Wallace, 2007). In addition, native vegetation acts as a potential genetic storage for the future improvement of crop species (Altieri, 1999; Fischer et al., 2006).

Revegetation techniques

Revegetation techniques generally fall into three categories: (1) natural regeneration, (2) direct seeding, and (3) tubestock planting. Natural regeneration is often the preferred method of revegetation because it is cost effective, doesn't require planting or management input, and has the added advantage of retaining the character and native species of an area (Curtis, 1990; Whisenant, 1999). It is underpinned by the process of succession and is based on the premise that once disturbances impacting on the ecosystem are ameliorated, plants will naturally re-establish through vegetative means, or natural seed fall (Miller et al., 2013). The best way to encourage natural regeneration of native trees and shrubs is to exclude

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stock from the area for at least 5 years (Curtis, 1990). However, severely degraded farmlands that have been intensively managed for long periods of time are very slow to regenerate with little to no regeneration capacity (Cummings et al., 2007). Eucalypts in particular, are difficult to re-establish in rural landscapes because seed survival is low, even following a heavy seed fall (Curtis, 1990). The first step towards successful natural regeneration often involves identifying the restoration barriers (biotic and abiotic), which prevent the transition of the degraded landscape into a more desirable state (Cummings et al., 2007; Whisenant, 1999).

Direct seeding is a relatively inexpensive technique, which lends itself particularly well to broad-scale revegetation in agricultural settings (Dalton, 1993; Florentine et al., 2011). Various methods are used to introduce seeds into new sites, including broadcast seeding, aerial seeding, hydro-seeding and seeding using commercial agricultural seeders and drill seeders (Greipsson, 2011; Whisenant, 1999). Past studies have shown high variability in the success of direct seeding as a revegetation technique, particularly in parts of New South Wales (Carr et al., 2009; Van Andel and Aronson, 2012). This is most likely because of prolonged dry periods, lack of defined winter wet season and inadequate weed control (Geeves et al., 2008).

In unpredictable and inhospitable environments where direct seeding operations seldom succeed, transplanting whole plants is the most viable alternative (Whisenant, 1999). Although the cost of tubestock plantings is high, the technique is often preferred because trees can be established in the landscape in a relatively short timeframe and the results are immediate (Rawlings et al., 2010). Seedlings can be planted using a mechanical planter consisting of a ripper tyne, furrow opener, plant delivery system and a press wheel. Alternatively, seedlings are planted by hand using either a Hamilton planter or a Pottiputki planter (Namoi Catchment Management Authority, 2013). Choosing healthy, disease-free seedlings with good root development is critical to short-term and long-term survival (Rawlings et al., 2010), along with good site preparation and weed control (England et al., 2013).

PROBLEMS AND SOLUTIONS

Revegetation is one of the most expensive natural resource conservation activities (Florentine et al., 2011). Since the mid-1990s the Australian government has invested millions of dollars in revegetation programs largely for vegetation re-establishment (England et al., 2013). For example, in 2000-2001 alone AU$36.4 million dollars was spent to re-establish native vegetation and to provide appropriate habitat for wildlife (Florentine et al., 2011). Despite this investment, little to no follow-up monitoring has been done to assess the effectiveness and success of revegetation projects (Atyeo and Thackway, 2009; England et al., 2013; Florentine et al., 2011). Of those projects monitored from the 1970s to the present, many have been unsuccessful in terms of survival (Freudenberger and Harvey, 2003). Factors affecting survival include climate, soil type, previous land use, and poor establishment techniques (Andrews, 2000; Close and Davidson, 2003; England et al., 2013).

My research is based on the Northern Tablelands of New South Wales, Australia. The primary aims of my research are: (1) to evaluate the success of past and present revegetation projects in terms of tree performance, (2) to investigate different planting and management techniques to increase the germination and survival rates of direct seeding and tubestock planting, and (3) to provide scientifically based guidelines for landholders, revegetation practitioners and NRM organisations. I discuss below some of the primary problems impeding the success of direct seeding and tubestock planting on the Northern Tablelands.

Poor species/provenance selection

Revegetating altered landscapes to achieve diverse, functional systems rich in native species requires vast quantities of seed (McKay et al., 2005). The decision as to where to source this seed has caused conflict among revegetation practitioners and organisations over past decades (Hancock and Hughes, 2012). Traditionally, the paradigm of “local is best”

has been widely advocated, and using local seed still remains a goal for much of the revegetation work undertaken in Australia (Broadhurst et al., 2008; McKay et al., 2005; Williams, 2007). It is based on the premise that local provenance seed delivers better revegetation outcomes because it is adapted to local conditions and, therefore, it reduces the risk of genetic pollution and outbreeding depression (Broadhurst et al., 2008; Hancock and Hughes, 2012; Williams, 2007). However, as revegetation targets increase and research into the effectiveness of current strategies intensifies, the appropriateness of the local provenance paradigm has come under review (McKay et al., 2005).

A fundamental problem underpinning the "local is best" paradigm is that there is no universal definition for "local" (Carr, 2008; Hancock and Hughes, 2012). Although local provenance is almost always determined by spatially explicit guidelines, these vary among revegetation practitioners and organisations (Hancock and Hughes, 2012). A second is that seed harvesting to meet growing revegetation targets is likely to impact on remnant populations by reducing seed availability for natural population turnover, or reducing plant vigour through collateral collection damage (Broadhurst et al., 2008). Moreover, as our understanding of the demographic and genetic effects associated with landscape fragmentation broadens, there is growing evidence that using locally adapted seed may be consigning the progeny to a genetic dead-end (Williams, 2007). Such evidence identifies the need for a less restrictive approach to seed collection, with a view towards composite provenancing or mixing seed from sources from within the species' natural range to minimise the risk of inbreeding and to promote genetic diversity within newly planted areas (Carr, 2008; Williams, 2007). The latter is critically important given that genetic variation is an essential prerequisite for evolutionary change and the persistence of species in changing environments, particularly in relation to current climate change predictions (Hancock and Hughes, 2014).

Inadequate ground preparation

Inadequate ground preparation is often the downfall of revegetation efforts in northern New South Wales (Andrews, 2000). This is probably because existing protocols are not being adhered to, or corners are being cut due to time constraints, poor planning, or unsuitable machinery and equipment. Characteristics of a well prepared site include friable, aerated soil, good moisture availability throughout the soil profile, and soil that is free from competing weeds and weed seed burdens at the time of planting (Andrews et al., 2004). Furthermore, the longer a site is prepared prior to planting the better the performance of the trees subsequently planted there (Andrews, 2000). Andrews et al. (2004) recommends the following protocols for establishing trees on the Northern Tablelands:

- Slash or crash graze the site to remove heavy accumulations of herbage.
- Commence fallow using a knock-down herbicide or by cultivation 10-12 months prior to planting. Maintain fallow.
- Deep rip (500-600 mm) planting beds soon after commencement of fallow. Soil should be dry to ensure a good shatter at depth.
- Mound and/or cultivate 6 months before planting once good root release is achieved from the first herbicide application.
- One month before planting apply a residual herbicide along with another application of knock down herbicide if needed.

Bad timing

Planting at the right time is essential for successful revegetation. Site conditions such as rainfall, temperature and soil moisture all play crucial roles, whether direct seeding of planting tubestock (Namoi Catchment Management Authority, 2013). When planting tubestock, past research has shown that adequate water availability at seedling establishment phase, along with good soil moisture stores prior to planting, enhance seedling survival (Andrews et al., 2004; McGinness et al., 2007). Therefore, taking advantage of windows of opportunity that maximise the likelihood of follow-up rain and minimise evaporation rates is an important strategy (McGinness et al., 2007). For the Northern

Tablelands and Northwest Slopes and Plains, spring planting is recommended, however when planting in free-draining soils, where soils dry out faster than seedlings can extend their roots, autumn planting is an option (Andrews et al., 2004).

The timing of direct seeding is complicated as germination is cued by a complex combination of soil moisture, temperature, light, day length, and chemical signals, often in a specific order (Carr et al., 2009). Most NRM organisations and revegetation practitioners recommend sewing at a time when there is the highest probability of these optimal conditions occurring. If germination is delayed by planting out of season, dormant seeds are more at risk of desiccation, predation and disease (Carr et al., 2007). Generally, direct seeding should occur in winter and early spring in winter-dominant rainfall zones, while in summer-dominant rainfall zones mid-spring or autumn plantings are recommended. However, sowing times will vary according to the species planted, and conditions at the time of planting. For example, often a compromise between sowing early to maximise soil moisture availability and sowing later when temperatures are high enough to stimulate germination, will need to be made (Carr et al., 2007).

Overall, bad timing may not only be attributed to a lack of local knowledge, but also the need to push revegetation projects through in order to meet annual revegetation deadlines. In addition, plantings often occur out of season to relieve pressures from nurseries needing to offload seedlings pre-ordered for different NRM organisations. Succumbing to these pressures should be avoided, as the success of all planned revegetation is fully dependent on the survival and establishment of germinants and seedlings.

Inhospitable climates

Low temperatures represent one of the most harmful biotic stressors affecting temperate plants (Janská et al., 2010). In areas of northern NSW, cold temperatures and severe frosts limit the survival and growth of native trees, particularly in parts of the landscape where cold air drainage occurs (Reid et al., 2012). We assessed the impact of the physical environment on the survival and growth of three eucalypt species: *Eucalyptus nitens, E. pauciflora*, and *E. viminalis* on the Northern Tablelands (Figure 1). Using multimodel inferencing we identified minimum temperature to be one of the main abiotic stressors affecting tree performance. Other studies have reported similar findings in relation to cold stress in eucalypts (Ball et al., 1991; Green, 1969; Harwood, 1980; Leslie et al., 2013; Paton et al., 1979). Extreme cold may restrict plant survival directly, through mechanical injury, or indirectly by shortening the growing season. Further, a reduction in the growing season does not allow adequate time for photosynthetic-driven carbon gain or for recovery from grazing or frost damage (Reid and Palazzo, 1990).

Figure 1. Failed revegetation as a result of extreme cold temperatures, Armidale, NSW, August 2013 (photograph by S.L. Brown).

Frost damage generally causes injury to plant cells, either through the mechanical rupture of the cell membrane and cell wall, or through an imbalance in electrolytes as freezing removes water from solution (Reid and Palazzo, 1990). However, there is an abundance of research, which has shown that cold temperatures also interact with light stress to damage plants (Ball et al., 1991; Blennow and Lindkvist, 2000; Godde et al., 1992; Hayden et al., 1986; Osmond et al., 1987). This phenomenon is known as cold-induced photoinhibition. Photoinhibition occurs in all photosynthetic organisms because light is the driving force for photosynthesis (Murata et al., 2007). Light induces the production of reactive oxygen species (ROS), which inactivate the photochemical reaction centre of photosystem II (PSII) (Murata et al., 2007). Under normal circumstances, the PSII is able to repair itself quickly and efficiently through the synthesis of proteins (Ball et al., 1991; Murata et al., 2007). However, exposure to low temperatures increases a plant's sensitivity to light and inhibits protein synthesis, so that the damage to PSII occurs more rapidly than it can be repaired (Ball et al., 1991; Blennow and Lindkvist, 2000; Murata et al., 2007).

Water availability

Water is an essential requirement for all living organisms. In herbaceous plants, the water content ranges from 70-95%; however, it is continually lost from a plant through the processes of transpiration and photosynthesis (Passioura, 1982). The importance of managing soil moisture conservation in all revegetation (before and after planting) cannot be overstated. This requires effective planning and management, including adequate ground preparation and weed control, mulching, and follow up watering (McGinness et al., 2007), although in reality, this rarely occurs. Knowing the site and the soil water holding capacity, along with choosing drought tolerant species are also important factors. Surprisingly, the benefits of artificial irrigation to seedling survival have not been widely studied. One Australian study undertaken at Gungahlin, ACT, demonstrated a high survival rate (84-96%) of eucalypt seedlings planted into a soil profile that was artificially filled with water (McGinness et al., 2007). An earlier study (Yantabulla, NSW) reported that summer irrigation maintained the survival of *Dodonaea viscosa* subsp. *angustissima* (syn. *Dodonaea attenuate*) seedlings (>80%) planted in a natural grassland compared to zero survivorship in the unirrigated control (Harrington, 1991).

We investigated the effect of deep watering on the survival of *E. populnea* and *Casuarina cristata* seedlings in Narrabri, NSW. A total of 120 seedlings of each species were planted along with an equal number of control seedlings. The planting took place in the summer of 2014 so we were able to affectively assess the impacts of heat stress and water availability on seedling establishment. Treatment seedlings were planted within paper pulp cocoons, a Dutch designed watering system that held 25 L of water made available to the seedlings through a nylon wick (Figure 2) (Land Life Company, 2015). Controls were planted using a Hamilton planter, with each seedling receiving 5 L of water through a ground spike before planting and an additional 5 L after planting. Five months post planting 95% of the treated seedlings had survived, compared to 0% survival in the controls. We concluded that watering protocols need to be established and the associated costs factored in at the initial planning stages of revegetation to ensure seedling survival. Seedlings should receive at 5-10 L of water at the time of planting to avoid transplant shock and further follow up irrigation at 4 week intervals (during summer) and 8 week intervals (during winter) if reasonable rain is not forecast (Namoi Catchment Management Authority).

Figure 2. Land life box trials, Narrabri, NSW, October 2014 (photograph by S.L. Brown).

Poor weed control

Revegetation practitioners identify poor weed control as one of the primary factors affecting the successful establishment of trees in rural landscapes (Andrews et al., 2004; Hall, 1985). Weeds can reduce early growth rates by up to 70% and decrease survival to as little as 10% (Greening Australia, 2003). It is the very nature of weeds that makes them a such problem, for example, weeds are usually early colonisers, exhibit rapid growth, reproduce prolifically and can withstand an array of environmental challenges. Consequently, weeds compete very effectively against planted seedlings for light, nutrients, and water (Figure 3). Effective weed control may encompass various techniques, including the use of mulches and weed mats, scalping, cultivation, flaming, hand removal, and chemical control (Greening Australia, 2003; Namoi Catchment Management Authority, 2013; Taylor, 2013). Weed control for direct seeded sites is particularly challenging, as it is difficult to avoid spraying the germinated seedlings. Traditionally, weeds have been managed by long-term weed control before sowing, or by applying a knockdown herbicide in the months preceding, followed by a residual just before sowing. However, this practice is not always effective (Semple and Koen, 2006; Taylor, 2013). This raises the question as to whether herbicide oversprays could be used as an effective method of weed control if some native species exhibit tolerance to them (Semple and Koen, 2006). Moore (1999) used this technique to demonstrate the tolerance of 21 of 26 native species to chlorthal and napropamide, while approximately half of the species tested were tolerant to chlorsulfuron and imazethapyr.

We investigated the tolerance of 12 native tree and shrub species to nine residual (broadleaf and grasses) herbicides oversprays. These trials took place in a temperature controlled glasshouse at the University of New England between December 2014 and February 2015. Our results were variable in that different species exhibited tolerances to different herbicides. Survival rate for seedlings treated with Jaguar® (diflufenican) was zero for all species. *Eucalyptus*, *Acacia*, and *Dodonea* species exhibited tolerance to Goal® (oxyfluorfen). *Casuarina*, *Senna* and *Acacia* species exhibited tolerance to Spinnaker® (Imazethapyr), while all species exhibited tolerance to Amitrole T (amitrole) and Balance™ (Isoxaflutole). We are currently investigating the effectiveness of herbicides against weeds and testing the tolerance of the same suite of species in situ. Clearly there is scope for continued research on herbicide tolerance of a broad range of species.

Figure 3. Infestation of thornapple (*Datura* sp.) at a direct seeded site 5 months post-planting Bingara, NSW, March 2015 (Photograph by S.L. Brown).

Planting against the contour

Although one would expect that planting along the contour would be a fundamental principle of successful revegetation, sometimes it is not practiced. The main problem associated with planting against the contour is the increased risk and severity of erosion, especially in the event of a rainstorm (Figure 4). Figure 4 shows a site that encompasses 10 km of direct seeding and tubestock in undulating basalt country near Ben Lomond, NSW. Two months after planting the area received heavy rains over a short period of time, resulting in widespread damage to the planting.

Figure 4. Erosion along direct seeded ripline after heavy rain Ben Lomond, NSW, April 2015 (photograph by S.L. Brown).

Survival and growth rates also differ in trees planted against the contour due to environmental gradients, which correspond to the natural topography of the landscape (Ferrero et al., 2013). Topography substantially modifies local environmental conditions by altering the regional climate and its interactions with soil properties (Ferrero et al., 2013). Consequently, trees planted along environmental gradients often display variations in their response to local environmental conditions, particularly in relation to frosts and

waterlogging (Davidson and Reid, 1985; Gilfedder, 1988). Inverted tree lines are very good visual representation of how patterns in tree height vary along an environmental gradient.

Planting along the contour not only rectifies these problems, but affords many other benefits. For example, contour plantings allow runoff to be collected along the prepared bed, which in turn allows water to soak in and directly benefit the survival of direct seeded germinants and tube stock (Namoi Catchment Management Authority, 2013). It also facilitates the retention of leaf litter, thus reducing the loss of natural resources from the site, improving the foundations for ecological functioning (Mullan, 2000).

CONCLUSIONS

The purpose of this paper is to provide practical solutions to problems that my research has identified as being detrimental to revegetation success. However, these problems are not new. The last 40 years of revegetation efforts have liberated an abundance of planting protocols and recommendations, which have been made available to a wide range of NRM audiences. The challenge, therefore, is to establish clear communication pathways between revegetation organisations, revegetation practitioners and landholders, so that advice can be freely available at all stages of the project, monitoring procedures can be established and implemented before and after planting, expectations can be clarified, and the risk of failure can be managed so that successful revegetation can be achieved.

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Climate variability and risk management in nursery production©

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INTRODUCTION

One of the key skills of a nurseryperson is the ability to observe small changes in plant growth and the growing environment, and to understand how these factors influence plant production systems. This training in intuitive observation may explain why some growers comment that the seasons are not what they were. They may perceive that winters are warmer; that flowering and seeding is earlier or that rainfall patterns have changed in their area. Is this anecdotal evidence of increased variability in our climate, or just random musings?

The theme of the IPPS Australian Region conference, "The Times Are Changing", provides an opportunity to be future focused, and arguably the most significant change of our times is our changing climate. If the seasons are changing, then our plants and our livelihood as growers of living products will also change. The purpose of this presentation is to highlight the latest scientific research relating to the effects of climate change both globally and locally. It introduces the concept of risk management relating to business planning and discusses how nursery businesses can source local climate projections to plan for climate variability. Finally, I suggest potential opportunities for nursery businesses to engage with climate issues and position themselves firmly as being part of the solution.

DISCUSSION

In the beginning…greenhouse gases and the atmosphere

We know that life on earth is powered by the sun. The average global temperature is a comfortable 15°C and this is due to the naturally occurring presence of a number of 'greenhouse gases' in our lower atmosphere; mainly water vapour, carbon dioxide, methane, and nitrous oxide. These gases absorb heat radiated back from our earth and this warms the earth's surface and lower atmosphere, creating the conditions that sustain life. NASA scientists (2015a) note that our earth's atmosphere has been through various phases of heating and cooling for at least 650,000 years. However, they make the point that atmospheric carbon dioxide (CO_2) has never been above 300 parts per million (ppm) during this period. Figure 1 illustrates the rapid increase in CO_2 levels in recent times and scientists believe that warming of the atmosphere is linked to the increase in CO_2 levels (2015b). Regular updates on current CO_2 levels are available online from http://co2now.org/Current-CO2/CO2-Now/. This data is from the Scripps CO_2 Program (2015) at the Mauna Loa Observatory in Hawaii – the longest-running, high-precision instrument record for atmospheric CO_2. In January this year their sensors recorded atmospheric CO_2 of 399.73 ppm and in April a preliminary monthly average of 403.26 ppm. So what does all of this mean?

The Intergovernmental Panel on Climate Change (IPCC) reports that: "The current warming trend is of particular significance because most of it is very likely human-induced and proceeding at a rate that is unprecedented in the past 1,300 years" (IPPC, 2007:5).

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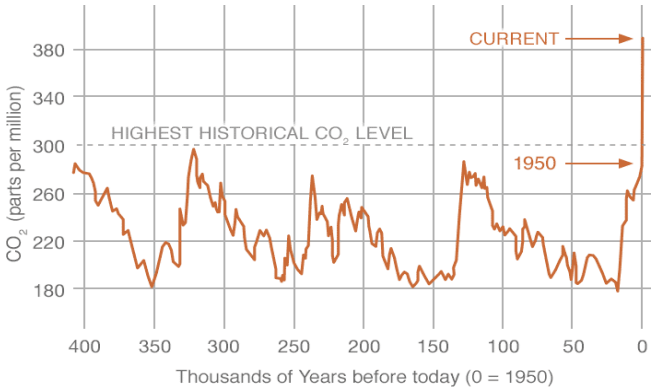

Figure 1. Graph of CO_2 levels during the last three glacial cycles, as reconstructed from ice cores. Data source NOAA. Accessed from NASA (2015b).

Human-induced impacts

Humans have always sought to modify their environment to provide for their needs and to ensure a greater chance of survival. However, since the industrial revolution, human impacts have increased with the extraction and burning of fossil fuels, the large scale burning of vegetation, increase in stock numbers producing methane and the use of nitrogen fertilisers. While the amount of methane and nitrous oxide has increased in the atmosphere, concern is focused around CO_2 levels. This is because CO_2 lasts for a long time in the atmosphere, and it is therefore considered to have a greater long-term warming effect (IPCC, 2014). New Zealand's CO_2 emissions are lower than Australia's because New Zealand has one of the highest levels of renewable electricity generation in the world. However, around half of its greenhouse gas emissions (which include methane and nitrous oxide) come from agriculture, creating debate about how to maintain agricultural development while reducing methane emissions (Ministry for the Environment, 2015). This compares with agriculture in Australia contributing approximately 16% of its greenhouse gas emissions (NGIA, 2014).

As climate scientists report a greater level of scientific certainty that global warming is mostly due to humans and not natural forces, governments are being called to show greater leadership to cut greenhouse gas emissions. The target is to attempt to keep any temperature increase below 2°C relative to pre-industrial levels. "If greenhouse gas emissions continue to increase at the current rate for a few more decades, we are likely to see average global temperatures warm by more than 4°C by 2100" (Ministry for the Environment, 2015).

Intergovernmental panel on climate change

There are many scientific research institutions around the world working to gather, analyse and report on atmospheric changes and their link to climate change. The lead organisation in this process is the Intergovernmental Panel on Climate Change formed by the United Nations and tasked with gathering together global research and reporting on this every 5 to 7 years. In 2014 the IPCC released its Fifth Assessment Report produced by more than 700 scientists and additional comments on drafts were received from 1,700 expert and government reviewers (Hughes, 2014). Summaries of the IPCC's Fifth Assessment Report have now been released by many countries' scientists and these deserve to be widely read (Hollis, 2014; Hughes, 2014). The latest IPCC report confirms:

"That human influence on the climate system is clear and growing, with impacts observed across all continents and oceans. Many of the observed changes since the 1950s are unprecedented over decades to millennia. The IPCC is now 95% certain that humans are the main cause of current global warming. In addition, the SYR (Synthesis Report) finds that the more human activities disrupt the climate, the greater the risks of severe, pervasive and irreversible impacts for people and ecosystems, and long-lasting changes in all components of the climate system" (IPPC, 2015).

Global effects of climate change

Some of the global effects of climate change reported by the IPCC (2014) are:

- Increased air temperatures and more frequent and intense heat waves:
  The global mean surface temperature change for the period 2016-2035 relative to 1986-2005 is similar for the four Representative Concentration Pathways (RCPs) or greenhouse gas concentration scenarios and will likely be in the range 0.3 to 0.7°C.
- Increase in the frequency and intensity of rainfall events over some regions
- Oceans are continuing to warm and become more acid:
  Since the beginning of the Industrial Revolution, the acidity of surface ocean waters has increased by about 30% (PMEL Carbon Program, 2015).
- Global sea levels are rising:
  Global sea level rose about 17 cm in the last century. The rate in the last decade, however, is nearly double that of the last century (Church and White, 2006).
- Plant and animal ranges, migration patterns and behaviors such as flowering have changed:
  "A large fraction of species faces increased extinction risk due to climate change during and beyond the 21st century…" (IPCC, 2014).
- Glaciers have shrunk, ice on rivers and lakes is breaking up earlier:
  NASA's data (2015a) shows that Greenland ice sheets lost 150 to 250 cubic kilometres of ice per year between 2002 and 2006, while Antarctica ice sheets lost about 152 cubic kilometres of ice between 2002 and 2005.

Key risks for New Zealand and Australia from climate change

For most people the global scale of climate change is almost too overwhelming to relate to. However, closer to home, information on the key risks from climate change in the 21st century is available for Australia and New Zealand. Hollis (2014) and Hughes (2014) both summarise aspects of the risks outlined in the IPCC.

Working Group II assessment report.

Three key risks for both countries combined are:

- Increased frequency and intensity of flood damage to settlements and infrastructure.
- Increased damage from wildfires .
- Increasing risks to coastal infrastructure and low-lying ecosystems from continuing sea level rise, with widespread damage if the more severe projections are realised.

Five other key risks for Australia only are:

- Damage to coral reef systems
- Shrinking mountain habitats and loss of some native species due to increasing temperatures and fire risk
- Constraints on water resources in southern Australia due to higher temperatures and decreased rainfall
- Increased illness, death and infrastructure damage during heat waves
- Reductions in agriculture production in the Murray-Darling Basin and south western and south eastern Australia due to dry conditions

Risks to nursery production

A sector based on the production of plants is particularly sensitive to climate change effects given that plants are living products. Changes in temperature, water, carbon dioxide levels, pollinators, and micro-organisms will have a significant impact on growth and reproduction. Nursery production is also affected by decreases in gardening demand due to bad weather; be it too wet, dry, hot or cold and also from damage to nursery physical structures and wider transport infrastructure from storm events.

Some other potential risks relate to the effects of increasing temperatures driving increased evaporation rates, making water resources increasingly scarce for production. Competition for water resources will increase as rainfall patterns change and some regions become drier. Less water and increased temperatures will also affect how biodiversity copes with climate change (CSIRO, 2014). A warming environment means that weeds, pests and

diseases may expand into new areas creating problems for growers.

A conference, "Species on the Move" to be held in Hobart in 2016, notes that "the global redistribution of our planets' species is widely recognised as a fingerprint of climate change." The IPCC (2014) also reports that "most plant species cannot naturally shift their geographical ranges sufficiently fast to keep up with current and high projected rates of climate change in most landscapes.

For those growers involved in native plant production, an interesting change to seed and cutting sourcing is emerging, known as climate adjusted provenancing. This concept is being discussed by geneticists as an important strategy for some species to improve their chances of adaptation and survival. Rather than using local seed, in some circumstances, it may be better to source genetic material from outside the area and to actively move genes into their potential future ranges. Concepts of provenance and its interactions with climate change, are being explored by organisations such as the Society for Ecological Restoration Australasia, and the Australia Network for Plant Conservation.

So the question is how do we prepare for increased uncertainty and what can we do to lower the risk to our businesses to ensure they are sustainable into the future?

Risk management

"Risk is the combination of the likelihood of occurrence and the magnitude of the consequence of a hazard. It is a useful concept for dealing with an uncertain future" (UK Climate Impacts Programme).

It is normal business practice to plan ahead to manage risk. Although the exact impacts of climate change are uncertain, they can be managed like any other business risk. The point is to start the discussion now with your staff and identify a few key risks and develop an action plan to make your business more resilient to these risks. Use the issue as a catalyst to review and focus on what you can control, rather than worrying about all the things that you cannot. Remember that climate variability will also bring business opportunities as well as threats as will be discussed in later sections of this presentation. Whether you're sceptical about climate change or not isn't the point, it is what your clients' think about the issue, that is the point.

The UK Climate Impacts Programme (UKCIP) has excellent resources for encouraging businesses to take a planned approach to climate change. They list six potential impacts that climate change has on businesses (UKCIP, 2010).

These are:

- Markets: e.g. demand, product mix, diversification
- Logistics: e.g. supply chains, utilities, transport
- Process: e.g. plant growth factors, resource use
- Finance: e.g. insurance, price positioning, costs
- People: e.g. consumer behaviour, demographics
- Premises: e.g. structures, design, energy needs

Businesses can adapt to climate change risks in a number of ways depending on what their key risks are. In regions that may become drier, securing additional water resources and fire mitigation may be a priority. For others in low lying areas, where extreme weather events such as flooding may increase, moving to alternative power sources such as solar power, having emergency backup supplies and other storm mitigation plans may be their focus. Protecting your business financially from climate impacts should include having adequate insurance and business continuity cover in place, as well as security for electronic records to minimise disruption due to adverse events. Training staff for emergencies and investing in their health and safety and professional development also builds resilience in your business.

There are a number of models for managing risk (Ministry for the Environment, 2008; UKCIP, 2010). In general, they involve a process of firstly identifying the potential hazards and risks involved; analysing these risks, and evaluating them against set criteria to prioritise and identify key issues; then identifying adaptive measures including the costs and benefits, and selecting action measures to implement. Whatever process of planning you use,

the key thing is to start now!

Getting started – where to look for information

To get started you will need the best resources, tools and support. The good news is that there is a wide range of helpful resources out there that are easy to access on the internet (Table 1). Some organisations and their websites worth looking at are:

Table 1. List of organisations with useful resources relating to climate change.

Organisation	Web address	Resources
Climate Change in Australia	http://www.climatechangeinaustralia.gov.au	Climate Futures Web Tool Users can manipulate data to try out various climate impact scenarios for their area Cluster brochures and reports Maps and climate change projections for all regions of Australia
Climate Council of Australia	http://www.climatecouncil.org.au/category/reports	Reports on climate change issues and projections of impacts on various areas
New Zealand Climate Change Centre	https://www.nzclimatechangecentre.org/	Climate information and a searchable database that links users to active climate change research
UK Climate Impacts Programme	http://www.ukcip.org.uk/decision-making-for-adaptation/	Business Areas Climate Impacts Assessment Tool Scoping impacts of climate change UKCIP Adaptation Wizard Online tool to help you adapt to climate change
National Oceanic and Atmospheric AdministrationNational Centers for Environmental Information (NCEI)	http://www.ncdc.noaa.gov/climate-information/climate-change-and-variability http://www.ncdc.noaa.gov/climate-information/statistical-weather-and-climate-information	Global climate at a glance Various maps e.g. temperature and rainfall changes and access to climate data records
National Aeronautics and Space Administration	http://climate.nasa.gov/evidence/	Climate change information and resources; mitigation and adaptation strategies and technologies

Future focused opportunities

Threats from increasing climate variability have been well covered by the media to the point of fatigue or disbelief depending on your point-of-view (Lloyd, 2015a, b; Asten, 2015). The scale of the problem is very serious, however, there are some positive opportunities specific to the nursery production sector.

1) Plants not only sustain all life and but as part of their photosynthesis processes they also absorb carbon dioxide. As more people equate greening the planet with saving the planet, plant propagators should be seen as the experts in plant based climate change solutions in their communities. The Nursery and Garden Industry Australia (NGIA) puts it plainly when they say "the sector can play a vital role in preventing, stabilising and reversing environmental degradation".
2) Urban greening campaigns such as 202020 Vision which aims to create 20% more green space in Australian urban areas by 2020, increase the public's understanding of the value of plants as well as stimulating demand. Similarly Greening Australia's joint project "One Million Trees" launched in 2014, will see the planting of one

million trees south of Perth, and in western Victoria. The trees planted will not only capture thousands of tonnes of CO_2, but also restore threatened habitats. Both projects have in common significant community involvement. The forestry industry also promotes the value of forests as carbon sinks.

3) As the climate changes in regions, there are opportunities to provide clients with new types of plants adapted for these conditions. Biodiversity conservation will become critical and the demand for native plant restoration skills and seed resources will increase.
4) The world population has now reached 7 billion and is expected to reach 9 billion by 2050. It is estimated that 70% more food will be needed to feed the world population by 2050 (Ministry for the Environment, 2015). Opportunities for food production exports, and therefore the starter plants for the vegetable and fruit production sectors, will increase. Unfortunately, crop and agricultural food production has already decreased in parts of Africa and Europe due to climate change (Hughes, 2014). New Zealand and Australian growers may have a competitive resource and climate advantage in meeting these food demands.
5) New growing technologies achieving energy, irrigation and spatial efficiencies are being developed all the time.
6) Many opportunities exist for growers to educate themselves about how to increase the sustainability of their businesses in the future. Nursery and Garden Industry Australia already offer accreditation schemes to improve nursery practices such NIASA-BMP, EcoHort®, BioSecure. In addition to this, policies on climate change and sustainability already exist in the nursery industry to provide additional guidance for the future (Nursery and Garden Industry Australia, 2011, 2014; Oregon Assoc. of Nurseries, 2011).

CONCLUSION

If the times are changing, what is your vision for the future? The good news is that climate researchers confirm that we have the ability to limit climate change and its risks, with many solutions that still allow for development. The sense of urgency about the level of change required is clear: "…stabilizing temperature increase to below 2°C relative to pre-industrial levels will require an urgent and fundamental departure from business as usual" (IPPC, 2015).

Growers can plan ahead to minimise the risks of climate variability on their businesses. There are some useful tools and resources that have been developed internationally, nationally, and no doubt at a local level, by local government and community leaders already planning for these risks in your area. Opportunities do exist for growers to position themselves positively and raise consumer awareness of the value of plants in their communities.

We need to continue to look innovatively at how we reduce our greenhouse gas emissions and examine our industry practices to reduce their impact on the environment. It may not be business as usual in the future, but there is a greater need than ever for plant production to be acknowledged as having an essential role in sustaining our collective future on this planet.

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Onwards and outwards©

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INTRODUCTION

Trees are an important and very valuable component of our urban landscapes. In a civic sense, they:

- Help reduce air temperatures
- Reduce electricity consumption (which in turn reduces the use of coal and demands on water)
- Sequester carbon
- Prolong the life of asphalt
- Improve the amenity value of our streets
- Help reduce storm-water runoff and much more.

On a more personal level, trees can improve our streets, enhance the quality of life we enjoy in our houses and gardens and add significantly to the value of our homes. When you look at the cost of the tree itself, the percentage of the total costs of planting is very small while the return on investment is huge.

Climate change with its associated increases in temperatures means that the contribution trees make will become increasingly important and the predicted increase in storm activity means that the structural integrity of our trees becomes even more important. The quality of the trees we grow is a key component of the success of those trees in our urban landscapes. All tree growth, both above and below ground, is determined by extension – the tree we plant is the foundation of the tree in the landscape. This is recognized by our knowledgeable end users and more enlightened growers from whim there is a strong push to improve the standard of trees grown and used in Australia.

Our industry is plagued with tree quality problems that relate to a reluctance to change existing production practices and the financial constraints and mechanisms we work under.

In the same way that the trees supplied by our advanced tree growers are the foundation of the trees in the landscape, the trees produced by our propagators are the foundation of these more advanced trees.

DISCUSSION

The value of trees

It has long been understood that trees provide a range of benefits to our urban environments. The benefits to the community include; reduced air temperatures, reduced electricity consumption (and the associated reduction in the use of coal and water) carbon sequestration, the increase in the lifespan of asphalt in our streets, capture rainfall in storm events therefore reducing runoff and the costs associated with dealing with that water, improve the amenity of our streets and increase tourism.

In a recent paper by Greg Moore (2016) some of these values have been quantified (see Table 1). Moore suggests that based on data collected for a community of 100,000 trees (a city roughly the size of Newcastle) each tree contributes of the order of $1400 per annum in measurable benefits — these do not include aesthetic and amenity values. Therefore, if we assume that these trees have a useful life of 40 years then each tree, in its lifetime, will contribute something of the order of $56,000 in measurable benefits to that community.

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Table 1. Quantified savings based on a community with 100,000 trees [based on figures found in Moore (2016)].

Benefit	Quantified benefits from 100,000 trees
Savings in electricity (based on a rate of $0.30 per kWh)	$1,000,000 per annum
Savings in water – as used to produce electricity only (based on a rate of $1.50 per kilolitre)	$450,000 per annum
Value of extending life of bitumen by 20-30 years, assumes 30% shade from the trees and that asphalt re-sheeted. (If old asphalt also needs to be removed, this figure is doubled and if full road reconstruction is needed, multiply it by four.)	$137,500,000
Value of sequestered carbon, based on a rate of $23 per tonne (as per the carbon tax)	$30,000,000

On a personal level, various real estate agents suggest that, for an average house worth around $500,000; a single tree in the garden can add anything from $5000-$25,000 to the value of the property, a garden of trees will add $25,000-$50,000 and established trees in the street will add up to an extra $150,000. While a Planet Ark survey found that 100% of buyers would pay an extra $35,000 if there were established trees in the street and 35% of buyers would pay an extra $100,000 (Table 2).

Table 2. Estimation of the value of trees and treed gardens [based on figures found in Moore (2016)].

Description of trees considered	Estimated increase in property value (For properties worth approx. $500,000)
Value added by a single tree	$5000
Real estate agent valuation	
Value added by a single tree [Real estate agent valuation]	5% of property value (or $25,000)
Value of established tree in the nature strip	30% of property values (or $150,000)
Value of a treed garden [Real estate agent valuation]	$50,000
Value of a treed garden [Real estate agent valuation]	5% of property value (or $25,000)
Value of a treed garden/street	$35,000 - $100,000

However, these amazing financial benefits are not gained by planting trees. They are gained through planting and establishing trees that succeed, in the longer term. Successful tree plantings are the result of a number of factors coming together – tree quality being one of them.

Planting substandard trees and therefore jeopardizing the tree planting in the process simply doesn't make sense. It is even more ridiculous when you consider the fact that the cost of the tree is typically a small percentage of the overall costs of the project and planting a poor tree is the same as planting a good tree.

For example, the recent removal of Hills Figs in Layman Street in Newcastle and replacement with new Hills Figs had a total cost of around $2,100,000 while the cost of the trees themselves was around $20,000 – less than 1%.

Climate change and tree quality

Google "climate change" and you will get something like 140,000,000 results in around 0.25 of a second. It is safe to assume that the predictions made by the scientists are real. We will be getting warmer and we will experience increased storm activity.

Increasing temperatures will make the benefits trees offer all the more important and the calculated $ values can only increase.

Perhaps more important will be the increased need for trees to be structurally stable so as to be able to withstand increased winds associated with increased storm activity

predicted.

As we better understand the importance of tree quality, as a component of success in the landscape and both The NATSPEC specification for trees (Clark, 2003) and the new draft Australian Standard AS 2303:2015 (2015) *Tree Stock for Landscape Use* (The Standard) now figure prominently in our industry, to continue to grow and supply trees that don't conform to these standards will be to leave yourself exposed to the possibility of legal action.

Stock quality as a component of success in the landscape

Successful tree plantings are the result of the following five critical components coming together:

1) Planning and design
2) Species selection
3) Stock selection (Stock Quality)
4) Planting and establishment
5) Maintenance

There is also a sixth critical component of successful tree plantings – communication. Those involved with the various stages of planting and establishment need to communicate with each other, if all stages are to come together cohesively.

While it is true that tree quality is not a guarantee of success, using only high quality tree stock gives you your best possible chance.

Specifications and standards NATSPEC

In 1996 the first NATSPEC specification for trees was published in a book called *Purchasing Landscape Trees* (Clark, 1996). This was immediately endorsed by the National Arborists Association of Australia, the people responsible for cleaning up the mess created when sub-standard trees are used and by the Olympic Coordination Authority, for trees supplied for the 2000 Olympic Games. However, growers found it variously; enlightening, confronting, confusing, and/or offensive.

Having benefitted from 7 years of use and significant contributions from senior arborists and landscape architects, the second version of NATSPEC was published in 2003 in *Specifying Trees* (Clark, 2003). The information was very much the same but the format had been simplified and de-mystified. This second version was endorsed by landscape architecture as well as the arborists.

The reaction from growers this time round was far more positive, with large numbers of growers now supporting it. However, there were still some objections to NATSPEC, falling into the following two broad categories:

- Misunderstandings
- A resistance to change

Misunderstandings

The bulk of issues arising from a misunderstanding are linked to the misuse of some indicative tables.

The second version of NATSPEC includes tables that show indicative height/calliper/container volumes that comply. (These were added at the request of our landscape architects.) While these tables are very useful, providing sensible descriptions of trees for use in ordering and tendering, these suggested combinations do not form part of the specification. Unfortunately, these indicative height/calliper/container volumes have, on occasion, been seen as requirements and potentially conforming trees rejected as a result.

A resistance to change

Many of the production practices we use in our industry, while firmly entrenched, are designed to benefit growers rather than trees. For example; growing trees at close spacings allows more trees to be grown in any given area (good for the grower). However, close spacing forces vertical growth, loss of lower foliage and often leads to trees being unable to support themselves and unable to add the necessary calliper and stem taper to ever be self

supporting (bad for the trees).

Objections to NATSPEC have resulted from grower's inability to produce complying trees, using their current production practices. The belief being that the specification must therefore be too tough.

NATSPEC remains our most stringent specification for trees and it is endorsed by the bodies representing our most knowledgeable end users – Arborists and Landscape Architects.

AS 2303:2015 — Tree stock for landscape use (The Standard)

The Standard has just been released (2015). The Nursery and Garden Industry (NGIA) initiated its development and it is based on NATSPEC.

Creating a document through committees and based on consensus is always difficult. After quite an involved process, the resulting document still needs work but is a very creditable first draft.

Areas that need to be upgraded include:

- Replacing the "less than or equal to" signs on figures relating to the figure relating to stem taper.
- Upgrading the criteria for stem structure relating to the relationship between the size of stems and branches.
- Upgrading the section on tree stock balance assessment from "should" to "shall".
- Revising and simplifying the inspection process.

While the current form of The Standard remains flawed it is a credit to both the NGIA and the Local Government Tree Resources Association. While "not quite there yet" The Standard is within striking distance of being a workable and effective document.

Conformance and litigation

With NATSPEC and The Standard now "out there", growers who choose to continue producing trees to lesser standards leave themselves open to the possibility of litigation.

Should trees fail in the landscape it is generally not difficult to conduct a post mortem to discover which fault, in the development of the tree, caused the problem. This fault can, in turn, be traced back through the stages of production to the grower responsible.

As a grower your best protection, against the possibility of legal action, is to grow conforming trees.

Quality problems — why do we have them?

If you head to your local nursery and take a serious look at the standard of trees being sold, you will probably find some that are poor, some that are OK and some that are good.

Why aren't all trees grown and sold in Australia grown to high standards? The answer is twofold; a resistance to change and commercial pressures.

Many of our tree growing practices have evolved to make tree growing easier and more profitable. For example: Growing trees close together means we can fit more into our nurseries. This is good, financially, for the grower but typically results in trees with poor stem taper and calliper.

These practices are well entrenched and, perhaps even more importantly, the market place has evolved with these less than ideal trees and come to expect and accept them. While there is a market for such trees it is difficult for growers to afford the extra time, effort and space needed to grow genuinely great trees.

Sadly, not all tree sales are made to knowledgeable buyers. Sales to the general public are governed by size, presentation and cost, rather than actual quality. The decision about which trees to but for major projects is often made by financial controllers or project managers. Understandably, these are not "tree people" and price will be the major consideration. It is only sales where the buyer is knowledgeable about tree quality issues (generally arborists or Landscape Architects) where quality will be the primary consideration. Sadly, these knowledgeable end users represent only a small part of the total market for trees.

The quality of trees grown by our industry remains variable because it is human nature to resist change and commercially difficult to implement it.

The adoption of rigorous specifications and standards raises the standard of demand. As a bigger and bigger proportion of the market place comes to demand a better product, the industry can then in turn justify producing a better product, safe in the knowledge that they can now do so and still be competitive.

Quality as it specifically relates to propagators

- Good trees in the landscape come from good trees grown by our growers of more advanced stock.
- Good advanced trees result from growing-on good smaller stock.
- Good smaller stock is the result of potting-on and growing-on good propagation material.
- Therefore, good trees in the landscape come from good trees from our propagators.

The majority of problems with propagation material results from root deformities. NATSPEC sets out clear criteria for acceptable root direction and development in all sizes, including tubes and small propagating containers. The Standard actually takes this a step further and officially outlaws J-roots, circling roots, kinked roots and girdling roots.

As with tree quality in general, the publication and adoption of acceptable criteria for quality, especially of root systems, for tubestock etc. means that poor stock will no longer be acceptable and growers will be required to put extra rigor into their production practices to ensure trees conform. The market place must, in turn, be prepared to pay for this extra effort.

Propagating trees with great root systems involves:

- Understanding what is required (see Moore, 2016; AS 2303:2015, 2015)
- Devising propagation practices that address these requirements
- Building-in an ongoing inspection/QA programme to ensure that these practices are working.

CONCLUSION

Trees are incredibly valuable components of our urban landscapes adding tens of thousands, or even hundreds of thousands, worth of calculable benefits to communities and individuals. These benefits are only realised if the trees succeed in our landscapes and this success is the result of key components of tree planting coming together — tree quality being one of them.

Given that the cost of the tree represents a small part of the total planting costs, cutting costs and quality of trees does not make practical, professional or financial sense.

With the increased temperatures associated with climate change, trees become even more important to us and, due to the increased storm activity predicted, their structural integrity more critical.

As our understanding for the need for quality increases and we have specifications and standards in place, we, as growers, have an increased responsibility to produce well grown, well structured trees.

The voice of knowledgeable tree users is getting louder and we, as an industry, must listen to that voice. With the NGIA now joining our arborists and landscape architects in the push for better trees, growers who don't take quality seriously will be left behind.

NATSPEC is widely used and respected, if at times misunderstood and The Standard has the potential to be a useful successor. Combined they help to define new benchmarks that the nursery industry can aspire to, competitively.

The quality of trees from our propagators is the beginning of the quality control process and quality, at this small starting point in tree production, is a key component of the success of all trees in our industry and in the landscape beyond.

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NATSPEC. www.natspec.com.au/.

Breeding for sterility in invasive ornamental plants©

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INTRODUCTION

Invasive plants are introduced species that can thrive in areas beyond their natural range of dispersal (USDA-NISIC, 2014). They naturalize over large areas, displace native plants, and disrupt natural ecosystems (Ranney, 2004). In Florida, over 1.5 million acres (approximately 600,000 ha) of public conservation lands have been invaded by introduced plant species (Figure 1), and approximately USA$7 million was spent on management and control of invasive upland plants in 2011. In the USA, control costs and production losses due to weeds was estimated at US $30.6 billion per year (Cusack et al., 2009). For example, purple loosestrife (*Lythrum salicaria*), was introduced from Europe to USA in the early 1800s. Purple loosestrife is now found in all continental states except Florida (Blossey, 2002) and accounts for USA$50 million per year in control costs and forage losses. Mexican petunia, *Ruellia simplex* (previously also known as *R. brittoniana*, *R. coerulea*, *R. malacosperma*, and *R. tweediana*), was introduced to Florida from Mexico sometime before 1940 (Hupp et al., 2009) and has now naturalized throughout the state, plus six other southern USA states, Puerto Rico, the USA Virgin Islands and Hawaii (USDA-NRCS, 2014). It is considered as a Category I invasive species in Florida because it is altering native plant communities by displacing native species and changing community structures or ecological functions (FLEPPC, 2013). However, there is no evidence that it is hybridizing with native species (Freyre and Tripp, 2014). Sales of *R. simplex* 'Purple Showers' in Florida were ranked third for herbaceous perennials after pentas and lantana (Rick Brown, Riverview Flower Farms, pers. commun.), so a breeding program aiming to develop sterile, non-invasive cultivars was established at the University of Florida in 2007 (Freyre et al., 2012a). This species will be described in more detail in this paper.

Figure 1. Lake Jesup area invaded by *Ruellia simplex*.

CHARACTERISTICS OF INVASIVE PLANT SPECIES

The most successful non-native species, those capable of displacing natives, share several characteristics: (1) Effective reproductive and dispersal mechanisms; (2) Competitive ability superior than that of the native; (3) Few to no herbivores or pathogens;

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(4) Ability to occupy a "vacant niche"; (5) Capability of altering the site by either significantly changing resource availability or disturbance regimes or both (Gordon, 1998). *Ruellia simplex* shows many of these characteristics. Plants flower within 3 months (Wilson and Mecca, 2003), and can produce fruits from either open or self-pollination. Under low light levels, plants can produce cleistogamous flowers, which have greenish-brown, very small corollas that do not open, and form fruits from self-pollination (Khoshoo et al., 1969). Capsules contain on average 20.6 seeds per capsule. Seeds do not have a dormancy period, and have 98% to 100% germination rate under ideal conditions of 30°C day and 20°C night. Moreover, seeds are capable of germination under a wide range of temperatures and under conditions of both light and dark (Wilson et al., 2004). Explosive dehiscence of the seed capsule results in seed dispersal distances from the parent plant of 2.5 to 3 m (Witztum and Schulgasser, 1995). Seeds become mucilagenous and adhesive when wet, aiding their dispersal by animals (Ezcurra and Daniel, 2007). Seeds can even germinate under water (personal observation).

Ruellia simplex plants have the ability to grow in a wide range of environmental conditions, from wetlands to almost xeric. In Florida, the species has been reported in five different plant community types: pine flatwoods, prairies; hardwood (hammocks, tree islands, etc.); freshwater marshes; rivers, springs; and salt marsh (Hupp et al., 2009). In the meantime, native *R. caroliniensis* is found primarily in dry native woodlands (Gilman and Landrum, 1999). A study comparing growth and development of *R. caroliniensis* and *R. simplex* established that under wet conditions in laboratory experiments, *R. simplex* exhibited several traits that favor efficient use of resources and high growth rates. It was therefore concluded that under typical wetland conditions *R. simplex* might be expected to out-grow and out-compete native *R. caroliniensis*, especially if the supply of nutrients is limited (Wilson et al., 2004). In several areas where *R. simplex* has naturalized, its coverage was found to constitute 50% of the infested stratum, thus changing community structure by adding a new stratum, or increasing plant density in the stratum by 5-fold. It was also probably altering the hydrology within plant communities (Hupp et al., 2009).

BREEDING METHODS TO OBTAIN STERILITY IN ORNAMENTAL PLANTS

For several years, ornamental plant breeders have been using a number of methods to develop sterile (or nearly sterile) plants that will not be invasive by seed dispersal including the following.

Selecting and breeding for double flowers

Many plant species have forms exhibiting double flowers, which have more than the normal number of petals in the corolla. The reproductive organs (stamens and carpels) are modified into additional petals, thus conferring sterility or near sterility. Many garden plants have been selected for having double flowers, for example roses, carnations, camellias, and double columbines, petunias, and impatiens. Recently, a molecular model that accounts for the formation of double flowers was described (Lohmann et al., 2001; Lenhard et al., 2001).

Induced mutagenesis

Induced mutations have successfully assisted in developing improved and new cultivars among both seed- and vegetatively-propagated crops (Jain, 2006). Mutations resulting from treatment with X-ray or gamma irradiation or chemicals such as ethylmethanosulfonate (EMS) can result in sterility. However, mutations are random, resulting in the need to screen large numbers of individuals. Irradiation treatments have been successful in inducing male and/or female sterility in several ornamental crops that are clonally propagated for commercial production, including *Chrysanthemum*, *Cineraria*, and *Verbena* (Broertjes and Dejong, 1984; Huang and Hong, 1995; Saito et al., 2005).

Wide hybridization

This involves interspecific or intergeneric crosses between distantly related individuals. Chromosome dissimilarities between the parental genomes can result in meiotic

failure during gamete formation, leading to sterility. Some examples include interspecific crosses between *R. caroliniensis* × *R. simplex* (Freyre and Tripp, 2014), and ×*Chitalpa*, an intergeneric cross between *Chilopsis linearis* × *Catalpa bignonioides* (see also ×*Chitalpa tashkentensis*) (Rusanov, 1964). In some cases, breeders may need to use ovule or embryo culture in vitro to obtain hybrid plantlets that would not otherwise survive (Bridgen, 1994).

Polyploidization and development of triploids

Ploidy manipulation is an important tool in plant breeding, exemplified by the development of seedless triploid sugar beet and water melon (Stebbins, 1956). The development of triploid plants (with three sets of chromosomes) involves first the induction of tetraploids (with 4 sets of chromosomes) from original diploid plants (with two sets of chromosomes) by use of the chemicals colchicine or oryzalin, followed by cross pollination between tetraploids and diploids. Triploids typically grow and function normally, but they have an inherent reproductive barrier in that the three sets of chromosomes cannot be divided equally during meiosis (Ranney, 2004). In ornamental plants, triploids have been bred in rose-of-sharon and spurflower (Brits and Li, 2008) and this approach has also been utilized to breed triploid sterile selections of invasive tutsan (Olsen et al., 2006) and lantana (Czarnecki and Deng, 2008).

BREEDING STERILE MEXICAN PETUNIA

Polyploidization experiments were performed at the University of Florida in Gainesville in 2008 using oryzalin on the apical meristem of seedlings of *R. simplex*. Ploidy levels were determined on mature plants using flow cytometry as described by Czarnecki and Deng (2009). Treatments of three applications of 25 or 50 µM oryzalin every 12 h were most successful in inducing polyploidy. Hybridizations were performed with plants of different ploidy levels, such as 4*x* × 2*x* and 2*x* × 4*x*, aiming to obtain sterile triploid plants. A total of 495 *Ruellia* plants were obtained in 2010 and initially evaluated in the greenhouse for growth habit, flowering, and lack of fruit formation. Fifteen *Ruellia* hybrids and five controls were selected for field trials and propagated vegetatively.

In 2011, plants were trialed in three simultaneous field experiments conducted at the North Florida Research and Education Center in Quincy, Florida, at the Plant Science Research and Education Unit in Citra, Florida; and the Indian River Research and Education Center in Ft. Pierce, Florida (northwestern, north central, and southeastern Florida, respectively). The experimental design was a randomized complete block with three blocks. Each plot consisted of three plants for each cultivar or breeding line, spaced 50-cm apart. Wild *R. simplex* (2*x*) and 'Purple Showers' (4*x*) were included as purple- flowered comparison lines, 'Chi Chi' (2*x*) as pink-flowered and 'Snow White' (4*x*) as white-flowered controls. Each plant was evaluated every 4 weeks, from May to October (24 weeks), for landscape performance, flowering and fruiting (Freyre at al., 2012a).

Three 4*x* plants with different flower colors were outstanding and better than their respective controls at all locations. The three selected breeding lines: purple-flowered R10-102, semi-dwarf pink R10-105, and white R10-108 were evaluated for female fertility by harvesting and germinating open pollinated fruits from the field, and by germinating seeds obtained from manual cross pollinations and self-pollinations in a greenhouse. Additionally, male fertility for each plant was determined by staining pollen grains with lactophenol cotton blue. It was estimated that R10-105 had 5% viable seeds per plant as compared to the invasive wild *R. simplex* and 6% as compared to female and male fertility than the existing commercial pink cultivar 'Chi Chi', and it was not approved for cultivar release by the UF/IFAS Invasive Plants Working Group. However, it was demonstrated that R10-102 and R10-108 are both female and male sterile. These lines were released as new cultivars 'Mayan Purple' and 'Mayan White', respectively (Freyre at al., 2012b), and were commercialized in 2013 (Figure 2).

Figure 2. ‘Mayan Pink’, ‘Mayan White’, and ‘Mayan Purple’.

Fruits were collected at the three field locations in 2011 from open pollination of pink-flowered R10-105. Seed was germinated obtaining 148 progeny, which were then trialed in the field in Citra in 2012. A total of 29 pink-flowered open pollinated progeny from R10-105 were selected for further trials based on performance and apparent low or no fruiting. These plants were propagated vegetatively and grown in a greenhouse in Gainesville. Nineteen plants were selected for 2013 field trials in Citra and in Fort Pierce, and for potted plant trials in Gainesville.

The plant R10-105-Q54 was selected as the best performing pink-flowered plant that also had low fruit count. In Citra it was observed that R10-105-Q54 produced some fruits from open pollination but they all seemed to abort prior to maturation. To confirm female fertility, 10 self-pollinations were performed in a greenhouse as well as 20 cross pollinations using either wild *R. simplex* or ‘Chi Chi’ as males. A few fruits were produced but they all aborted before maturation, with the exception of one fruit which matured and dehisced naturally. This fruit contained 14 seeds but they did not germinate. Additionally, it was determined that R10-105-Q54 had only 10% pollen staining compared to wild *R. simplex* with 69%. Since it was demonstrated that R10-105-Q54 had extremely low to null fertility, it was approved for release as a new cultivar by the UF/IFAS Cultivar Release Committee and the UF/IFAS Invasive Plants Working Group. This line will be commercialized under the name ‘Mayan Pink’ (Freyre and Wilson, 2014).

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Semi-selective herbicide use in nursery weed control©

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BACKGROUND

Herbicides fall into three practical categories (groups):

A. Pre-emergent.
B. Non-selective (knockdowns), semi-selective refers to the use of non-selective knockdowns at ultra-low concentrations to control weeds and to avoid off target damage in bushland and nursery situations.
C. Selective: selective relates to target species or types of weed control within cereal crops, grass selective, etc.

A considerable body of science in the use of semi-selective herbicide use has been developed by scientists and practitioners in Western Australia to combat particular environmental weeds in quality bushland. The intention has been to find effective weed controls using herbicides without off target damage. This work over many years has lead to the development of very successful techniques which may have application to nursery weed control.

This presentation is our introduction of the use of non-selective knockdown herbicides at ultra-low concentrations for nursery weed control.

DISCUSSION

Products

1. Western Australia.

The following are some of the knockdown herbicides that are currently being used in semi-selective mode with Western Australia (WA) bushland; these are permitted for off label uses in WA:

- Metsulphuron (Brush Off®)
- Triasulphuron (Logran®)
- Clopyralid (Lontrel®)
- Halosulphuron (Sempra®)
- Haloxyfop (Verdict™)

2. New Zealand.

I could find only one reference to the use of a herbicide in semi-selective mode — Metsulphuron for use in Onehunga control on golf courses (Massey/University of NZ) .

New Zealand herbicide brand name match:

- Metsulphuron: Associate®, Agrpro®, Muturon®, etc.
- Triasulphuron: Titan, Genfarm
- Clopyralid: Versatill™
- Halosulphuron: Enviromax, Nufarm
- Haloxyfop: Hurricane, Ignite

Objective of trials

- Determine if control could be achieved without off target damage.
- Which chemical would provide best overall results and which was best for particular weeds.
- If mortality was not achieved, was it possible to prevent weed seed set.

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MATERIALS AND METHODS

Preparation and application

The following is a guide for nursery application:

- Accurate measurements by weight critical.
- Use clean filtered water.
- Granular herbicides — use warm water to aid dissolution.
- Waiting period for watering will apply.
- Avoid spraying on warm days.
- Mix in 20-L volume and dispense to smaller units.
- Apply to strong plants.
- Apply once, avoid double spray.
- Target weeds as best possible.

Trial outline

- Various application rates and mixtures were trialled on individual plants, including combinations of two herbicides given their compatibility.
- Nine species of Perth natives chosen for weed treatment.
- Settled on the following:
  - o Triasulphuron at rate 1. 2 g 20 L^{-1}
  - o Metsulphuron at rate 0. 6 g 20 L^{-1}
  - o Combo of triasulphuron and metsulphuron (50/50)

Weeds targeted

Scientific name	**Common name**
Cardamine hirsuta	Flick weed
Chamaesyce spp.	Asthma weed, cats hair
Gnaphalium spp.	Cudweed
Oxalis spp.	Wood sorrels
Sagina procumbens	Pearlwort
Marchantii polymorpha	Liverwort
Bryophyte	Mosses

RESULTS

Logran results

- Effects in place within 1 to 2 days for cudweed and flick weed.
- Cud weed species were heavily affected; within a week most wilted off.
- Stunted and discolouration of *Oxalis* spp.; weeds left in an inferior state, roots and stems still in place with leaves wilted off.
- Liverworts and sponge-like moss displayed changes by the 2nd week and treatment appeared to be effective.
- No abnormal changes in grass-like moss (parlwort).
- Successfully achieved aims; no off target impact.

After 1 month.

Weeds	**Impact**
Flick weed	Decayed/rotted off/eradicated
Asthma weed	Stunted growth, yellowing of leaves
Cudweed	1 to 2 days; strong signs of wilt, decayed
Wood sorrels	Stunted growth, yellowing of leaves
Pearlwort	No effect, seed set of pearlwort not effected
Liverwort	Eradicated
Moss	Stunted growth

Metsulfuron results

- Took 2 to 3 weeks for changes to be observed.
- Successful on flick weed and cud weed species; most wilted, off completely by the end of the month.
- Similar to the effects of Logran on *Oxalis* spp.; roots and stems still in place.
- Successfully achieved aims.

Weed results after 1 month:

Weeds	**Impact**
Flick weed	Stunted growth, strong signs of wilt
Asthma weed	Stunted growth, signs of rot
Cud weed	Eradicated
Wood sorrels	Stunted growth, yellowing of leaves

Logran and metsulfuron mix results

- Effects take up to 3 to 4 weeks; slow to act compared to other trials.
- Cud weed did not wilt off completely within a month compared to other trials.
- Good against flick weed species; by the end of the month most had wilted off completely.
- Effective against *Oxalis* spp.; able to produce adverse effects on infestations.
- Possibility that Logran and Metsulfuron are working against each other.
- Aims achieved but not best option.

Weed results after 1 month:

Weeds	**Impact**
Flick weed	Stunted growth, strong signs of wilt
Asthma weed	Stunted growth, yellowing of leaves
Cud weed	Stunted growth
Wood sorrels	Stunted growth, yellowing of leaves
Pearlwort	No effect
Moss	Stunted growth

CONCLUSION

Summary of results

- Earlier stages of trials are positive.
- Trials show that logran and metsulfuron act better on certain weeds.
- Same mode of action, different active constituents; affect different weed species at different rates.
- Ongoing trials: Liverwort regrowth, time it takes for new weed growth after application.
- More trials to be done with different Group B herbicide products.
- Repeat current trials for conclusive evidence.

Potential with caution

- Encouraging results.
- Impacts on succulents/herbs may be adverse.
- May be more relevant to natives and strong ornamentals.
- Suggest small scale trials with very low concentrations, then upscale to achieve weed morbidity and assess off-target impact.

Towards new nursery industry protocols for *Phytophthora* control on supply of stock for restoration and revegetation©

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Abstract

The threat to Australian plant life and biodiversity from existing and potential additional forms of *Phytophthora* is real and well documented. Some 50% of Western Australia s endangered flora is susceptible to *Phytophthora* dieback. Whilst there is a range of potential methods of *Phytophthora* pathogen transfer to valuable conservation areas, a very obvious and likely source is transmission via nursery sourced plant stock. The current nursery accreditation standards and compliance, though better than none, are no longer considered adequate to address the current and prospective threat to Australian flora posed by *Phytophthora* from nursery stock. The need for nurseries and buyers of plant stock to know and understand their responsibilities to the environment, the nursery industry and each other, requires broad engagement and consultation with the shared intention of moving forward to a higher level of pathogen management and in particular for those supplying stock to valuable conservation areas. This paper will outline the writer views on achieving improvement to nursery *Phytophthora* protocols for supply to conservation areas.

BACKGROUND

The writer has been involved in the production of plant stock for restoration and revegetation for the last 12 years and founded a Perth native plant nursery and associated environmental contracting and consulting business. His concerns for plant quality and hygiene standards within the industry, led to a presentation on the subject at the Australasian Plant Conservation (APC) Conference in Perth in 2010. The writer has not become aware of any improvement in hygiene standards within the industry or in any particular nurseries supplying stock for restoration /revegetation in the 4 years since.

Plant science has continued to isolate additional species of *Phytophthora*, some of which have the potential to cause significant additional damage to both native flora as well as commonly used exotic flora.

The recently released National *Phytophthora* Threat Abatement Plan contains numerous mentions of the risk of pathogen transmission from nursery stock.

DISCUSSION

The light bulb moment for the Natural Area Nursery, operated by our family company, arose in early 2014 with three significant clients seeking nursery soil samples for *Phytophthora* testing.

Supply of plant stock for restoration — current industry deficiencies

- Stock often sourced from non accredited nurseries.
- Accreditation system compliance inconsistent across nurseries.
- No agreed methodology for soil testing.
- No agreed or documented system for recovery from positive *Phytophthora* testing.
- The need for a higher standard for supply to conservation areas is yet to gain acceptance.

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Risk sources for nursery infection

- Drainage runoff entry to nursery.
- Nursery ground soil or water supply.
- Contaminated soil from vehicles, boots, tools, and equipment.
- Inadequate sterilisation of recycled plant containers.
- Plant stock from other nurseries or harvested from external sites.
- Soil suppliers.

Risk management

Some of the main issues that arise from requests for *Phytophthora* testing by clients are seen as follows:

1) To accept or refuse.
   i) To accept leaves the nursery very exposed should a positive result be returned. How would a nursery deal with such result? What could the impact be on the business? It can be expected to be anything from significant to devastating. Does the nursery have a plan for recovery after a positive result? Was a control sample retained by the nursery to enable verification of the result?
   ii) To refuse may raise suspicion and a test may still be undertaken by the client when stock is received. If positive, it is likely that payment for the stock would be in dispute. A refusal is likely to lead to the loss of future orders.
   iii) The reality is that testing is likely to become more prevalent in the future and the industry needs to accept that these risks need to be managed and systems established. Dealing with the issue in a proactive way is preferable to retrospective fire fighting. A positive test for *Phytophthora* is likely to have a debilitating effect on any nursery supplying the restoration/revegetation market.
   iv) It has become obvious that the current accreditation standard does not satisfy prominent clients involved in restoration of the conservation estate.
2) To take control.
   i) It is preferable that testing by clients not take place at all as nurseries will not have sufficient control of the process, the outcome, and the impact. The issue is to find a way to satisfy clients relative to hygiene standards and discourage testing.
   ii) The only prospect of achieving this is by undertaking a comprehensive review of hygiene protocols, up-scaling them to a standard above accreditation and implementing testing systems which are controlled by the nursery. The up scaled protocols can be detailed to customers, visitation encouraged and the nurseries policy re testing outlined and acceptance gained.
3) Potential for damage to the industry.
   i) Positive tests of nursery stock could force a curtailment of buying and moves to increase the use of direct seeding for revegetation. There is also the risk of infection transfer between nurseries. All nurseries involved in the supply of revegetation stock could see a substantial impact on their business.
   ii) Furthermore, growing incidences of *Phytophthora* detection in revegetation nurseries may result in increased scrutiny and testing of nurseries focussed on supply of landscape stock.

Recommended new nursery protocols for supply to conservation estate areas and for restoration and native species revegetation

- All plant stock to be off the ground at sufficient height to avoid root contact with the ground surface and water splash from ground surface.
- All soil batches to be tested to standards established by Murdoch University, Centre for *Phytophthora* Research. Control sample to be retained by nursery.
- All production to be tracked and matched to individual soil batches to enable recovery action in event of positive test result. Soil batches not to be mixed.

- No exposed ground areas within nursery, i.e. either hard stand or 100-mm aggregate cover.
- No growing medium to be recycled.
- Stock bought in to be from accredited suppliers.
- Recycling of containers to be a dual stage process, any two of chlorine solution inundation, steam sterilisation, commercial grade dishwasher hot wash min 80°C. (Solarisation, pressure cleaning and hand washing are not acceptable).
- Existing accreditation requirements to apply; i.e.
    - All soil and potting mixes to be sourced from accredited suppliers.
    - Soil mix to be housed within a clean and contained storage facility with no potential ground water inflow.
    - Chlorination of all nursery water.
    - Clean down stations and foot baths at nursery entry points.
    - Restricted vehicle entry and designated plant despatch area as a distinct hygiene area.
    - Nursery tools and equipment to be exclusively for nursery use.
    - A distinct quarantine area to be maintained for stock from outside sources

Issues arising from suggested protocol up scale

1. Cost.

The significant cost associated with nursery benches is acknowledged. In some cases it may suit to utilise recycled plastic pallets to maintain stock ground clearance.

A quality Stage 2 *Phytophthora* test will likely cost approx $300 per test. In the Natural Area Nursery situation, the added cost per plant based upon an average year is 0.5¢ per tube. A Stage 1 test results would not normally be available for 2 weeks, hence the need for soil batches not to be mixed and all production tracked relative to each batch. In event of a positive test, this would allow subject stock to be isolated/dealt with.

2. Effectiveness with clients.

Proposed changes may not be acceptable to some clients and business decisions will be made within nursery management in dealing with each. However, it is the firm view at Natural Area that once in place, we do not intend to compromise the new arrangement by acceding to testing in the hands of others.

Once stock has been accepted as in good condition and has left the nursery, the client may then carry out testing but this would not affect the client obligation to pay for the stock. Should testing prove positive in this case, we would argue that we cannot be held responsible for stock out of our control.

Marketing

It is intended that Natural Area actively market the up scaled protocols as a positive initiative to the benefit of clients, the environment and the nursery industry in general.

Industry acceptance

There is no doubt that some, maybe many in the nursery industry will not accept that these proposed changes are necessary or in their interest to implement. However, having seen the impact elsewhere on Phytophthora introduction to a production nursery, the writer has no doubt that the introduction of higher standards is very much a sound risk management exercise.

It is expected that the restoration/revegetation industry sector will establish design, operational and supply standards to projects and these are expected to include demanding standards on suppliers of seed and plant stock. Reference to recent Society for Ecological Restoration Australasia (SERA) and Revegetation Industry Association of Western Australia (RIAWA) conference proceedings will confirm.

Implementation

We are currently road testing the concept with clients in the lead up to 2015 supply and responses are awaited. The Directors of Natural Area Holdings/Natural Area Nursery expect to fully implement the up scaled protocols by end of second quarter 2015.

CONCLUSION

The writer would appreciate constructive criticism and comment on the proposal from those involved in academic, nursery, and revegetation activity in this space.

In vitro Grevilleas©

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INTRODUCTION

The genus is named after Charles Francis Greville and is predominantly Australian with some species from neighbouring countries. Grevilleas are a member of the family *Proteaceae*. There are three groups; the Banksia Group, the Rosmarinifolia Group, and the Toothbrush Group. From these groups there are also a number of interspecific hybrids.

Importance

The plants are a range of prostrate shrubs to trees mostly woody natives. Flowering ornamentals with attractive flowers and foliage and those with nectar attract pollinators and native birds. They make excellent native garden plants from ground covers to shade trees. They have a range of flowers available to suit everyone's interest.

There are many species and hybrids with different plant forms and a range of flowers available to suit everyone's interest. There also a few varieties that are commercial timber species.

Adaptability

Grevilleas will grow in most environments and love sunshine and well drained light soil low in phosphates.

PROPAGATION

Seed propagation is ok for straight species. Generally it is fairly easy and young plants could be selling around $2.00 each. Genetic variation not of any concern.

Vegetative propagation is a must for hybrids due to sterility and need to maintain consistency of appearance. Need to be aware of seasonal issues. Can be done in three ways: cutting, grafting, and micropropagation.

Semi-hardwood cuttings

Difficult due to low strike rates in many cases as low as 10%. Will translate to $3.00-$10.00 per young plant.

Grafting

Very difficult to do which in many cases results in $15 or more per plant. Silky oak (*Grevillea robusta*) is considered to be the best root stock.

Micropropagation or plant tissue culture

This is a technique of growing isolated organs/tissues and cells of plants in a defined nutrient medium under controlled conditions of light, temperature, and humidity.

Advantages of tissue culture:

- Rapid cloning (clones are identical plants)/uniformity
- Production of large numbers in a small space and time
- Freedom from seasonality of production
- Production of clean/disease free plants
- Less expensive in many cases, compared to grafted plants
- Induce juvenility
- Accelerate maturity and early flowering

Why tissue culture grevilleas? As detailed above normal vegetative propagation is difficult and results in expensive young plants. Tissue culture gives the following benefits:

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- Fast and reliable multiplication/cloning
- Avoid segregation of hybrids
- Generate clean/disease free plants
- Induce juvenility for accelerated cutting production
- Uniformity of the plantlets
- Early flowering
- Reduces cost of production

Major steps in tissue culture production of grevilleas. There are basically four major steps in micropropagation of grevilleas: initiation, multiplication, rooting, and acclimatization/hardening.

1. Initiation.

First step is to initiate the plants. This involves getting micro cuttings clean, sterile, and stable on the agar. The agar or growing medium is very important. *Grevillea* initiation can take place in different media like MS Medium (Offord and Tyler, 1998), WPM medium (Bunn and Dixon, 1992), half strength MS medium with $1/10^{th}$ KH_2PO_4 supplemented with low levels of cytokinin alone (BAP 2.0-5.0 µM) or a combination of NAA or IBA and BAP at ratios 1:5 to 1:10 in the range of BAP (5.0-10.0 µM); 2iP was also useful. A 16-h photoperiod at 50-100 µmol m^{-2} s^{-1} light is adequate.

It can take 1-3 months before initiation takes place. The most significant problem at initiation step is contamination. The pubescent nature and/or the waxy stem harbours a lot of contaminants. These can be a major issue in the clean sterile growing environment. Combination disinfection treatments with ethanol followed by bleach worked better than a single treatment. However, tissue death is an issue with some of the species and hybrids during the decontamination of explants. There is a large variety of contaminants that need to be removed or at least controlled.

2. Multiplication.

Once the plants are stable and growing on the agar they can be put into multiplication mode whereby the number of units rapidly increases.

Again the make-up of the agar or growing media is a key issue. Grevillea multiplication can take place in different media like WPM + 5 µM Kin + 0.5 µM BAP — shoot multiplication (Bunn and Dixon, 1992), or ½ MS + 10 µM BAP + 0.5 µM IBA (adventitious shoots on leaf explants of *G. scapigera* (Bunn and Dixon, 1992). Also ½ MS and WPM was helpful along with 1-4 µM BAP and 0.01-0.02 NAA in the case of some grevilleas. Seventeen species of grevilleas have been multiplied on MS medium containing 1.0-1.5 µM BAP alone (Offord and Tyler, 1998).

3. Rooting.

Once the plants have multiplied two or three fold some are put into a rooting agar and some back into multiplication. Those on rooting will produce small agar specific roots. In vitro rooting is reasonably easy in ½ MS medium containing 5.0-10.0 µM IBA. In some cases added charcoal (0.5-2.0 g L^{-1} also is helpful. Sometimes the new cuttings bypass this stage in the laboratory. This is referred to as ex-vitro rooting.

Ex vitro rooting with IBA powder at 1 g kg^{-1} (1000 ppm) or 3 g kg^{-1} (3000 ppm) in a fogged glasshouse at 90-95% humidity, gave good results (Bunn and Dixon, 1992; Offord and Tyler, 1998).

4. Acclimatisation.

The hardest and most risky part of the process is the acclimatisation or hardening off where the young plants are weaned off agar and removed from the moist controlled atmosphere of the laboratory and “taught” to grow in a normal medium in regular greenhouses. In vitro rooted grevilleas acclimatised in greenhouse with fogging initially but misting after 2 weeks from deflasking. High porosity of the potting mix is critical for success.

The propagatability index

The question arises on whether the selected taxon of *Grevillea* is better by tissue culture or conventional means. To assist in this there is the propagatability index (PI). This is the product of success rates at each stage an equals: [Initiation (I)] × [multiplication M)] × [rooting (R)] × [acclimatization (A)] or (PI = I×M×R×A) where I = success rate at initiation, M = multiplication rate per month × R = Rate of rooting and A = rate of establishment at hardening stage. For example: cultivar "A": I = 0.50, M = 4.0, R = 0.90, A = 0.80 which gives 0.5×4.0×0.9×0.8 = 1.44; the PI is 1.44. In general, tissue culture of a species in demand with a PI over 0.70 is commercially viable.

CONCLUSION

Grevilleas can be done by micropropagation although not all cultivars have a success rate that is commercially viable.

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Seed propagation of two native Australian species important for land restoration©

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INTRODUCTION

There has been substantial investment in revegetation and restoration of native biodiversity in eastern Australia in recent decades (Close and Davidson, 2003). Incentive programs run through agencies such as Catchment Management Authorities encourage community-based management of natural resources and restoration of native vegetation communities to support biodiversity conservation (Hallett et al., 2014; Local Land Services, 2014). However, more effort is required to achieve restoration at landscape scales. The main limitations to landscape-scale restoration are associated with costs, incompatibility with existing agricultural practices, deficiency of straight financial profits from restoration activities, and inappropriate incentives to change the land management practices (Morrison et al., 2008).

Successful revegetation requires an understanding of species biology and ecological requirements. Large scale revegetation can be achieved using tubestock planting of seedlings, and direct seeding. Direct seeding is more convenient than other methods as it is cost-effective due to less investment of work and material costs and permits use of a diverse seed mix incorporating a range of plant species and growth forms (Dalton, 1994; Gibson-Roy et al., 2007; Hallett et al., 2014). However, direct seeding for restoration requires sufficient ecological knowledge of seed collection, quality, viability, persistence, storage, germination, and other ecological aspects for a wide variety of species.

There are many knowledge gaps to be addressed in order to increase the success of direct seeding revegetation (Baskin and Baskin, 2004; Budelsky and Galatowitsch, 1999; Hossain et al., 2014; Long et al., 2015). These knowledge gaps include effective treatments to break dormancy, seed responses under different seasons and environmental conditions, and suitability of seed to be direct seeded. As a preliminary attempt to fill some of these gaps, we present the results of viability and germination studies of two native plant species with ecological and economic value: *Eremophila debilis* and *Capparis lasiantha*.

Eremophila debilis has a broad geographic range and importance in ecological communities of the arid zones, where it is often dominant or codominant of wide areas. It is drought, fire, frost, salinity, and grazing tolerant and palatable to stock despite its low growth habit. However, its germination is unreliable and that limits its use in direct seeding revegetation programs (Cunningham et al., 2011). Two factors are assumed responsible for unreliable germination in this species: (A) Inappropriate environmental conditions and inability to overcome physical dormancy; (B) A chemical inhibitor of the seed, seed coat, or fruit (Richmond and Chinnock, 1994).

Capparis lasiantha is palatable to livestock and native fauna and has cultural value in aboriginal communities as its fruit is palatable to man (Cunningham et al., 2011). This species appears to be adaptable to abandoned farming fields and is a key component of several important ecological communities in Australia, including some endangered ecological communities (Department of Environment and Heritage, 2006; Fensham and Fairfax, 1997). *Capparis lasiantha* is drought tolerant (Walters, 2015). Studies done on other species from this genus show that the physical constraint imposed by the seed coat may be responsible for seed dormancy and removing it partially will allow germination (Pascual et al., 2004; Sozzi and Chiesa, 1995).

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METHODOLOGY

Viability

Seeds of *E. debilis* were manually extracted from fruit after cracking the fruits using a vice grip. Seed coats of *C. lasiantha,* were removed after cracking the seed coat and peeling it off. Seed viability was tested using the standard tetrazolium staining technique (ISTA, 2003). Three seed lots were tested for each species with three replicates of 30 seeds per seed lot. Seeds were obtained from Field's Native Nursery®, in Uralla, NSW. *Capparis lasiatha* were collected on 2012 (SL1), 2013 (SL2), and 2014 (SL3) test and *E. debilis* seeds were approximately 1 year old. Both were stored in a cool room until test.

Pre-treatments and germination

Germination experiments were carried out using the seed lot with the highest viability (according to the tetrazolium test) for each species. All seeds with evidence of damage were discarded. Three replicates of 50 seeds were used for each treatment and control.

Trials with seeds of *E. debilis* consisted of two treatments: (1) the fruit was manually nicked by cutting the apex of the fruit horizontally with a blade to permit water and oxygen to reach the seed and allow imbibition; and (2) naked seed were obtained from the fruit. This was done by cracking the hard fruit with a vice and posterior manual extraction of the seed from the release locules. The control consisted of untreated seeds within the fruit.

Capparis lasiantha seeds also had two treatments: (1) seed coats were punctured using a dissecting needle, punctures were placed at an edge of the wider section of the seed to avoid damaging the embryo, and (2) seed coats were completely removed from the seed. The control consisted of complete untreated seeds.

Seeds were placed on moistened filter papers over wettex sponges in petri dishes and incubated in growth cabinets, with temperatures set at 25/15°C for *C. lasiatha* and 35/25°C for *E. debilis* at 12 h light/darkness. Temperature regimes were set based on previous results and bibliographic records that suggest the regimes employed here provide the best germination results for these species. The seeds and fruits were irrigated with 10 mL of tap water at the start of the experiment and when required. When naked seed was used, only healthy plump firm seeds were selected for the germination tests. Germination was recorded daily over a 4-week period in the case of treated seed and 8 and 12 weeks for untreated seed of *C. lasiantha* and *E. debilis,* respectively.

RESULTS

Viability

The three different lots of *E. debilis* had apparent similar mean viability results (Figure 1). Up to eight seeds were recovered per fruit within the four locules, however differences among seedlots are statistically significant ($p=0.01$).

Two seed lots of *C. lasiantha* collected in past years had no or very low viability and although the tetrazolium test of the third (fresh) seed lot collected the same year indicated 100% viability of the seed tested, 25% of the seeds had to be discarded due to damage or incomplete seeds, and this lowered the apparent viability. Differences among the seedlots were highly significant $p>0.001$ (Figure 1).

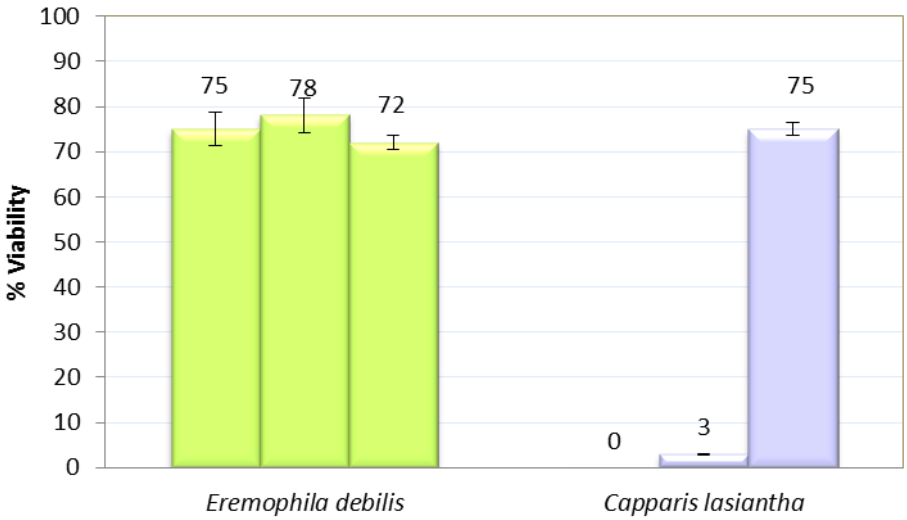

Figure 1. Mean percent viability of three different seed lots (SL) of *Eremophila debilis* and *Capparis lasiantha.*

Germination

The highest germination of *E. debilis* was obtained from naked seed, it was also the fastest as it took an average of 8 days for seeds to germinate compared to the 24 days and up to 80 days for the cutting treatment and control, respectively. The cutting treatment improved germination compared to the control, but had lower germination than the naked seed. In contrast, *C. lashiantha* had low germination from naked seed and highest when seed was punctured. The germination of the control treatment was also noteworthy based on the accepted standard for seed germination of 80% (Association of Official Seed Analysts, 2005). However, these results were obtained by using only healthy-looking seed. Germination percent per treatment was significantly different ($p>0.01$) in both species (Table 1).

Table1. Mean percent germination of two species after two seed pre-treatments and control.

Treatment	Germination (%)	
	Eremophila debilis	*Capparis lasiantha*
Fruit/seed section	42	98
Naked seed	81	65
Control	24	91

DISCUSSION

Plant propagation from seed is encouraged whenever possible because of time and cost savings and to preserve genetic biodiversity (Gibson-Roy and Delpratt, 2014). However, knowledge of seed ecology of native Australian plants can be a limitation for many species. Furthermore, some procedures used to prepare seed and increase germination are time consuming and unrealistic for large scale plantings. Both scientific knowledge of plant physiology and specific technical skills acquired through experience are vital to determine cost effective methods for seed treatment and germination. This implies involvement of scientists, academics, field personnel, nursery managers, and others involved in all aspects of growing and planting native plants.

Previous research with other species of *Eremophila* (Richmond and Ghisalberti, 1994) and *Capparis* (Pascual et al., 2004) suggest that mechanical scarification will give similar results to treatments involving partial cutting of the hard fruit or seed coat like those investigated here. A possible practical approach could be to conduct seed scarification using

mechanic scarifiers that have already been successfully implemented in species of commercial importance (Liu, 2007).

Although dormancy breaking mechanisms for various species are better understood, further research in the direction of practical applications for large scale direct seeding and plant production is required to achieve appropriate outcomes.

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Effects of rare sugars on growth and development in *Phalaenopsis* tissue culture©

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INTRODUCTION

Research on the effects of rare sugars in plant tissue culture is limited (Fukai and Saruta, 2004). In this study, effects of rare sugars on growth and development in *Phalaenopsis* (syn. *Doritaenopsis*) tissue culture were examined.

MATERIALS AND METHODS

Roots of in vitro plantlets were used as the source of explants. These plantlets were derived from seeds of *P.* (syn. *Doritaenopsis*) Little Gem Strips "No1" × *P.* (Yu Pin Fireworks) "3146" hybrids.

Root tips (<0.5 cm) were dissected from plantlets and were cultured on full strength Murashige and Skoog (1962) medium supplemented with 40 g L^{-1} sucrose and 8 g L^{-1} agar. One root tip was cultured in a grass tube (40 mm diameter × 130 mm) containing 30 mL of medium. The pH of medium was adjusted to 5.8, and all media were autoclaved for 15 min at 120°C. Cultures were incubated at 24±2°C under cool-white florescent lamps at an intensity of 50 µmol m^{-2} s^{-1} photosynthetic photon flux (PPF) 16 h day^{-1}.

Experiment 1

Effects of D-tagatose on growth and development in *Phalaenopsis* root tissue culture. D-tagatose (0 or 5 mg L^{-1}) was added to the medium described above. Fifteen tubes were used for each treatment.

Experiment 2

Effects of D-psicose on growth and development in *Phalaenopsis* root tissue culture. D-psicose (0 or 1 mg L^{-1}) was added to the medium described above. Fifteen tubes were used for each treatment.

RESULTS AND DISCUSSIONS

Experiment 1

The root-tip explants cultured on Murashige and Skoog medium supplemented with or without D-tagatose did not show any response. All of them did not survive more than 4 weeks of culture (Table 1).

Table 1. Effects of D-tagatose on growth and development in *Phalaenopsis* tissue culture after 4 weeks of culture.

D-Tagatose (mg L^{-1})	Survival rate (%)	Root formation rate (%)
5	0	-
0	0	-

Experiment 2

Higher percentage survival and morphogenic response of root tips cultured on Murashige and Skoog medium supplemented with D-psicose was observed (Table 2). On Murashige and Skoog medium supplemented with 1 mg L^{-1} D-psicose 40% of root tips

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survived and 26.7% of them developed new root.

Based on these results, D-psicose had little effect on root regeneration in *Phalaenopsis* tissue culture.

Table 2. Effects of D-psicose on growth and development in *Phalaenopsis* tissue culture after 4 weeks of culture.

D-psicose (mg L^{-1})	Survival rate (%)	Root formation rate (%)
1	40.0	26.7
0	26.7	6.7

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In vitro shoots formation by inflorescence apex culture of *Primula ×polyantha*©

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INTRODUCTION

Primula ×polyantha hort. selections are important pot flowering plants in Japan, however, homogeneous seed production is difficult on account of cross-fertilization (allogamous) plant. On the other hand, commercial vegetative propagation is also not possible because of low reproduction rates. Micropropagation is also difficult. In shoot apex culture, contamination occurs frequently because the shoot apexes occur close to the surface level of the soil.

In primulas, adventitious shoots were obtained by the flower-bud culture. We tried flower-bud culture, using *P. veris* L., *P. vulgaris* Hudson, and *P. juliae* Kusnetsow and obtained a few adventitious shoots from only *P. vulgaris* and *P. juliae* (Matsumoto and Ohashi, 2014).

In primulas, the inflorescence is the only elongated stem and those are an indefinite inflorescence; there is an apical meristem in the apex. Actually, a bud is formed on the tip of the inflorescence after flowering, in primulas such as *P. malacoides* franchet, Bull., *P. obconica* Hance, *P. sinensis* Sabine ex Lindley, and *P. modesta* Bisset & Moore.

In the present study, we tried in vitro shoots formation by inflorescence apex culture of *P. ×polyantha*.

MATERIALS, METHODS AND RESULTS

Inflorescence elongation instruction experiment

The selections of *P. ×polyantha* are distributed between the following three types by inflorescence elongation.

1) Polyanthus Type (PT): elongated both inflorescence and pedicel in flowering
2) Acaulis Type (AT): non-elongated inflorescence and elongated pedicel in flowering
3) Sham Acaulis Type (SAT): elongated inflorescence by temperature conditions

First, we examined the inflorescence elongation induction condition because the important cultivars of *P. ×polyantha* are distributed in the AT or SAT elongation types.

The six plug seedling forms of *P. ×polyantha* "Claudia" were purchased from Sakata Seed Corporation in the autumn of 2011 and 2012. These were planted in plastic pots (diameter 12 cm) containing a mix of equal parts of bark compost and pumice for gardening called "kanuma" soil, and cultured on bottom water supply trays in a greenhouse, and treated with gibberellin water solution mist from the flower bud appearing stage. Gibberellin Meiji (Meiji Seika Pharma Co., Ltd., Japan) was used as the gibberellin, and the concentration of water solutions were 50 mg L^{-1} in 2012 and 100 mg L^{-1} in 2013. The treatments were carried out monthly in 2012 and every 2 weeks in 2013, spraying 1, 3, or 5 times per plant (Tables 1 and 2) to the cluster of flower buds.

Table 1. Amount of gibberellin (mg $plant^{-1}$) in mist treatment to *Primula ×polyantha.*

Year	Gibberellin concentration (mg L^{-1})	1 spray	3 spray	5 spray
2012	50	0.031	0.039	0.156
2013	100	0.062	0.187	0.311

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Table 2. Day of carried out gibberellin mist treatment to *Primula* × *polyantha*.

Year	Cycle of spray treatments	Day of carried out treatment				
2012	Every 1 month	January 4	January 19	February 25	May 20	April 20
2013	Every 2 weeks	December 25, 2012	January 7	January 23	February 5	February 19

The length of elongated inflorescences more than 5 mm were measured at the treatments carried out after second treatments.

Figure 1 shows the results of the 2012 gibberellin treatment experiment. Most of elongated inflorescences were not observed on 19 January and 25 February, then, observed all treatments including control on 20 March. It is thought that the elongation of inflorescence was caused by the rises in temperature not an effect of gibberellin and "Claudia" forms are SAT types.

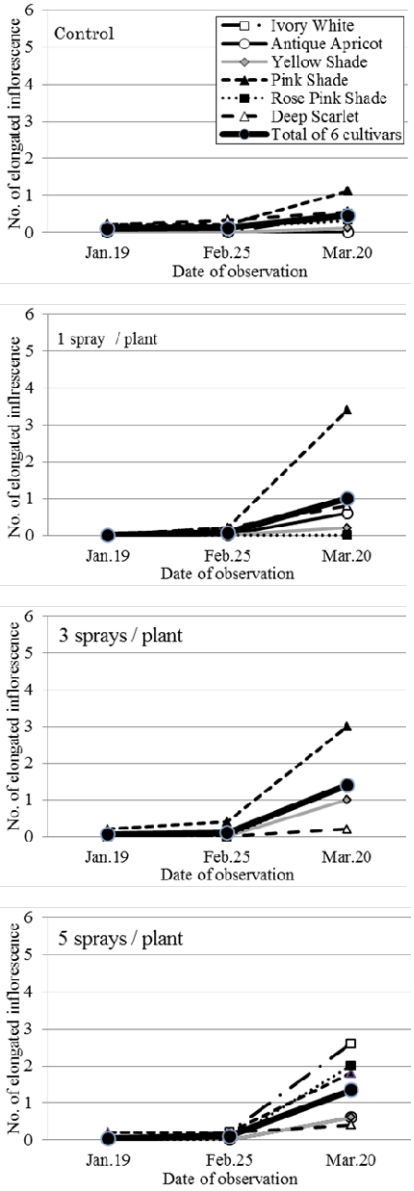

Figure 1. Effect of gibberellin mist treatments on inflorescence elongation in *Primula* ×*polyantha* (2012).

However, Figure 2 shows the results of the 2013 experiment, in which most of the elongated inflorescences were observed on the treated plants and not control plants on 19 February. It is thought that the elongation of inflorescence was caused by an effect of gibberellin, and increased on 8 March; observed elongated inflorescences per plant were 2.75-2.95 on average.

It will be necessary to investigate the following things in the future, optimum concentration point and interval of gibberellin treatments, and plant type difference.

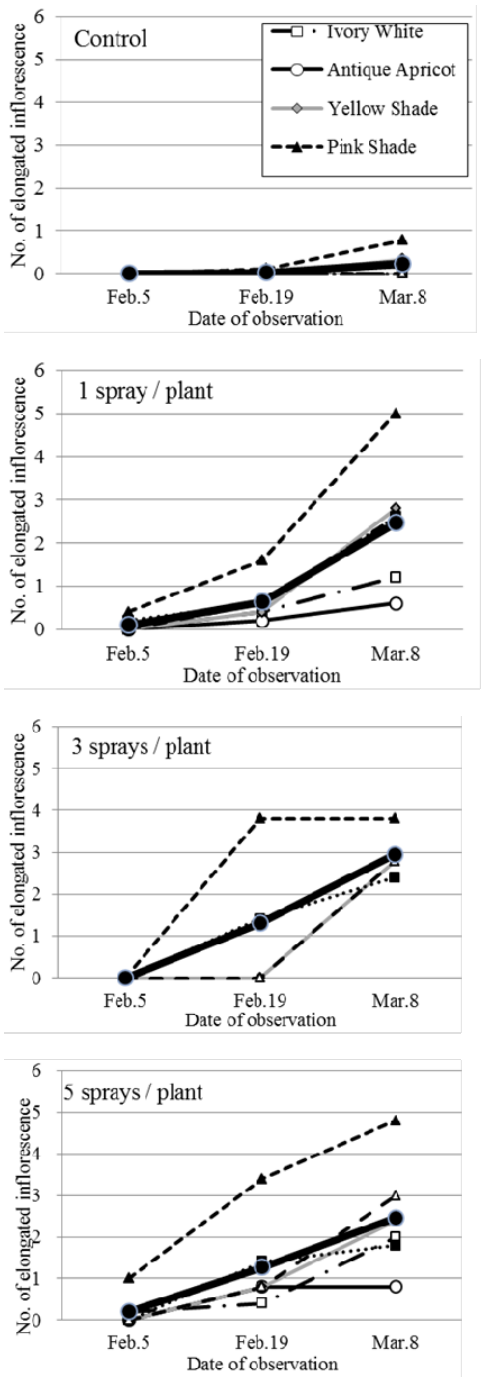

Figure 2. Effect of gibberellin mist treatments on inflorescence elongation in *Primula* ×*polyantha* (2013).

Inflorescence apex culture

Basal medium for inflorescence apex culture was MS medium (Murashige and Skoog, 1962) supplemented with 30 g L^{-1} sucrose and 1, 2, or 4 mg L^{-1} of 6-benzylaminopurine (BA) alone and in combination with 0.1 mg L^{-1} 1-naphthylacetic acid (NAA) as plant growth regulators (Table 3). The pH were adjusted to 5.8±0.1 and 2.5 g L^{-1} gellan gum (Wako pure Chemical Industries, Ltd., Japan) was added before dispensing 10 mL per test tube (25 mm diameters; 120 mm height).

The harvested elongated inflorescences consisting of flowers and flower buds were dipped in sodium hypochlorite solution (1% available chlorine) for about 8 minutes and rinsed with sterilized water. The inflorescence apexes were removed and put and placed one per testtube on each medium.

These were incubated under 20±2°C and16 h per day white fluorescent lamp illumination (about 2,000 Lux) condition, and then observed for 45 and 90 days after inoculation for shoot formation.

After 90 days the rate of contaminated explants was under 10% in spite of the simple method of surface sterilization. By addition of NAA, the rates of explant survival rose, and the rates of shoot formed on explants was 50%, and we obtained 1.8 shoots per inflorescence apex (Table 4).

Table 3. Combination of plant growth regulators for inflorescence apex culture of *Primula* ×*polyantha*.

		BA (mg L^{-1})		
		1	2	4
NAA (mg L^{-1})	0	O	O	O
	0.1	-	O	-

NAA = 1-Naphthylacetic acid, BA = 6-Benzylaminopurine, O = added grow regulator.

Table 4. Contamination rate and effect of plant growth regulators for callus and shoot formation in inflorescence apex culture of *Primula* ×*polyantha*.

Combination of plant growth regulators		Explants (no.)	Non contami-nated explants (no.)	Contamination rate (%)	Surviving explants (no.)	Rate of survival explants (%)	Rate of callus formed explants (%)	Amount of callus per explant	Rate of shoot formed explants (%)	No. of shoots per explant
NAA (mg L^{-1})	BA (mg L^{-1})									
0.1	2	48	44	8.3	30	68.2	65.9	1.5	50.0	1.8
0	1	49	45	8.2	12	26.7	4.4	0.1	20.0	0.7
0	2	51	47	7.8	13	27.7	4.3	0.1	14.9	0.6
0	4	52	50	3.8	9	18.0	2.0	0.0	10.0	0.3

CONCLUSIONS

In this study, we understood that we could lengthen an inflorescence by gibberellin treatment, and could obtain shoots by the inflorescence apex culture at a high rate. It will be necessary to define more closely the optimum point about the above points. In that case, it may be possible to perform the micropropagation of selected primula polyanthus plants.

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Induction of polyploidy plants through colchicine treatments in balloon vine (*Cardiospermum halicacabum*)©

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Abstract

The balloon vine (*Cardiospermum halicacabum*) is a popular plant with balloon-like fruits, but there are no cultivars. Polyploidization could be used to breed new cultivars. Here, we tried to breed polyploid balloon vines for improved horticultural value. Polyploidization was induced by immersing germinated seeds in colchicine solution. The frequency of tetraploids depended on concentration and treatment time. No tetraploids were obtained from seedlings treated at 1 mM. Some tetraploids were obtained from seedlings treated at 10 mM for 24, 36, or 48 h. All were diploid–tetraploid chimeras. Some chimeras had bigger leaves and fruits than diploids, but others showed no difference. Tetraploid seeds were obtained from chimeras and their progenies. From tetraploid seeds treated with 10 mM colchicine for 24 or 48 h, five survived, including one tetraploid–octoploid chimera. It had thicker leaves and grew more slowly than diploids and tetraploids.

INTRODUCTION

The balloon vine (*Cardiospermum halicacabum* Linn.) is a vine plant in the Sapindaceae. It is popular in horticulture for its balloon-like fruits and heat tolerance, but there are no cultivars. The *Cardiospermum* genus comprises 16 known species with pantropical distribution (Ferrucci, 2000). There is no report of interspecific crosses within the genus for improving horticultural value. Polyploidization is a valuable technique for breeding. *Cardiospermum halicacabum* is diploid (x=11, $2n$=22) (Sugiura, 1931). Many other species in the *Cardiospermum* genus are also diploid, and only *C. bahianum* is known as polyploid ($2n$=$4x$=36) (Urdampilleta et al., 2013). Polyploidization could be useful for breeding new and valuable cultivars of balloon vine. Here, we bred tetraploids and octoploids by polyploidization for improved horticultural value.

MATERIALS AND METHODS

Balloon vine seeds were soaked in 95% sulfuric acid (Wako Pure Chemical Industries, Ltd, Japan) for 1 h and then germinated on wet filter paper in a glass Petri dish at 25°C in constant light. Polyploidization was carried out by immersing germinated seeds in colchicine (Wako) solution containing 10% (v/v) dimethyl sulfoxide (Nacalai Tesque, Inc., Japan). To induce tetraploid plants, we treated diploid seeds with 1 or 10 mM colchicine for 12, 24, 36, or 48 h (Table 1). The treated seeds were sown in a 1:2 (v/v) mixture of perlite and BM2 culture soil (Berger Peat Moss Ltd., Canada), and then grown in a glasshouse under natural day length. To induce octoploid plants, we treated tetraploid seeds with 10 mM colchicine for 24, 48, or 72 h. Tetraploid seeds were obtained from diploid–tetraploid chimeras and their tetraploid progenies. Ploidy level was determined from young leaves with a PA flow cytometer (Partec GmbH, Germany) according to the manufacturer's instructions. Signals with a relative fluorescence intensity of <20 were ignored as they were mostly noise.

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Table 1. Effects of colchicine treatment on induction of polyploids in diploid balloon vine.

Colchicine conc.	1 mM			10 mM		
Duration (h)	24	48	12	24	36	48
No. of seeds treated	59	20	35	59	23	20
No. of survivals	44	13	16	25	7	10
No. of diploids	44	13	16	19	6	8
No. of tetraploids	0	0	0	6	1	2
Survival ratio (%)	74.6	65.0	45.7	42.4	30.4	50.0
Polyploidization ratio (%)	0	0	0	10.2	4.3	10.0

Polyploidization ratio (%) = No. of tetraploids / No. of seeds examined × 100.

RESULTS AND DISCUSSION

Two hundred and sixteen (216) diploid seeds were treated with colchicine solution to induce tetraploid plants and ploidy level of 115 surviving plants were determined with a PA flow cytometer. Untreated (diploid) plants showed a single peak relative fluorescence intensity of about 100 (Figure 1A). Some surviving plants showed two peaks of about 100 and 200 (Figure 1B). Flow cytometric measurement of young leaves can show two peaks (Galbraith et al., 1983), due to rapidly dividing G2 and M phase cells, in contrast to the single peak of G1 phase cells. Peak of the M phase cells should be lower than the G1 peak. In our results, cell counts of each peak were nearly the same, so these plants were deemed to be chimeric plants with both diploid and tetraploid cells. Treatment with 10 mM colchicine for 12 h or 1 mM for 24 or 48 h produced no tetraploid plants. Treatment with 10 mM for 24 h produced six diploid-tetraploid chimeric plants (polyploidization ratio of 10.2%), treatment for 36 h produced one plant (4.3%), and treatment for 48 h produced two plants (10.0%) (Table 1). As colchicine is a toxic chemical that prevents cell division by inhibiting mitosis (Taylor, 1965), the optimum concentration for polyploid induction reduces survival. Survival after 1 mM treatment was relatively high; it is possible that 1 mM was not high enough for polyploid induction.

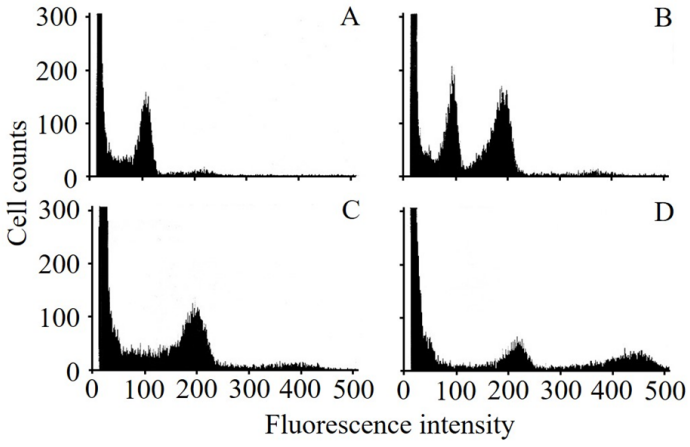

Figure 1. Flow cytometric histograms of balloon vines with different ploidy levels: (A) diploid, (B) diploid–tetraploid chimera, (C) tetraploid, (D) tetraploid–octoploid chimera.

We compared the morphological characteristics between diploid–tetraploid chimeric plants and diploid plants (Figure 2). Balloon vine has biternate leaves. Leaflets of diploids do not overlap one another (Figure 2A). The leaflet size and shape of the chimeric plants were separated into two types. One was very similar to diploids, and leaflets didn't overlap (Figure 2B). The other had larger and thicker leaflets that overlapped one another (Figure 2C). The

chimeras with larger and thicker leaflets invariably produced larger fruits than diploids. Organ size varied among plants and shoots, even though all chimeras had tetraploid cells. In general, the shoot meristem of plants has three cell layers (L1, L2, and L3), which are maintained after differentiation into organs. In leaves, the epidermis consists mainly of L1 cells, the palisade layer mainly of L2 cells, and the spongy parenchyma mainly of L3 cells (Sussex, 1989). According to Dermen (1960), cytochimeras with an L1–L2–L3 ploidy of 2*x*–4*x*-2*x* are much more likely to be tetraploids than those with either 2*x*-2*x*-4*x* or 4*x*-2*x*-2*x*. Adaniya and Tamaki (1991) reported that cytochimeric *Allium wakegi* with an L1-L2-L3 ploidy of 2*x*-4*x*-4*x* showed similar growth characteristics to tetraploids, but 4*x*-2*x*-2*x* plants were similar to diploids. In our study, the ploidy of each cell layer was not measured, but the ploidy of L2 may be important for determining organ size in the balloon vine, too.

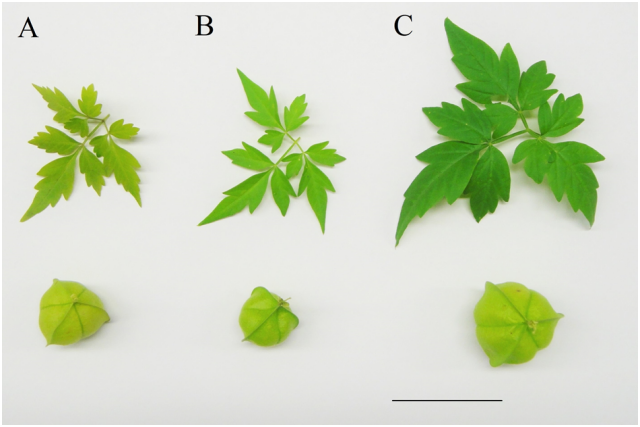

Figure 2. Leaves and fruits of balloon vines with different ploidy levels: (A) diploid, (B, C) diploid–tetraploid chimeras. Scale bar = 5 cm.

Tetraploid seeds obtained from diploid–tetraploid chimeras and their progenies were used for octoploid induction by treatment with 10 mM colchicine for 24, 48, or 72 h (Table 2). Five plants survived 24 h treatment, but none survived 48 or 72 h. The survival ratio in 24 h treatment (2.8%) was much lower than that of diploid seeds under the same condition (42.4%). The tetraploid balloon vines grew more slowly than the diploids, so their growth characteristics should be different. We suspect that the colchicine concentrations that trigger growth inhibition were also different. Treatment at a lower concentration or for a shorter duration might be optimal for octoploid induction. Untreated tetraploids had a peak relative fluorescence intensity of about 200 (Figure 1C). One survivor of treatment with 10 mM for 24 h showed two peaks of about 200 and 450 with nearly the same cell counts (Figure 1D), and thus appears to have been a tetraploid-octoploid chimera. The other four survivors had a peak at about 200 and are likely to have been tetraploids. The tetraploid–octoploid chimera had thicker, crumpled leaves and grew more slowly than both diploids and tetraploids.

Table 2. Effects of colchicine treatment on induction of polyploids in tetraploid balloon vine.

Colchicine conc.		**10 mM**	
Duration (h)	24	48	72
No. of seeds treated	177	47	22
No. of survivals	5	0	0
No. of tetraploids	4	0	0
No. of octoploids	1	0	0
Survival ratio (%)	2.8	0.0	0.0
Polyploidization ratio (%)	0.6	0.0	0.0

Polyploidization ratio (%) = No. of tetraploids / No. of seeds examined × 100.

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Effects of light intensity, soil acidity, and nitrogen concentration on the vegetative growth of pitaya seedlings©

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INTRODUCTION

Pitaya (*Hylocereus undatus*) is a little-known tropical fruit in Japan. Pitaya fruit contains important nutrients to enhance human health, including dietary fiber, vitamins, minerals, and polyphenols (Mahattanatawee et al., 2006). The fruit also contains oligosaccharides known to improve the intestinal environment (Wichienchot, 2010). Red pitaya fruit contains betalains, pigments with antioxidant activity that are used as natural dyes and to remove active oxygen species (Wu et al., 2006; Tenore et al., 2012).

The pitaya flower opens at night and wilts the next morning. It resembles the queen of the night flower (*Epiphyllum oxypetalum*), which is approximately 25-30 cm in length. The queen of the night flower usually blooms 3-4 times a year, whereas the pitaya flower blooms 5-6 times a year, from spring to autumn.

Pitaya plants tolerate temperatures as low as -4°C; thus, they could grow during the winter season in the warmer regions of western Japan (Fumuro et al., 2013). Pitaya requires relatively simple management practices and is expected to increase in popularity.

Comparisons of pitaya cutting propagation techniques have identified an auxin that promotes rooting (Fumuro, 2011). Fumuro (2015) measured the effects of growth regulators on pitaya growth and reported that gibberellin promoted cladode growth. Alternatively, spraying cladodes with ethephon and applying 1-naphthaleneacetic acid (NAA) inhibited cladode growth (Fumuro, 2015).

Pitaya is a type of epiphytic cactus native to tropical forests. Pitaya growth is weak in the presence of strong solar radiation and requires cheesecloth to provide shade. The optimal light intensity varies by species (Le Bellec et al., 2006). The optimal light intensity in Japan, which is at higher latitude than the location where pitaya is normally found, has not been determined.

Pitaya is grown in a wide range of climates, from arid to high-rainfall areas (Le Bellec et al., 2006). Although slightly acidic soil is common in Japan, there is a difference from most strongly acidic to alkaline soils. Pitaya is adapted to a wide range of soil types, and is minimally affected by soil acidity. However, the relationship between seedling growth and soil acidity is unclear. In addition, the optimal method of fertilization for pitaya cuttings has not been established.

This study was conducted to measure the effects of light intensity, soil acidity, and nitrogen concentration on pitaya growth to improve the efficiency of pitaya cutting production.

MATERIALS AND METHODS

All experiments were performed in 2006 and 2007 using rooted cladodes growing in pots (13.5 cm diameter, 11 cm height) in a greenhouse (6.3 m width, 9.6 m length; about 60 m^2) at the experimental farm of Kinki University. Cladode cuttings were collected from 4- and 5-year-old trees grown in the greenhouse (6.5 m wide, 20.8 m length; about 135 m^2). Each cutting was trimmed to 11-12 cm, sprayed with a solution of 500 ppm of benomyl and 150 ppm of streptomycin, and placed in a shaded, well-ventilated location for 48 h to allow the wounds to heal. The cuttings were dipped into a 2,000 ppm solution of NAA (Wako Pure Chemical Industries, Osaka, Japan) for 10 s to promote rooting. Each cutting was planted at a depth of 4 cm in a container (22 cm wide, 65 cm long, 18 cm deep) filled with a soil mixture

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(mountain soil, peat moss, and vermiculite mix (2:1:1, by vol.), and grown in a greenhouse under 50% shade. Cladode cuttings were rooted about 1 month after preparation. Using the cuttings propagated as described above, the following experiments were conducted.

Experiment 1. the effect of light intensity

In 2007, the 3- to 4-month-old rooted cuttings with one new cladode, approximately 10 cm long, were used. Ten rooted cuttings were shaded with three types of cheesecloth to create variations in light intensity; 10 untreated cuttings were used as controls.

The light intensity prior to shading was measured on 14 April, inside and outside the greenhouse, using a solar radiation meter. The light penetration rate (LPR) of the controls (non-shaded) was 86% outside the greenhouse. Light penetration through the three types of cheesecloth was 10, 45, and 71% outside the greenhouse. If a new cladode started to sprout from the old cladode it was removed immediately. The average lengths of the old rooted cladodes for the three types of cheesecloth and the control were as follows: 10%, 12.1±0.7 cm (mean±standard deviation); 45%, 12.6±1.2 cm; 71%, 12.3±2.1; and 86% (controls), 11.8±1.2 cm.

The plants were watered once daily, and 5 g of delayed release fertilizer (10 N:10 P_2O_5:10 K) was applied to each pot at the start of the experiment. The greenhouse was ventilated by a fan when the internal air temperature reached 30°C to maintain the temperature below 35°C. Both the side windows and skylights remained open from April to November.

The degree of sunburn occurrence was rated using five levels based on the percentage of new cladode death: 0 (0%), 1 (10%), 2 (20%), 3 (30%), 4 (40%), and 5 (more than 50%). The lengths of all new cladodes were measured 30 (14 April), 60 (14 May), 90 (13 July), and 120 days (12 August) after shading. The plants were separated into new cladodes, old cladodes, and roots on the last day of measurement. Fresh weights were recorded before drying to a constant dry weight in an oven at 80°C The weights were recorded after drying, and the dry matter percentage of each organ was calculated.

Experiment 2: the effect of soil acidity

In 2007, 3- to 4-month-old rooted cuttings with one new cladode, approximately 10 cm long, were used in this experiment. Soil pH adjustment was performed by adding hydrated lime to mountain soil, which was strongly acidic (around pH 4.5). Acidic soil with a pH of 5.5, neutral soil with a pH of 7, and alkaline soil with a pH of 8 were prepared.

On 14 May, the plants were transplanted to larger pots (18 cm diameter, 15 cm height) using the adjusted and unadjusted soils as described above. Similarly, on 26 July the soil was replaced with recently adjusted soil or unadjusted soil to correct for any changes in soil acidity. Ten rooted cuttings were used for each soil acidity level.

To prevent sunburn, the seedlings were covered with cheesecloth to provide 50% shade. The seedlings were watered once daily, and liquid fertilizer (N:P_2O_5:K = 6:10:5), diluted 500-fold, was applied at 200 ml per pot on 22 May and 9August. On 22 October, the new and old cladode lengths were recorded and separated into new cladodes, old cladodes, and roots. Their fresh and dry weights were measured as described above. The average lengths of the old cladodes were as follows: strongly acidic soil, 12.1±2.2 cm; acidic soil, 12.5±1.0 cm; neutral soil, 12.5±1.2 cm; and alkaline soil, 11.9±1.8 cm.

Experiment 3: the effect of nitrogen concentration

In 2006, 2-month-old rooted cuttings without new cladodes were used in this experiment. The mountain soil did not contain inorganic or organic components, and the electrical conductivity was almost 0. The nitrogen concentration was adjusted to 25, 50, and 100 ppm, applying 150 ml of ammonium nitrate solution per pot once a week from 6 July to 31 January of the following year. As a control (N = 0 ppm), tap water was applied. With respect to phosphorous and potassium, liquid fertilizer (0 N:6 P_2O_5:4 K) lacking nitrogen was applied biweekly by diluting it 500-fold (120 ppm P_2O_5 and 80 ppm K) under 50% shade. Fifteen plants were used for each nitrogen concentration.

The lengths and rate of new cladode occurrence were measured once a month until 7 February. They were separated into new cladodes, old cladodes, and roots on the last day of measurement. Their fresh and dry weights were measured as described above. The average lengths of the old cladodes of the seedlings were as follows: 0 ppm, 11.2±0.5 cm; 25 ppm, 11.6±0.5 cm; 50 ppm, 11.3±0.4 cm; and 100 ppm, 10.9±0.7 cm.

RESULTS AND DISCUSSION

Experiment 1: the effect of light intensity

The extent of sunburn occurrence for 86 and 71% LPR was 1.4±0.7 and 0.1±0.3, respectively. Sunburn did not occur at 10 or 45% LPR. The new cladodes were the longest for both 71 and 45% LPR, followed by 86 and 10% LPR (Figure 1). The fresh weight of new cladodes was the highest for 71% LPR, followed by 45 and 86% LPR, and the lowest for 10% LPR (Table 1). The root fresh weight was the highest at 71% LPR, and there were no significant differences between other LPRs. The total fresh weight was the highest for 71% LPR, followed by 45 and 86% LPR, and lowest for 10% LPR. As for flesh weight, the dry weight of the new cladodes was the highest for 71% LPR, followed by 45 and 86% LPR, and the lowest for 10% LPR. The root dry weight was also the highest for 71% LPR, and there were no significant differences between other LPRs. The dry weight percentages of old cladodes were higher for 71 and 86% LPR than that of 10% LPR. The new cladode dry weight percentages were the highest for 71% LPR, followed by 45 and 86% LPR, and the lowest for 10% LPR (Table 2).

Table 1. The effect of light intensity on the flesh and dry weights of each organ in pitaya seedlings.

Light penetration rate (%)	Flesh weight (g)[1]				Dry weight (g)[1]			
	Old cladode	New cladode	Root	Total	Old cladode	New cladode	Root	Total
10	64.2 a[2]	60.3 c	4.5 b	129.0 c	5.0 a	4.1 c	0.8 b	9.9 c
45	63.7 a	158.1 b	4.7 b	226.5 b	5.5 a	13.8 b	0.9 b	20.2 b
71	56.6 a	188.3 a	6.8 a	251.7 a	5.5 a	19.4 a	1.4 a	26.3 a
86	51.8 a	58.3 b	4.5 b	214.6 b	4.9 a	14.2 b	1.0 b	20.1 b

[1]Measured after 4 months of the treatment.

[2]Values in a column followed by the same letter are not significantly different (P<0.05) by Tukey-Kramer's multiple range test.

Table 2. The effect of light intensity on the dry weight percentage of each organ in pitaya seedlings.

Light penetration rate (%)	Old cladode (%)	New cladode (%)	Root (%)
10	7.8 c[1]	6.9 c	19.6 c
45	8.6 b	8.7 b	19.9 b
71	9.8 a	10.3 a	20.1 b
86	9.4 ab	9.0 b	20.9 a

[1]Values in a column followed by the same letter are not significantly different (*P*<0.05) by Tukey-Kramer's multiple range test.

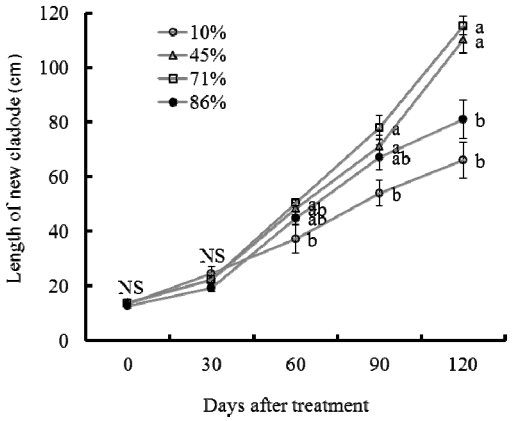

Figure 1. The effect of light penetration rate on the elongation of new cladodes in pitaya seedlings. Vertical bars represent ± SE. NS and values followed by the same letter are not significantly different (P<0.05; Tukey-Kramer multiple range test).

Pitaya, which is native to tropical forests, does not tolerate strong solar radiation. Pitaya also does not require a high photosynthetic photon flux density for photosynthesis (Nobel and Barrera, 2004). Raveh et al. (1998) reported that the most favorable conditions for growth and fruit production were 30% shade for *H. polyrhizus*, while, for *Selenicereus megalanthus*, 60% shade was optimal. This study showed that 30% shade is best for *H. undatus*, as reported by Raveh et al. (1998) for *H. polyrhizus*.

Experiment 2: the effect of soil acidity

There were no significant differences in new cladode length, or in the flesh and dry weights of each organ (Figure 2, Table 3). Cladode growth was not affected by soil acidity at a pH range of 4.5 to 8.

"Kanuma-tsuchi" and "Akadama-tsuchi" gardening soils are popular in Japan and are frequently used to grow horticultural crops. The former is strongly acidic (pH = 4-5), and the latter is slightly acidic (pH = 6-7). Soils mixed with vermiculite, peat moss, or compost are also commercially available. They are often adjusted to a pH that is slightly acidic to neutral. However, the cost of plant production is high because these soils are expensive. Since there is little influence of soil acidity on plant growth, field soil could be used to reduce the production cost.

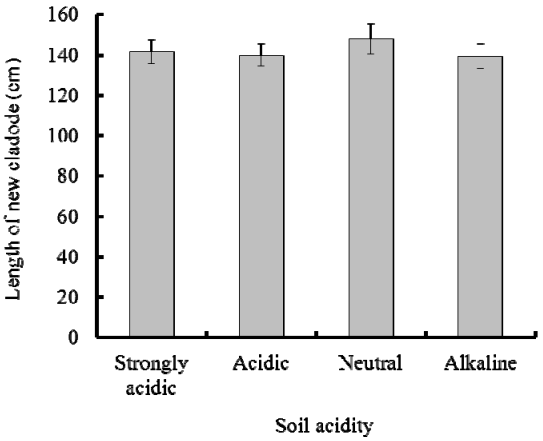

Figure 2. The effect of soil acidity on the elongation of new cladodes in pitaya seedlings. Vertical bars represent ± SE.

Table 3. The effect of soil acidity on the flesh and dry weights of each organ in pitaya seedlings.

Soil acidity	pH	Flesh weight (g)[1]				Dry weight (g)[1]			
		Old cladode	New cladode	Root	Total	Old cladode	New cladode	Root	Total
Strongly acidic	4.5	57.0	193.0	8.6	258.6	5.8	21.6	1.7	29.1
Acidic	5.5	57.9	212.8	9.6	280.3	5.6	22.9	1.9	30.4
Neutral	7.0	59.7	218.1	9.3	287.1	6.2	23.7	1.9	31.8
Alkaline	8.0	55.2	206.6	7.9	269.7	5.8	23.2	1.6	30.6
Significance[2]		NS	NS	NS	NS	NS	NS	NS	NS

[1]Measured after 5 months of the treatment.

[2]NS: non-significant at P=0.05.

Experiment 3: the effect of nitrogen concentration

The rates of new cladode occurrence after 1 month of treatment were approximately 60-90%. The rate increased as the nitrogen concentration increased (Figure 3). New cladodes were observed at all nitrogen concentrations after 2 months of treatment. The new cladodes were longer as the nitrogen concentration increased (Figure 4). The growth of new cladodes in 0 ppm nitrogen stopped after 2 months of treatment, and growth in 25 ppm nitrogen remained low after 2 months of treatment. As the nitrogen concentration increased, the fresh and dry weights of the total plant, new cladodes, and roots increased (Table 4).

Several reports have been published regarding the effect of chemical fertilizer application on pitaya growth. The application of chemical fertilizers, including nitrogen, improves tree growth and the fruit yield and quality of pitaya (Muchjajib and Muchjajib, 2012; Chakma et al., 2014). However, research focusing on a suitable nitrogen concentration for potted seedlings has not been reported. The results of this study indicate that the nitrogen concentration for seedling production was sufficient at 50-100 ppm.

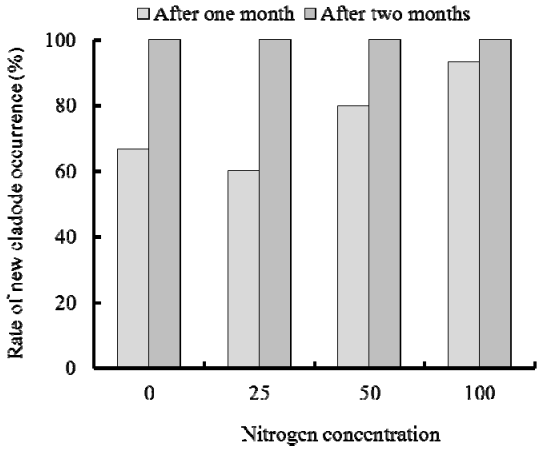

Figure 3. The effect of nitrogen concentration on the rate of new cladode occurrence in pitaya seedlings.

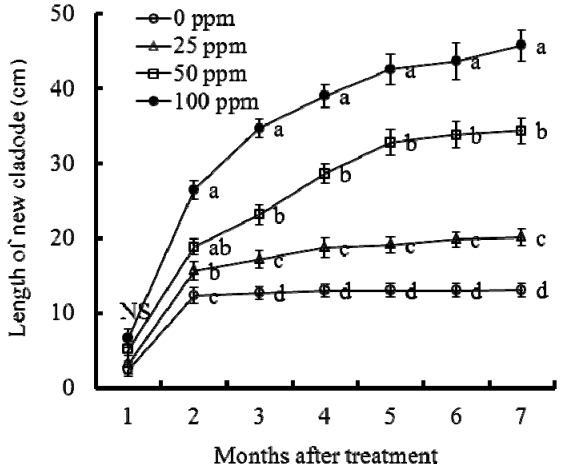

Figure 4. The effect of nitrogen concentration on the elongation of new cladodes in pitaya seedlings. Vertical bars represent ± SE. NS and values followed by the same letter are not significantly different (P<0.05; Tukey-Kramer multiple range test).

Table 4. The effect of nitrogen concentration on the flesh and dry weights of each organ in pitaya seedlings.

Nitrogen concentration (ppm)	Flesh weight (g)[1]				Dry weight (g)[1]			
	Old cladode	New cladode	Root	Total	Old cladode	New cladode	Root	Total
0	53.3a[2]	23.3d	1.1c	77.7d	6.0a	2.5d	0.2c	8.7d
25	56.8a	42.2c	1.9b	100.9c	6.3a	4.6c	0.4b	11.3c
50	59.0a	65.6b	2.5b	127.1b	6.6a	7.1b	0.5b	14.2b
100	56.0a	82.6a	3.2a	141.8a	6.4a	9.1a	0.7a	16.2a

[1]Measured after 5 months of the treatment.

[2]Values in a column followed by the same letter are not significantly different (P<0.05) by Tukey-Kramer's multiple range test.

CONCLUSIONS

The results of this study indicate that a suitable light intensity for the growth of pitaya seedlings is about 70%. Seedling growth was unaffected by soil acidity, and 50–100 ppm was found to be a suitable nitrogen concentration.

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Improvement in rooting of cuttings of FDR-1, a dwarfing rootstock for kaki©

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INTRODUCTION

The 'Fuyu' Japanese persimmon (*Diospyros kaki* Thunb.) tree grafted onto FDR-1 (Fukuoka Dwarfing Rootstock No. 1) showed a semi-dwarfing growth habit in the orchard of Fukuoka Agriculture and Forestry Research Center. After cutting off from the rootstock, the roots sprouted root-suckers. The explants (buds) were collected from the root-suckers and were micropropagated easily. Young trees of 'Taishu' Japanese persimmon grafted onto the micropropagated FDR-1 rootstocks showed dwarfing growth (Haranoushiro et al., 2010). Although the cutting propagation of kaki in the mist system was shown to be a cheap and commercial propagation (Tetsumura et al., 2011), the rooting percentages of cuttings of FDR-1 was low in our preliminary experiments. Hence, the objective of this study was to improve rooting of cuttings of FDR-1.

MATERIALS AND METHODS

Root-suckers from FDR-1 roots and shoots of FDR-1 hedges were collected from May to September in 2011-2015. Single-node stem cuttings with one leaf and one bud were prepared from the root-suckers and the shoots, dipped at their bases in 50% aqueous ethanol with 3000 ppm indole-3-butyric acid (IBA) for 5 s, planted singly in a plastic pot (EG-90, 300 ml, Minamide Inc., Japan) which was filled with Metro-Mix® 360 (Sun Gro, Horticulture Distribution Inc., Washington D.C.), and then placed under a vaporized aluminum netting in a propagation frame covered with plastic film. The propagation frame was intermittently misted (30 s mist and 15 min stop in the daytime) and was ventilated with fans when the ambient air reached 38°C. In addition to this mist system, a fog system, in which the unit with humidification spray nozzles (Mini Fogger II, Spraying Systems Co., Japan) intermittently produced fog (30 s mist and 1 min stop in the daytime) under a vaporized aluminum netting in a propagation frame covered with plastic film without fans, was used in this study. Data loggers (TR-72i, T&D Corporation, Japan) measured the temperature and the relative humidity in the systems. In vitro shoots of FDR-1 tended to root better when dipped in 2.5 mM IBA, which was twice as high as the concentration normally used (Tetsumura et al., 2015b). Hence, some FDR-1 cuttings were dipped in 6000 ppm IBA for 5 s. Ten cuttings per each treatment in each year were used. The rooting percentage and number and length of roots were investigated 2 months after cutting, and then the rooted cuttings were transplanted singly to a plastic pot (EG-105, 400 mL, Minamide Inc., Japan). The pots were filled with Metro-Mix® 360 and were placed in a propagation frame covered with plastic film but opened at the sides. Soon after transplanting, leaf SPAD values of the rooted cuttings were measured with a chlorophyll meter (SPAD-502, Minolta Camera Co., Japan). The survival of rooted cuttings was investigated in April of the following year.

RESULTS AND DISCUSSION

Only 6% of the cuttings collected in July, August, and September rooted. Although 30% of the cuttings collected in mid-May rooted, the bases of some cuttings were damaged by the treatment of high concentration IBA solution possibly because they were soft. Collecting the cuttings in May did not seem to be practicable, because at that time we collected fewer cuttings from the stock plants, shoots from which did not elongate well.

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Table 1 shows the results of the cuttings collected at the beginning of June in 2012, 2014, and 2015, in which the stock plants produced many shoots for cuttings and the irrigation systems worked well. The rooting percentages was higher in the cuttings from root-suckers than those from hedges (Figure 1.). The fog system was superior to the mist system in the rooting percentages of FDR-1 cuttings. The rooting percentages of the cuttings treated with 6000 ppm IBA was higher than those with 3000 ppm IBA.

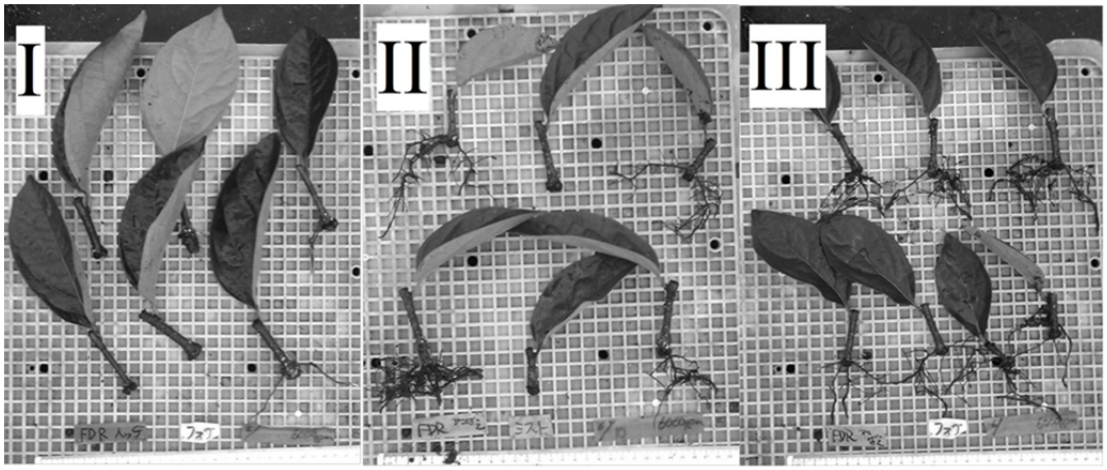

Figure 1. FDR-1 cuttings 2 months after planting on 10 June 2014: (I): the cuttings collected from hedges, irrigated by fog, and treated with 6000 ppm IBA; (II): the cuttings from root suckers, treated with mist, and with 6000 ppm; (III): the cuttings from root-suckers, treated with fog and with 6000 ppm.

The cuttings from root-suckers of 'Nishimura Wase' Japanese persimmon and 'MKR1' dwarfing rootstock for kaki also rooted better (Tetsumura et al., 2001, 2011, 2015a). The relative humidity in the mist system dropped below 50% in the daytime of summer because the fans ran to keep the temperature setting. On the other hand, the relative humidity and the temperature in the fog system closed by plastic film were maintained over 95% and below 40°C, respectively. The amount of water used per 10 m^2 in the fog system was 3.6 L h^{-1}, which was one-fifth of those in the mist system. The small amount of water provided by the fog system possibly reduced leaching from the leaves of cuttings, which showed higher SPAD value (51.1 of the leaves of the rooted cuttings in the fog system vs. 42.9 of those in the mist system in 2014 and 50.9 vs. 43.7 in 2015), because SPAD values of persimmon leaves were highly correlated with N concentration of the leaves (Choi et al., 2011). In these conditions, photosynthetic rates of the leaves in the fog system may have been higher than those in the mist system, and consequently the rooting percentages of the former were assumed to be higher than those of the latter.

The treatments improving the rooting percentages did not improve the number and length of roots (Table 1). Almost all of rooted cuttings survived 1 year after cutting. FDR-1 nursery plants propagated by cutting were seemed to be more vigorous than 'MKR1'.

In conclusion, rooting of FDR-1 cuttings was improved when they were collected from root-suckers at the beginning of June, put in the fog system and treated with 6000 ppm IBA. However, there were annual variations in the rooting percentage. For example, 80% of the cuttings collected from hedge, put in the fog system and treated with 6000 ppm IBA in 2012 rooted, while 20% of those cuttings rooted in 2014. Hence, we should investigate other factor influencing rooting of FDR-1 cuttings.

Table 1. Effects of stock plant, irrigation system and concentration of IBA on rooting of cuttings of FDR-1, a dwarfing rootstock for kaki, collected at the beginning of June in 2012, 2014 and 2015. (The data of Root + Fog + 3000 ppm and Root + Fog + 6000 ppm were collected in 2014 and 2015).

Stock plant	Irrigation system	Conc. of IBA (ppm)	Rooting (%)	Roots per rooted cutting	Total length of roots (cm)
Hedge	Mist	3000	17±10	1.9±0.1	14±0
		6000	27±12	2.1±0.2	11±1
	Fog	3000	20±6	3.1±1.1	24±5
		6000	47±17	2.3±0.1	18±1
Root	Mist	3000	33±5	2.0±0.3	15±2
		6000	60±9	4.0±0.9	26±5
	Fog	3000	45±8	1.5±0.4	11±3
		6000	80±7	3.3±0.6	22±3

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Production of interspecific hybrid plants between *Hydrangea scandens* subsp. *chinensis* and *Hydrangea macrophylla* via ovule culture©

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BACKGROUND

McClintock systematically described the genus *Hydrangea* (McClintock, 1957). She included 23 species with a disjunctive distribution in both eastern Asia, eastern North America, and South America. *Hydrangea macrophylla* (Thunb. Ex J.A. Murr.)Ser. is the most popular of the species, and it is one of the most commercially important flowering shrubs in the world.

Hydrangea macrophylla native to Japan and China was cultivated in Japan long before introduction into Europe in the 1800s (McClintock, 1957; Wilson, 1923). For this species, numerous cultivars with showy colorful flowers have been bred since the early 1900s through selection of natural mutants and intraspecific crosses among a limited number of early ancestral taxa.

BREEDING

Although breeding of *H. macrophylla* has been successful, further improvements in flower shape, flower color, and growth habit are desirable. *Hydrangea scandens* subsp. *chinensis* is a small shrub that is native to south and southeast Asia and valued for its evergreen foliage, remontant flowering or reblooming, and broad adaptability in mild climates.

Cross-pollination between *H. scandens* subsp. *chinensis* and *H. macrophylla*, and subsequent ovule culture in half-strength MS medium (Murashige and Skoog, 1962) without any plant growth regulators resulted in the production of three interspecific hybrid plants (Figure 1). The hybridity of these plants were confirmed by RAPD analysis. The hybrid plants had flower and leaf morphologies intermediate between the two parental species. Since the hybrid plants showed more vigorous growth than both parents, had evergreen foliage, and flowered in winter to early spring, it has sufficient horticultural merit for commercialization and may be suitable for greenhouse pot culture.

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Figure 1. Interspecific hybrid plant between *Hydrangea scandens* subsp. *chinensis* and *H. macrophylla*.

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Study of native *Hosta* species on Shikoku Island, Japan©

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Hostas are perennial herbs native to eastern Asia including Japan, Korea, and China. About 20 species are recorded in Japan now. There are about 12 *Hosta* species on Shikoku Island. Eleven species are recorded just for Kochi Prefecture, among 13 species on Shikoku Island, including: *H. alismifolia* (baran-giboushi), *H. capitata* [syn. *H. nakaiana* (kanzashi giboushi)], *H. sieboldiana* var. *montana* [syn. *H. montana* (ohba giboushi)], *H. sieboldii* (koba giboushi), *H. longissima* (mizu giboushi), *H. longipes* [syn. *H. longipes* var. *caduca* (saikoku iwa giboushi)], *H. gracillima* [syn. *H. longipes* var. gracillima (hime iwa giboushi)], *H. kikutii* var. polyneuron (sudare giboushi), *H. kikutii* var. *caput-avis* (unazuki giboushi), *H. kikutii* var. *tosana* (tosano giboushi), and *H. tardiva* (nankai giboushi). Hostas are used as garden plants, materials for flower arrangements, and for vegetables. Hosta plants are known as a vitamin C rich vegetable in Japan with *H. tardiva* a popular vegetable with slight bitterness in Kochi, Shikoku. However, in Europe and America hostas are used as garden plants with very high popularity for a long time. Philipp F. B. von Siebold introduced Japanese hosta cultivars to Europa at the end of 17th century.

In this study, we investigated the ecology of *Hosta* taxa native to Shikoku Island, Japan. Among four 1A (CR) ranked endangered species in Kochi, we confirmed four sites of native populations of *H. alismifolia* including central Kochi area (three sites), one site in the eastern area (Figures 1 and 2). There are three sites for *H. sieboldii*, and two sites for *H. longissima*, respectively. There was only one site for *H. longipes*. *Hosta sieboldiana* var. *montana* is an important species for horticultural points of view. We confirmed native population in Shikoku Island including three sites for Ehime and Tokushima, and two sites for Kochi. We confirmed wide range of distribution of *H. kikutii* var. *caput-avis* around central and eastern Shikoku area at Tokushu and Kochi. There were sites for native populations for *H. capitata* in Kochi and Tokushima, respectively. There was a wide population range for *H. kikutii* var. *polyneuron* in Kochi and Tokushuma. This species showed much morphological variation in native habit. *Hosta tardiva* was cultivated near farmer's houses, however, this species in Shikoku is rare except for Kochi. We confirmed native population of *H. gracillima* along Shimanto river (western part of Kochi), Ehime. However,we never confirmed population of Kagawa. Classification of *Hosta* species is considered difficult, because there are many variations in native *Hosta* species, which shows large gene flow. We are checking characteristics of *Hosta* by application of DNA analysis. *Hosta alismifolia* which is distributed in Kochi showed strong relationship with *H. longissima*. We will try system analysis of Shikoku hostas.

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Figure 1. *Hosta alismifolia*.

Figure 2. Habitat of *Hosta alismifolia* in Kochi Prefecture.

Both the stable production and stable supply get trust from the consumer – production system of Central Rose Co. Ltd.©

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I started to product rose seedlings about 40 years ago and changed to potted plant production of mini roses 27 years later. Now our company supplies about 30% of the potted mini roses in Japan (about 2,000,000 pots per year Figure 1).

Figure 1. Examples of various mini roses from Central Rose Co. Ltd.

When I started rose production, I contracted with a major company and supplied rose seedlings. However, because of my physical condition I gave up seedling production. After that I went to Europe in 1989 and inspected the European market and production systems. I observed the mini-rose production system, which had just started in the Netherlands, and became intrigued by it. And so I decided that, "I'll start this mini rose production which no one had yet begun in Japan." I began production in a greenhouse of about 1,000 m^2. I'm expanding the scale of the greenhouse every year with floral demand expanding and now I am producing about 20,000 m^2 at present.

An advanced production system from the Netherlands was introduced into this production. To be able to do stable rose production, I applied a linear factory automation

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system to all the greenhouses. I introduced both liquid culture system and moving bench system and succeeded (Figures 2 and 3).

Our work is caring for nature. However, both the stable production and stable supply are essential to satisfying the request from vendor. This requires that the greenhouse environment not be influenced by the outside weather. I believe that our customer's trust contributes to the future sales.

Figure 2. Automatic watering (left) and cultivation of mini roses in pots (right).

Figure 3. Mini roses, just before shipping.

Effects of soil conditioner FFC-Ace© on inhibited plant growth under acidic soil conditions©

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INTRODUCTION

Since 1984, Akatsuka Garden Company has focused on the behavior of certain ions, especially the iron ions in water, and interactions of water molecules with them. We have continued research on various solutions to not only accelerate plant growth, but also activate physiological functions of plants. Based on this research, we have developed FFC materials such as "FFC-Ceramics" (for water improvement), "FFC-Ace©" (for soil improvement), and others.

In addition, many agricultural producers in Japan have been utilizing FFC materials to rejuvenate plants and increase profits. Those producers have also explored many other possible methods of using FFC materials and consequently found good ways that benefit their actual production sites.

As a result, they have obtained many advantages over years of use, such as improved productivity, cost reduction, decreased dependence on agricultural chemicals, among others. Additionally, it has been reported that FFC-Ace enhances the growth of plants under laboratory conditions while improving disease resistance, drought resistance, and salt stress tolerance (Ichikawa and Fujimori, 2012, 2013; Ichikawa et al., 2014; Fujita et al., 2010; Hasegawa et al., 2006; Konkol et al., 2012; Shiraishi et al., 2010; Toyoda et al., 2010).

Andosol, which occupies half of field soil area in Japan, contains much organic matter and the soil easily forms an aggregate structure. However, andosol contains much alumina. As the soils acidify, soluble aluminum ions dissolve and inhibit root growth at micromolar concentrations. As a result, crop production decreases (Shoji, 1984; Yamamoto, 2002; Matsumoto, 2003).

In this study, we researched the effect of the soil conditioner FFC-Ace on both the inhibition of plant growth by artificially acidified andosol and on the inhibition of root growth by aluminum ions.

MATERIALS AND METHODS

Experiment 1: using acidified andosol

Sixty grams of andosol was mixed with 1.4 g of $FeSO_4.7H_2O$ to decrease pH value to 4.5 and then was moistened by addition of 10 ml distilled water for acidic andosol. FFC Ace treated andosol was made by addition of 6 g of FFC Ace to the acidic andosol. Sixteen seeds of komatsuna [*Brassica rapa* var. *pekinensis* 'Osaka-shirona' (Japanese mustard spinach)] were sown in FFC Ace treated andosol as well as the acidic andosol. After cultivating the komatsuna under fluorescent light for 11 days (12 h light-dark cycle for 11 days at 25°C, humidity 75%), the length of both shoots and roots were measured.

Experiments 2: water culture using solution containing aluminum ions

Three grams of FFC Ace were mixed with 100 mL of 10.5 mg L^{-1} (as Al) of aluminum chloride solution and was left overnight. The particles of FFC Ace were removed from the immersion water by filter paper and a cellulose syringe filter. The filtrate was diluted three times with distilled water. Distilled water was used as a control as well as 10.5 mg L^{-1} (as Al) aluminum chloride solution diluted three times with distilled water. Seeds of *Brassica rapa* var. *pekinensis* 'Osaka-shirona' (shirona) were sown in each solution, and after cultivating it

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for 4 days, the length of both shoots and roots were measured.

Experiment 3: re-elongation of roots by using water immersed with FFC Ace

Seeds of shirona were sown in about 350 mL of 3.5 mg L^{-1} (as Al) aluminum chloride solutions and were germinated. After 2 days, the seedlings were transferred to distilled water (control). FFC Ace treated water was prepared removing the FFC Ace from water immersed with FFC Ace that was left overnight. As with the control, the germinated seedlings were transferred to FFC Ace treated water for the measurement of shoot and root length after 4 days.

RESULTS AND DISCUSSIONS

In Experiment 1 using andosol which was acidified artificially by $FeSO_4.7H_2O$, the average shoot length of komatsuna grown in FFC Ace treated andosol was about 1.4 times as long as the control, and the average root length was about twice as long as the control. FFC Ace reduced inhibition of plant growth under acidic soil stress such as an acidified andosol.

In Experiment 2, the root elongation of shirona grown in aluminum chloride solution was inhibited. On the other hand, in FFC Ace treated aluminum chloride solution root elongation was not inhibited and we observed elongation to be more accelerated.

Generally, it is said that inhibition to root elongation by aluminum ions is irreversible (Clarkson, 1965; Morimura et al., 1978; Matsumoto and Morimura, 1980). Seedlings of shirona which germinated in aluminum chloride solution were transferred to distilled water or FFC Ace treated water. As shown in Figure 1 of the Experiment 3, further root elongations of seedlings in distilled water did not ever occur in any measurable amount. On the other hand, root elongations of seedlings in FFC Ace treated water restarted and some of the roots grew.

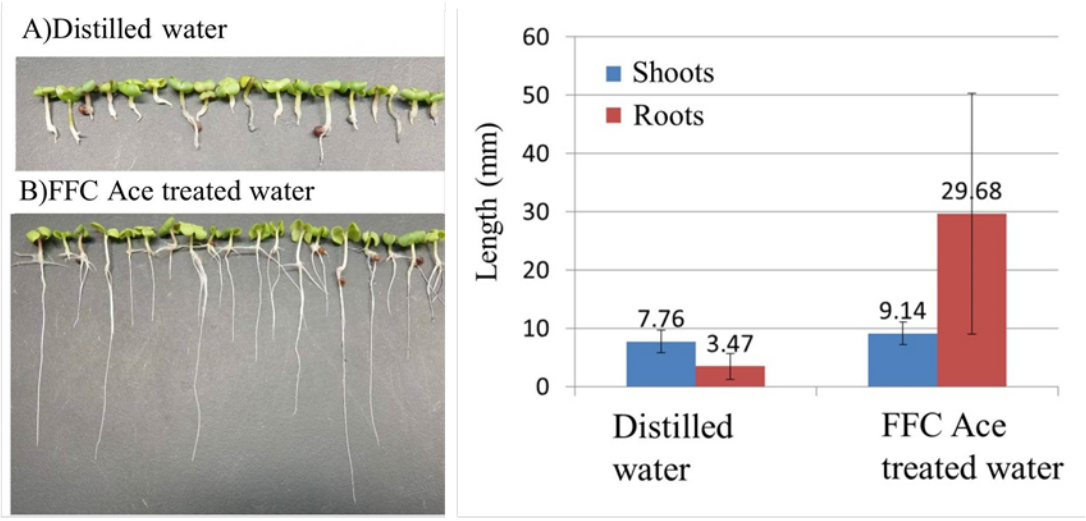

Figure 1. Re-elongation of roots *Brassica rapa* var. *pekinensis* 'Osaka-shirona' (shirona) using FFC® Ace treated water (Experiment 3): left shows seedlings and right shows graphical results.

The results suggest that FFC Ace treated water was effective in restarting the root elongation which had been stopped by aluminum ions. Based on the above results, FFC Ace was effective in reducing growth inhibition of plants under conditions of acidic andosol or existence of aluminum ions. In cultivated land where acidification of soil is accelerated, the application of FFC Ace should enable a noticeable increase in crop productivity.

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The role of botanical gardens in plant conservation©

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INTRODUCTION

Botanical gardens are much about colourful display and arboreal grandeur. Until relatively recently, though, most botanical gardens were largely collections of exotic plants arranged for the pleasure of the public — not unlike the zoos of the past. In other words, exotic eye candy to entertain the customer and not so much about conservation. University-based botanical gardens have always provided special gardens and research collections for the education of experts, but these were mostly inaccessible. Essentially, the typical garden visitor would have no clue as to the value of the plants beyond any intrinsic beauty or other esthetic appeal they might have. It's worth noting that the earliest European botanical gardens were cloistered herb gardens administered by Latin-speaking monks. The walls and yew hedges surrounding them were meant to keep the knowledge in and the riff-raff out. Such academic traditions have been broken — although not always completely — the various kinds of interpretive signage common in modern botanical gardens being an indicator of the newfound willingness to communicate with the public.

(Getting back to our fixation with the exotic...) In many cases, botanical gardens ignored their own regional floras, in deference to the appeal of the foreign and unfamiliar in public displays. The collect-one-of-everything mentality (known in the botanical world as "stamp collecting"), is not in itself destructive, nor even without value on occasion, but it probably indicates the baser instinct to acquire for the sake of acquisition, and then to show off about it. Historically, greed among competing collectors and the imperialistic tendencies of governments sometimes resulted in what was essentially the opposite of conservation.

Nowadays, modern botanical gardens have a better understanding of the potential destructiveness of wholesale collecting and, indeed, of any kind of collecting. Seed collection, for example, which is generally seen as a relatively benign activity, can have serious impacts on the health of some plants in the wild, particularly where seed is the only means of reproduction and natural seed production is limited. This is easily illustrated with plants that require cross pollination to produce fruit: remove enough plants and viable seed numbers decline. Once seed is unavailable in the wild, reestablishment suffers. It is not, of course, appropriate or fair to blame only collecting for the loss in biodiversity that makes conservation so obviously important. Habitat loss through clearing for industrialization, forestry, and large-scale agriculture and over-grazing and over-cutting because of an ever-shrinking resource base, are the most significant factors in the reduction of biodiversity around the world.

CONSERVATION OF WILD PLANTS

The conservation of plants in the wild normally encompasses two broad categories: ex-situ conservation and in-situ (i.e., habitat) conservation. Habitat conservation is generally the purview of botanical gardens that have wild areas or that have the resources to be able to purchase or manage wild habitats. A slightly more arms-length approach to in-situ conservation includes education about and advocacy for threatened habitats. Like other botanical gardens, we have a garden feature at UBCBG — the Garry Oak Meadow and Woodland Garden that features plants from a local endangered ecosystem. This gives us a platform so we can inform the public not only about this important community, but also about biodiversity and conservation in general. Conserving biodiversity in-situ usually starts with documentation, followed by a conservation assessment. Such expertise is often found in botanical gardens. If the area is remote, the ability to train people on the ground in those regions, and thus, build capacity, is also an important aspect of conservation. The Flora of

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Nepal, which was undertaken by Royal Botanic Gardens Edinburgh, is a celebrated example of this approach. Most gardens cannot hope to initiate or maintain projects on that scale, but collaborations are always possible. Indeed, staff at UBC Botanical Garden was involved in biodiversity inventories and conservation assessments for a proposed park in the Hoang Lien Mountains of northern Vietnam in 2004.

Ex-situ conservation is another matter. In ex-situ conservation, propagules are collected and stored or grown out in plantings. Royal Botanic Gardens Kew's Millenium Seed Bank is a good example of ex-situ conservation, having currently banked 13% of the world's wild species. Botanical gardens often make a big deal out of ex-situ measures, but in many cases, these amount to little more than stamp collecting.

There are generally two goals in ex-situ conservation. The first is to perpetuate a species. In the best-case scenario, the entirety of a species' genetic makeup would be included in what's saved, so that it might have in its genome sufficient variation to survive the rigours of current and future environmental disturbance. Such a species will have a better chance of surviving repatriation, which is typically the second goal. Effective ex-situ conservation generally requires significant numbers of seedling plants from across the species' geographical, edaphic (soil-related), and elevational range. This is no easy task, either in collecting or having the space to grow the seedlings out. It becomes simpler and less problematic where populations are already reduced, but in these cases, there are often questions about whether it might not be worth the effort. Perhaps the largest ex-situ initiative is the International Conifer Conservation Program, administered by Royal Botanic Gardens Edinburgh, which encompasses surveys, assessments, propagation and taxonomic research and a network of in-situ and ex-situ planting sites. It goes without saying that some expertise in propagation is often required in plant conservation, as seed recalcitrance and the availability of materials from which to propagate are often contributing factors in conservation status.

EX-SITU CONSERVATION AT UBCBG

In 2010, UBCBG took part in a botanical expedition to the Hengduanshan Mountains of Sichuan Province in China to observe and collect wild *Acer pentaphyllum*. DNA collection for genetic fingerprinting was carried out on this trip and a good supply of seed was collected, as well. Wild populations of *A. pentaphyllum* are being reduced in number and its original distributional range contracted. Known as a “genetic bottleneck,” such a diminishment in numbers notably decreases genetic diversity in the species. In other words, there is a lesser probability that offspring from the remaining populations will display the range of variability that the species once exhibited over its original range. This can mean that traits that confer cold hardiness, drought- or heat-tolerance, or resistance to a particular disease could be lost to the species, particularly if the remaining habitat does not bring those evolutionary pressures to bear. Actually, a number of traits may already be lost. It is, therefore, critically important (if saving the species is the goal) to collect seeds from as many individuals and as many populations as possible and plant them out in a variety of environments. Quarryhill Botanical Garden in California has already established a large ex-situ planting of *A. pentaphyllum* seedlings from two previous expeditions.

UBC Botanical Garden’s plants are derived from 12 different seed collections from the populations of *A. pentaphyllum* that remained in 2010. More than one hundred seedlings have been planted out in various sites and in a field trials area at UBCBG since that time. We certainly don’t expect all or even most of our seedlings to survive the vagaries of Vancouver’s climate, but if a few thrive, they will represent genetic expression that may not be represented by surviving collections elsewhere (such as in California, Belgium, Pennsylvania, or Sichuan). Ultimately, when plants are returned to the wild, they would represent the widest possible genetic complement.

CONSERVATION OF CULTIVATED PLANTS

Botanical gardens have a role in conserving diversity, whether that diversity is embedded in historical cultivars that speak to regionally or locally significant plant breeding

efforts, the rich First Nations legacy of plant selections or the diversity represented in collections of ornamentals. Botanical gardens are first and foremost “gardens” and gardens are places of beauty. Even food gardens have a certain appeal (who isn't fond of eating?) and it's worth pointing out that there is plenty of genetic diversity in cultivated plants. However, as various grass-roots seed-saver organizations have shown us, heritage food varieties, like their wild relatives, are under threat. Botanical gardens are stepping up to demonstrate and explain the value of conservation of food crops, even facilitating seed-sharing events such as “Seedy Saturdays”, which help to preserve and proliferate historically and regionally important open-pollinated taxa.

Ornamental plants represent through their cultivated ranks an enormous diversity. Together with the Plant Collections Network (PCN, an initiative of the American Public Gardens Association), North American botanical gardens have gotten together to identify and assess "national collections" of many such plant groups. The value here is that modern botanical gardens are generally committed to both record-keeping and the dissemination of information about the plants they grow. UBCBG is part of two multi-institutional PCN plant collections: maples and magnolias.

Independently, UBCBG has embarked on a propagation project to conserve the rare ornamental cherries of Vancouver. The goals of the program are to maintain the diversity of cherry trees in the Vancouver area and to identify propagation protocols that facilitate improvements to the health and longevity of the cultivars. There is some evidence that the various incompatibilities and differential rates of growth inherent in grafted plants are contributing to disease susceptibility. Cherries are well adapted to conditions in the Vancouver area and make excellent small-to-medium sized urban trees. However, brown rot and bacterial canker are serious diseases that limit the effective lifespan of these trees. More seriously, older rare cultivars are being lost and, because of stringent plant protection legislation (e.g., limited importations of stone fruits), these plants cannot easily be replaced. Working with the Vancouver Park Board, the Biotechnology Program at the British Columbia Institute of Technology and UBC Botanical Garden Nursery, we now have 35 cultivars, many of them the rarest cherry cultivars, growing on their own roots at the Nursery and in the Garden.

Botanical gardens have come a long way since the days of the monks’ cloistered garden. Conservation initiatives require the broadest engagement and highest level of communication for success. Whether through ex-situ collections, surveys and assessments, propagation research, education about biodiversity and habitat protection or the cultivation of heritage plants, botanical gardens are making a difference.

Plant propagation in the Micronesian region: challenges and measures for sustainable production©

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Abstract

This paper reports on plant propagation of select staple and cash crops in the Micronesian region. While discussing various climatic, socio-economic and technical issues that limit agricultural production, the paper emphasizes the feasibility of plant tissue culture techniques for sustainable plant propagation in the region. The findings include development of successful in vitro plant propagation methods and field transfer techniques for regional cultivars of banana, taro, cassava, sweet potato, pineapple, and black pepper. Plant propagation systems developed for crops at the Micronesia Plant Propagation Research Center serve as a foundation for establishing sustainable agriculture practices and attaining food self-sufficiency in Micronesia.

INTRODUCTION

Agriculture is an important industry and it could greatly help in the economic development and growth in the Micronesian region. Micronesia, lying just on the Equator, enjoys a tropical climate with relatively even, warm temperatures throughout the year. Rainfall is generally plentiful reaching up to 330 in. of rain per year. Nevertheless, drought conditions do occur periodically throughout the Micronesian region, especially when the El Niño condition moves into the western Pacific. At these times groundwater supplies even dwindle to emergency proportions. Tropical typhoons constitute an annual threat, particularly to the low-lying islands. Increasing climate variability has resulted in harsh weather calamities in the form of wave surges, salt water flooding and drought that continually pose challenges for the local farmers who struggle to attain food self-sufficiency by growing crops on their small household farms.

The common food crops in the region include: breadfruit, banana, taro, cassava, yam, sweet potato, pineapple, and citrus. The cash crops include: black pepper, kava, coconut, coffee, and noni. Limited farming, occurring mostly in the form of small farms developed at individual family level with inadequate access to appropriate agricultural resources and trained professional advice along with the frequent climate surges in the region, render the agricultural production of these crops insufficient for supporting the island communities.

Even though Micronesia is free of major insects, pests and pathogens, crop yield is not sustainable in the region. Continued use of traditional planting materials such as suckers, runners and cuttings without any decontamination or revival for years, and lack of knowledge of phytosanitary measures has resulted in pathogen accumulation in locally grown crops. The possibility of procuring seedlings is often obscure because of cost ineffectiveness. Moreover, the quarantine measures are very strict and entry of any planting material is strictly prohibited. Thus, the non-availability of disease-free and elite seedlings has become one of the major bottlenecks in quality production of vegetatively propagated staple and cash crops in the Micronesian region.

Considering the difficulty to maintain disease-free parental stocks in the tropical islands, plant tissue culture is increasingly being appreciated as a potential means of germplasm preservation and production of elite and disease-free planting materials on a mass scale. The Micronesia Plant Propagation Research Center (MPPRC) plays a vital role in germplasm collection, in vitro multiplication, distribution, and conservation of economically

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important staple and traditional medicinal and cash crops in the region. Directed under the United States Department of Agriculture Land-Grant Program, the MPPRC is the only North-Pacific regional facility that is actively engaged in plant biotechnology research, extension and outreach in the region. This paper will share the impact and outcomes of some of the major research projects that are successfully undertaken by the MPPRC and have served as the foundation for developing sustainable agriculture practices in the region.

MATERIALS AND METHODS

Plant material

Healthy explants of traditionally-preferred regional cultivars and selected salt tolerant cultivars of banana (*Musa* species), taro (*Colocasia esculenta* L. Schott and *Cyrtosperma merkusii* H. Schott), cassava (*Manihot esculenta* Crantz), sweet potato (*Ipomoea batatas* L. Lam.), pineapple (*Ananas comosus* L. Merrill) and black pepper (*Piper nigrum* L.) were collected from the field and were thoroughly washed with running tap water prior to surface sterilization by immersion in 70% ethanol followed by a treatment with 2% sodium hypochlorite solution with 5 drops of Tween 20. Apical and/or axillary meristems were excised from sterilized explants for in vitro culture establishment.

Culture medium

Murashige and Skoog (1962) medium (MS medium) supplemented with different concentrations and combinations of growth regulators was used as a basal medium for establishing aseptic cultures of all crops. All media contained 0.8% agar and 3% sucrose. The pH was adjusted to 5.8 prior to autoclaving.

Micropropagation

Aseptic cultures of all collected cultivars were established for multiplication on MS medium supplemented with different concentrations and combination of cytokinins such as thidiazuron (TDZ), 6-benzylaminopurine (BAP) and 6-furfurylaminopurine (KIN) and auxins such as 2,4-dichlorophenoxyacetic acid (2,4-D), indole 3-acetic acid (IAA) and 1-naphthaleneacetic acid (NAA). MS media augmented with IAA or without any growth regulator were used to induce rooting in multiple shoots. Each experiment was replicated three times with minimum 30 replicates per treatment. Complete plantlets were transferred onto sterilized potting mix for acclimatization in the greenhouse for 1-4 months. Completely acclimatized plants were transferred in the screen house where they were kept until field transfer. A one-way analysis of variance was used to determine the level of significance between experimental treatments in all crops. Statistical significance of the results was determined using the least significant difference (LSD) test by Tukey (1953) at 5% level of significance.

RESULTS AND DISCUSSION

Micropropagation methods were developed or optimized for banana (5 µM BAP - Verma, 2008, 2009, 2010), taro (5 µM TDZ or 5 µM IAA and 7.5 µM BAP - Verma, 2008, 2010, 2013; Verma and Cho, 2010), cassava (1 µM BAP), sweet potato (5 µM KIN - Verma, 2008, 2010, 2013), pineapple (9 µM BAP or 2 µM NAA), and black pepper (8 µM BAP or 3 µM IAA)(Figures 1A, B, D, G, I, J, and N). Disease-free and elite seedlings of traditionally-preferred regional cultivars and salt tolerant cultivars of some crops were produced in bulk quantities to ensure the year round availability of high quality planting material. More than 95% survival rate was observed for all crops after 8 weeks of acclimatization in the greenhouse. Acclimatized plants exhibited healthy growth in the nursery where they were kept until field transfer (Figures 1C, K and O). Upon transfer of fully acclimatized plants into the field, healthy and vigorous growth was observed (Figures 1H and L) and excellent and healthy yield was obtained after harvest (Figures 1E, F, H, L, and M).

Figure 1. Taro micropropagation (A and B), taro acclimatization (C), sweet potato micropropagation (D), harvested sweet potatoes (E and F), black pepper micropropagation (G), black pepper cultivation (H), banana micropropagation (I), cassava micropropagation (J), cassava acclimatization (K), banana cultivation (L), harvested pineapples (M), pineapple micropropagation (N), and pineapple acclimatization in greenhouse (O).

Development of successful micropropagation methods of various food and cash crops at the MPPRC reaffirms that tissue culture is of great advantage for mass propagation of vegetatively propagated crops for which traditional breeding methods are time-consuming and disease-free planting materials are in short supply. Advantages of in vitro propagation include: a high plant multiplication rate, physiological uniformity, the availability of disease-free material throughout the year, and safe and rapid dissemination of new salt tolerant plant germplasm (Food and Agriculture Organization, 2010; Verma, 2013).

CONCLUSION

Development of in vitro multiplication methods for traditionally preferred regional cultivars of banana, taro, cassava, yam, sweet potato, pineapple and black pepper through projects run by the MPPRC provide an effective rapid method of plant propagation in the Micronesian region where procuring disease-free seedlings is a major hurdle for sustainable agriculture production. In vitro propagation of these staple food and cash crops through tissue culture provides an excellent advantage over traditional propagation methods and serves as a first step towards developing sustainable agricultural practices to ensure food

self-sufficiency in the Micronesian region.

ACKNOWLEDGEMENTS

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Growth and development of container grown crops in coir based soils©

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INTRODUCTION

As commercial propagators all of us are engaged in the business of selling plants. Some of us grow seeded plugs, while others produce cutting-grown liners to sell to other growers and some of us specialize in difficult-to-propagate plant material by means such as tissue culture. The methods we employ are as varied as are the multitude of plants we seek to reproduce. One thing we all have in common, though, is the need for a medium that meets our own specific needs. Usually, our propagation, potting and canning soils are made up of various organic materials that are combined at different ratios to achieve the desired physical properties for a successful outcome. Components such as sphagnum peat moss, fir mulch, perlite, vermiculite, compost, pumice, rice hulls, loam soil, sand, etc. are some products that come to mind. Over the past few years, a new product has been emerging that has caught the attention of many growers. This product is often times referred to as "coco peat," "coir pith," or simply called "coir."

WHAT IS COIR?

Coir is a byproduct of the coconut industry. The coconut palm, *Cocos nucifera,* bears what we commonly refer to as a coconut. This coconut is covered by a husk that, once removed, is further processed into other products. The long fibers contained in the husk are used to make coco mats, ropes, stuffing for upholstery, etc. However, between these fibers is a corky material that is left behind and is known as coir or coco peat and is what we use in the horticultural industry.

Coir has some very beneficial qualities that make it an excellent component for use in soilless media.

- High water-holding capacity
- Excellent air porosity
- Decomposes slowly
- Rewets easily after getting dry
- Less costly than sphagnum peat moss
- Renewable, sustainable

Coir also has some challenges that need to be addressed before it can be used as an amendment for soilless media.

First, coir contains high amounts of sodium, potassium, and chlorides that need to be buffered to prevent it from having a negative effect on plant growth. This "buffering" is generally achieved using a calcium nitrate solution either before or after incorporation into the mix. Secondly, coir is generally shipped to growers as compressed blocks, usually 5 kg, which need to be hydrated to expand the coir and make it ready for use. Most often, it is during this wetting and expansion process that buffering takes place. This process takes time and space to achieve. There is some specialized equipment on the market that can process the coir blocks which can be easily incorporated into a continuous soil mixing line. The equipment is expensive and choice is limited.

CHALLENGES

Our current soil mix

For the past 4 years at Duarte Nursery, we have been growing our crops in a standard

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peat and rice hull (4:1, v/v)-based soilless medium. Prior to that time, our mix was peat and perlite (7:3, v/v) and the change from perlite to rice hulls came after many trials showed that our crops grew just a well, if not better in some cases, when less costly rice hulls were used instead of perlite. Our crops, including grafted grapevines, rootstocks used for the production of almonds, walnuts, pistachio and other stone fruit trees all performed up to expectations in the peat and rice hull mix. However, there were some other cultural problems we were experiencing with such a light and airy mix.

1) It was difficult to get the proper compaction of medium in our pots without some physical interaction during the pot-filling process.
2) Significant settling of the medium in the pots after planting required labor to refill the pots. We thought this to be a necessary step to allow more volume of soil for greater root development.
3) It was difficult to manage medium moisture in the bottom of the pot which was causing some root rot issue in our *Citrus* crops.

Coir, could it alleviate our problems?

1. Citrus.

In 2014, we began to run extensive trials on the crops we were growing. First, we needed to address the root rot issues in our citrus crops. We had already been given some suggestions of how much coir to add to our current mix by a citrus grower from Spain who had experience with coir. We began adding coir at 30% and 50% (v/v) to our standard peat and rice hull (4:1, v/v) mix and measuring plant growth. At different times during the plant's growth after transplanting, we measured plant height, stem caliper, and foliage and root dry weights. We also made observational notes on root health, overall plant growth and development. Additionally we were interested in how the mixes physically held up in the containers. The end result showed that a coir, peat, and rice hull (5:4:1, by vol.) mix provided the best results.

2. Bench grafted grapevines.

Budbreak vs. no budbreak.

After our initial success with the addition of coir to our potting mix for citrus, we began to look at other crops we were growing in the standard mix of peat moss and rice hulls. We grow large numbers (8 million in 2015) of bench grafted grapevines of *Vitis vinifera* on various rootstocks of *Vitis* spp. for the wine industry in California and the Pacific Northwest. Again we had some distinct challenges that we were interested in seeing if coir could help to improve our production.

Much of our propagation material for the production of grapevine transplants is sourced from vines that we farm and sell the grapes to wineries in California. I am of the opinion that the farming techniques for growing quality grapes for making wine is not always the best for producing quality propagation material! Because of this and the fact that we produce many scion/rootstock combinations throughout the production season, we inherently are challenged with a phenomenon that we term budbreak vs. no budbreak! Briefly, what that means is that the scion portion of the grafted vines begins to break out of dormancy during the callusing process, while others do not. The importance of this "budbreak" is used to determine the probable success of that particular grafted lot and whether or not we will meet the projected need for that order or if it is necessary to graft more of those vines.

So, we were curious to see if coir would have any effect on the "budbreak/no budbreak" vines after being transplanted into a coir based medium. We know what works for citrus, but wanted to see what effect, if any; varying amounts of coir might have on grafted vines. Soil trials were designed with varying amounts of coir ranging from as little as 30% to as high as 80% in combination with peat and rice hulls and in some trials even 100% coir was investigated.

At planting, bundles containing 100 grafted vines were sorted into lots of those that broke bud in callusing and those that had yet to break bud and were potted into the various soil mixes. Data on plant height, stem caliper, root and foliage dry weights, and bud break counts were taken and recorded at various times during the production cycle.

Scion and rootstock combinations.

Because of the vast number of rootstock and scion combinations that are used by our customers, we wanted to look at some of those combinations that had been ordered to see how they performed in soil mixes containing various amounts of coir. For this particular trial, specific clones of *V. vinifera* and the rootstocks used were as follows:

- Pinot Noir 23/1103P (*V. berlandieri* × *V. rupestris*)
- Pinot Gris 04/Freedom (1613 (Solonis × Othello) × Dogridge
- Pinot Gris 04/Salt Creek (*V. candnicans* × *V. rupestris*)
- Chardonnay 76/101-14 MG (*V. riparia* × *V. rupestris*)
- Pinot Noir 2A/3309C (*V. riparia* × *V. rupestris*)

The media for this trial had two sizes of coir, one described as being ¼-in. size, the other as ¾-in. size. Amounts of coir were combined with our standard mix at rates of 70% or 80% (v/v) in addition to 100% coir and our standard mix. Average root dry weights were recorded on 8/20/14 at the end of the trial.

A third trial of a single scion/rootstock combination involving PG04/Salt Creek and potted into our standard mix containing the two different sized 1/4-in. and 3/4-in. coir at rates of 30, 50 and 70% coir (v/v) in addition to 100% coir and our standard mix was also observed. In this single combination, average height, caliper, foliar and root dry weights, and saleable plants were recorded.

Trends.

In most of the trials we conducted with grafted grapevines, coir had a positive impact on growth and development. One exception was in the budbreak vs. no budbreak trial, the vines that had already broken bud in callusing, did much better in our standard soil mix versus the vines potted into the 100% coir. In contrast, however, in the very same trial, the "no budbreak" vines after being potted into 100% coir actually had a higher percentage of buds to break out of dormancy and grow better than the "no budbreak" vines potted into our standard peat and rice hull (4:1, v/v) standard soil mix. This may have been due to the fact that we "buffered" the coir in the pots with a calcium nitrate solution drench immediately after planting and a second drench 7 days after the initial drench, whereas our standard mix (UTC) did not receive a calcium nitrate treatment.

In trials in which we looked at various scion/rootstock combinations of grafted vines, the results varied greatly as to the individual scion/rootstock combination and the amount of coir in the mix. The results were not all that surprising since each individual rootstock in the trial, had their own distinct characteristics and thus one might have expected them to react differently to the amounts of coir.

In the single scion/rootstock trial of PG04/Salt Creek, if a decision had to made strictly on the results of the number of saleable plants, then the ¾-in. 100% coir produced the most saleable plants, with the 50% and 100% ¼-in. close behind.

SO, WHERE DO WE GO FROM HERE?

Currently, because of the other crops we grow besides grapevines, we have transplanted substantial numbers of our other crops in the same coir, peat, and rice hull (5:4:1, by vol.) mix in which our citrus is grown. We will continue to observe the growth of our rootstocks used in the production of almonds, walnuts, and pistachios in this mix as compared to the standard peat and rice hull (4:1, v/v) mix and most likely continue to examine the growth of our crops in 100% coir!

We know coir is in our future as a major component of our soil mix based on its cost and what we've observed so far in trials. As to how much to use; only trials and time will tell!

ACKNOWLEDGEMENT

I want to thank the Horticultural Research Staff at Duarte Nursery for all their time in setting up trials and collecting data that was used in preparation of this manuscript.

Slow sand filters: a biological treatment method to remove plant pathogens from nursery runoff©

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Abstract

Slow sand filters (SSF) are an effective technology, capable of developing high-quality water from untreated sources including irrigation runoff. The sand serves as a substrate on which a microorganism community grows. This microbial community can breakdown a wide range of pollutants including plant pathogens. This report reviews results on the removal of *Phytophthora* spp., *Fusarium oxysporum,* and tobacco mosaic virus. We were interested in the capacity of these filters to remove different kinds of plant pathogens from captured irrigation run off. Our experiments removed *P. capsici* after the microbial community was established (2 weeks) and after a simulated 7-day pump failure in previously established SSFs. However, SSFs did not remove *F. oxysporum* after 7 weeks. In our tests, the SSFs were also able to remove tobacco mosaic virus from inoculated runoff water after 6 to 9 weeks of exposure.

INTRODUCTION

Captured runoff may contain plant pathogens and it is necessary to remove them prior to reuse for irrigation to prevent disease spread. Treatment can include chemical compounds, such as chlorine, radiation (from UV light), thermal (using heat from steam or other sources) and biological treatment methods such as slow sand filters (SSF). These filters have been use for a very long time, originally to produce drinking water and in the last decade or two are being used in horticultural production at an increasing rate.

Slow sand filters are a biological treatment method that is simple to set up, requiring little or no chemical or energy inputs. As its name implies, flow rates are slow. For each square foot of sand bed surface, 0.06 to 0.2 gal of water can be treated per min. So a round tank that is 12 ft in diameter can treat about 10,000 gal per day. Any container that holds sand and water can be used: steel water tanks, septic tanks, or earthen lined reservoirs. At the bottom of the container is a manifold of pipes to collect the treated water (Figure 1). The manifold is buried in coarse gravel to facilitate collection of the treated water. Above the gravel are several layers of sand, graduating from coarse at the bottom to fine at the top, to prevent the filtration sand from entering the gravel layer. Finally, at the top, is the bed of sand. A pump may be necessary to move the treated water into or out of the sand filter as it may not be possible to rely on gravity for both inflow and outflow. While filtration takes place particulates may clog the filter; this needs to be prevented with sedimentation ponds and other pre-filtering treatments.

The sand serves as a substrate on which a community of microorganisms grows and water should flow continuously through the sand bed for optimizing treatment volume. Key characteristics of slow sand filters include:

- Round grains, uniform size of about 0.3-0.6 mm. Uniformity is important to maintain water flow through the sand. Sharp sand can pack and restrict flow, so round grains are necessary
- One meter deep sand bed. Maintenance can require removal of a few centimeters of the top layer of sand. When 0.5 m of sand is left after many “cleanings”, the sand bed should be rebuilt.
- One meter deep water head above the sand. These filters are gravity driven, so the water head is needed to push the water through the bed.

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- Flow control. To obtain most efficient treatment, flow needs to be controlled to balance water quality and flow. Slow rates improve treatment, but reduce the volumes of water treated.
- Recommend two filters. While one filter is being serviced the other can remain in operation.

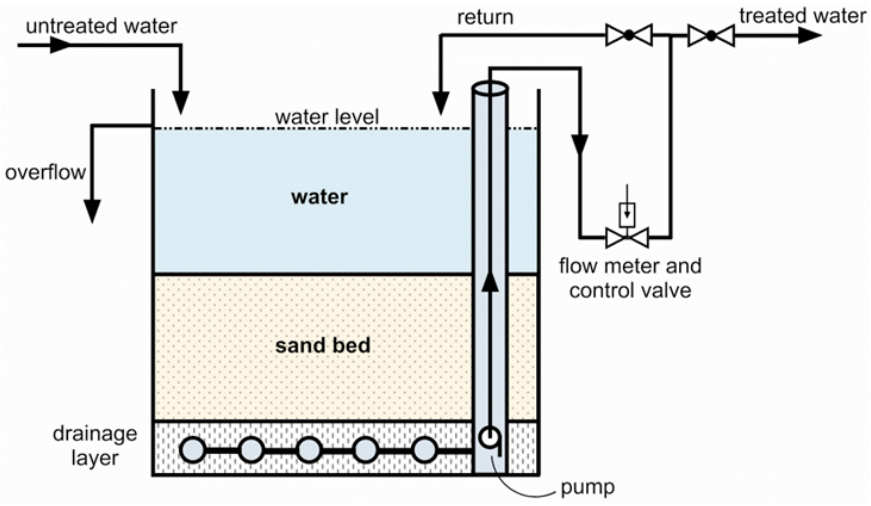

Figure 1. The slow sand filter (SSF) consists of a drainage layer that includes a pipe manifold assembly buried in pea gravel to collect treated water. Several layers of sand of gradually decreasing size cover the pea gravel so the SSF sand doesn't become incorporated in the gravel. The manifold is connected to a pump that moves the filtered water to storage. If topography enables it, the system may be entirely or partially gravity driven. Flow control is key for effective treatment.

To study how SSFs work in removing plant pathogens, three sets of experiments were conducted: (1) treatment performance, (2) pathogen switch, and (3) virus removal. The first set of experiments was used to determine the time required for treatment to occur. Although it is a biological treatment method, it is not necessary to inoculate the filters. Because the sand in a new filter is essentially sterile it takes time for the microorganism community to develop in response to the pollutants present. In the second set, it was not known if a sand filter established exposed to one pathogen can remove another type of pathogen if it suddenly appears. It was also not known if treatment would be compromised when water supply to a SSF system was shut down inadvertently, mimicking a pump failure. In the third experiment, since there was little information on the ability of SSFs to remove plant pathogenic viruses, work was done to assess this.

MATERIALS AND METHODS

Treatment performance

Slow sand filters were constructed using 4-in. PVC pipe that included sampling valves located just above the sand bed (unfiltered), at 20 cm intervals down the depth of the bed and below the sand bed (filtered) (Figure 2). Flow rates were set at 150, 250, and 500 L m^{-2} h^{-1}. The recommended flow rate of 150 L m^{-2} h^{-1} corresponded to a flow of 20 mL min^{-1} retrieved from the column. Irrigation runoff was generated by irrigating plants on a tray in a greenhouse (Harris and Oki, 2009). Captured runoff water was inoculated with *Phytophthora capsici* zoospores and then provided to the SSF columns. Water samples were collected every 5 days beginning on the day that water was introduced to the SSFs. The water samples were then analyzed for the presence of *P. capsici* colony forming units (CFUs) using culture media that selects for *Phytophthora* and *Pythium*.

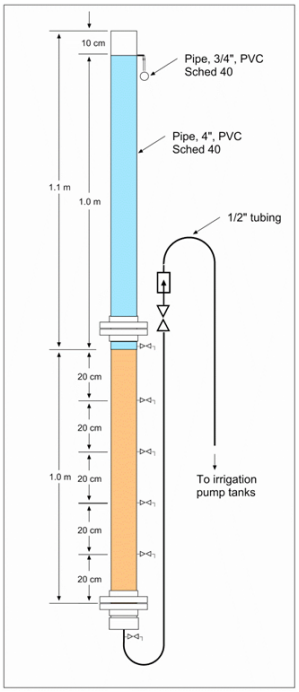

Figure 2. Slow sand filters for experimentation were constructed of 4-in. PVC pipe and included valves that enabled collecting samples above the sand bed, at 20-cm intervals down the sand bed, and below the bed.

Pathogen switch

Two sets of columns were set up, one set exposed to *P. capsici* inoculum in runoff water and the other exposed to *Fusarium oxysporum* f. sp. *lycopersici* for 6 weeks (Lee and Oki, 2013). Then, the pathogen inoculum for each treatment was switched (Figure 3). To simulate a system failure after 12 weeks, the pumps supplying water to the columns were turned off for 7 days, then restarted, and allowed to run for 6 more weeks. The entire experiment lasted a total of 19 weeks. Flow rates were set at 20 mL min^{-1}. Water samples were collected from above (unfiltered) and below (filtered) the sand bed and analyzed on culture plates to determine pathogen concentration and were also tested by bioassay on tomato and pepper plants to test for *F. oxysporum* and *P. capsici*, respectively.

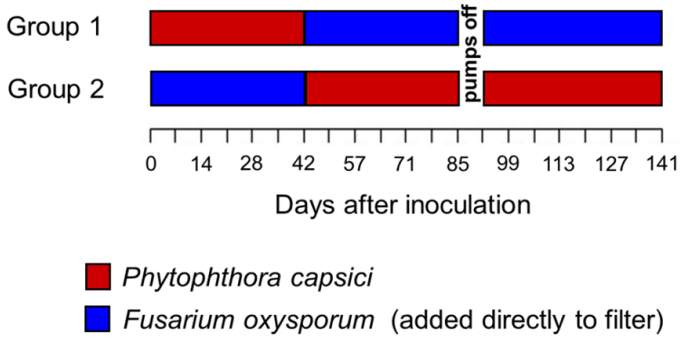

Figure 3. Two sets of slow sand filters (SSF) were set up. One set was initiailly exposed to *Phytophthora capsici* for 6 weeks then *Fusarium oxysporum*. The other SSF set was the opposite: initially exposed to *Fusarium* then *Phytophthora*. In addition, after 12 weeks the pumps were shut off for 7 days to simulate a pump failure.

Virus removal

Sand filters assembled in the same manner previously described were installed in the greenhouse at the UC South Coast Research and Extension Center in Irvine, California (Mathews et al. in preparation). A suspension of tobacco mosaic virus was introduced and mixed into the water above the sand beds. Samples of water were collected weekly from above and below the sand bed for 12 weeks and were analyzed by ELISA and bioassays. Bioassays used leaves of *Nicotiana glutinosa* and *Chenopodium quinoa* and whole plants of *N. tabacum* 'Turkish' and *N. benthamiana*. A pilot study utilized a single filter and a subsequent study involved three filters. The leaf assay for TMV was not conducted in the second study.

RESULTS

In all of the flow rate treatments in the first set of experiments, pathogen concentration declined steadily over time. Complete removal was apparent after 15 to 21 d and continued through the 30-d duration of the experiment (Figure 4).

In the pathogen switch study, during the first 6-week test period of the *P. capsici* was not detected in the samples collected at Week 2 and thereafter, but *F. oxysporum* was always recovered from the SSFs. When the pathogens were switched, those filters initially exposed to *F. oxysporum* were able to immediately remove *P. capsici* but *Fusarium* was always recovered from the SSFs. After the pumps were shut off for 7 d and then restarted, P. *capsici* was not detected, but *F. oxysporum* was recovered from treated water.

Although this is mainly an aerobic biological, interrupting water flow for 7 d did not diminish the ability of the filters to remove *P. capsici* when water flow was resumed. So it appears that these systems are resilient when experiencing flow interruptions caused by pump failures, for example, as long as the biological layer does not dry out.

In the virus removal pilot study that utilized a single filter, TMV was not detected in the samples collected at Week 9 and later. In the subsequent experiment using three filters, the virus was not detected in samples collected from Week 6 and later.

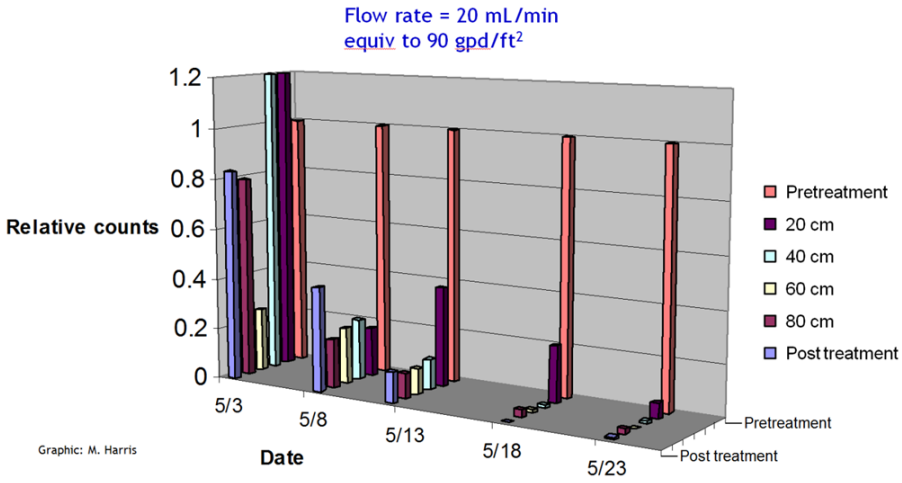

Figure 4. Recovery of pathogens measured as CFUs. Counts are relative to the pretreatment amounts. Removal occurred after 15 d in this experiment conducted in May. In the cooler fall season, treatment took 21 d to appear.

DISCUSSION

All of the treatment flows resulted in removal of *P. capsici* after about 14-21 d. But it was difficult to maintain the desired flow with SSFs running at the highest flow rate. Although pathogen removal is possible at greater than recommended flow rates, more frequent maintenance is necessary. Further study is needed to determine if the maintenance frequency is a factor of either the volume of water treated or the time interval between maintenance events.

The pathogen switch study showed us how these filters perform in the removal of a pathogen when the microorganism community is allowed to develope when exposed to another. The selection of the pathogens was based on their cellular composition. Specifically, the cell walls of *Phytophthora* are composed of β1,3 glucans, whereas those of *Fusarium* are of chitin, so it was posited that the microorganism communities that mitigated each of the pathogens would be distinct. Although the filters were not able to remove *F. oxysporum*, they were immediately able to remove *P. capsici* when it was introduced, but the opposite was not the case. This may suggest that organisms that can mitigate *P. capsici* are also present in the treatment of *F. oxysporum*, but organisms that can remove *P. capsici* may not be involved in *F. oxysporum* removal. Since *F. oxysporum* wasn't removed in this study, this is only speculation.

The experiments inoculating SSF with TMV are the first demonstrations of the removal of this virus using slow sand filters. Since TMV is so robust, there is a very high probability that most other plant pathogenic viruses can also be removed from captured irrigation runoff.

CONCLUSIONS

Slow sand filters are effective in removing a wide range of plant pathogens from water molds to viruses. As with other reports, we weren't able to remove *Fusarium* from the water. However, there are other reports indicating that removal can be attained after a long exposure or with pretreatment using chitin. Although these systems have high initial costs and can consume a large area, there are no other chemical or energy inputs required other than lifting the water into the filter and periodic cleaning.

ACKNOWLEDGEMENTS

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Landscape plant irrigation trials©

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Abstract

With its large population and Mediterranean climate, California's water supply is a valuable, but limited, resource that has been made even more apparent during the current multi-year drought. About half of the water consumed in residences is provided as irrigation to landscapes. In the past, plants used in landscapes were chosen only for their ornamental value, but recently more consideration is also given to their water needs. To contribute to information on plant water use, an ongoing study at the University of California, Davis developed irrigation requirements. Plants were installed in a field in the fall of the year and provided ample amounts of water during the first summer. During the second summer, from April to October, four irrigation treatments at 20, 40, 60 and 80% of reference evapotranspiration (ET_0) (CIMIS) were provided to the plants. Evaluations of plant size, appearance, and other quality parameters were measured each month. Recommended irrigation rates were developed from the evaluations and reported to funding sources and posted online.

INTRODUCTION

California is one of five Mediterranean climates zones in the world characterized by cool, wet winters and hot, dry summers. This means that most urban landscapes require irrigation during the summer. But since water is a precious and limited resource, a fact that has been made even more apparent during the current multi-year drought, there have been mandates limiting the amounts of water that can be applied to these landscapes. Even though the first State legislation requiring more attention to urban landscapes planning was passed in 1990 (Clute, 1990), further legislation was necessary to emphasize the need for conservation (Laird, 2004, 2006). The latest legislation (Laird, 2006) led to the Model Water Efficient Landscape Ordinance (MWELO) that requires an estimated annual water use for the new or renovated landscape. This calculation requires information on plant specific water use (species coefficients, K_S), similar to that used in crop water use estimations, to develop landscape water use coefficients (K_L) and refers directly to the Water Use Classifications of Landscape Species (WUCOLS) document (Costello, 2014).

This document includes information on only about 3,500 plants, which is the largest assembly of this type of information, but still leaves a void of data on many thousands of other plants that could be used. The method used to compile the information in WUCOLS was the convening of horticultural experts in six different climate zones in California. These committees evaluated plants on a list and agreed on a category describing their water use in those zones. These categories were aligned with a percentage of reference evapotranspiration (ET_0), which is essentially a species coefficient (K_S) (Table 1) that can be used to calculate water needs.

The project discussed here developed information based on a scientifically based replicated experimental field set up where plants were first established and then exposed to four irrigation treatments. Physical measurements and quality assessments were made every month and a group assessment was conducted at the end of the study period. Recommendations for irrigation of these plants were derived from these measurements (Reid and Oki, 2008).

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Table 1. Categories of landscape plant water needs. Plants were placed into one or several of these categories based on knowledge of the plants by local horticultural experts. Irrigation needs of the plants were rated relative to reference evapotranspiration (ET_0), that is the amount of water needed by a well irrigated cool season turf in that region.

Category	Percentage of ET_0
High	70-90
Moderate	40-60
Low	10-30
Very Low	<10

MATERIALS AND METHODS

Plants are selected for examination to be planted in either the field (Figure 1) or under 50% shade cloth on the UC Davis campus. Plants are arranged in rows 2 m apart and within the rows also 2 m apart. Rows are covered with 3-4 in. of mulch. Each row is provided four water lines so that any treatment can be delivered to each plant. Two 2 gph emitters are provided to each plant.

Plants selected for evaluation are planted in the field or shade house in October. Plants are allowed to establish during the first year after planting with regular irrigation during the summer. Establishment period irrigations of 8.3 gal per plant are applied at about weekly intervals, depending on weather. This volume of water replaces 50% of the water holding capacity (WHC) of a cylinder of the Yolo silty clay loam soil of 1 m diameter and 0.5 m deep.

Deficit irrigation treatments are 20, 40, 60, and 80% of ET_0 and are applied during the second growing season after planting from April through September. The recommended irrigation rate for cool season turf is 80% of ET_0 and 60% of ET_0 is the recommended irrigation rate for warm season turf, as a reference. Both treatments and planting arrangements are designed in randomized complete block designs.

Figure 1. Plants are randomly organized and spaced 2 m in rows 2 m apart. Irrigation lines provide one of the 20, 40, 60, 80% of ET_0 treatments to each plant during the second summer after planting.

ET_0 is measured continuously at a nearby CIMIS (California Irrigation Management Information System) weather station located on campus and is checked on a daily basis. ET_0 is multiplied by the treatment factor (0.2, 0.4, 0.6, or 0.8) and those values are accumulated separately for each treatment. When the accumulated value reaches the triggering level, irrigation is initiated. The level that triggers irrigation is equivalent to 50% of the soil water holding capacity of the root volume (assumed to be a cylinder 1 m in diameter with a depth of 1 m). The amount of water applied (16.6 gal plant^{-1}) is also equal to 50% of the soil water holding capacity. So, volumes are fixed and provide a deep irrigation, but the interval between irrigations varies depending on the treatment factor and the weather.

Each month, a plant growth index (PGI) is determined to quantify the comparative growth of plants using the formula [(l+w)/2+h]/2, where *l*, *w*, and *h* represent length, width, and height of the plant (Figure 2). Height is measured from the ground to the tallest leaf. Length and width are measured along the row (in a north-south direction) and across the row (in an east-west direction), respectively, using the outermost leaf in each direction. The means of plant growth indices for each treatment and species are calculated and graphed as both a change in the value of the PGI over time, and as a PGI relative to the starting PGI. Aesthetic ratings for foliage quality, flower quantity, vigor, health and overall appearance rating (OAR) on a 1-5 scale (1: very poor, 5: excellent) are assessed each month.

Near the end of the study, local horticulturists are invited to view and rate the performance of the plants (Figure 3) and that data is integrated with the monthly measurements to develop irrigation recommendations.

Figure 2. Project manager Jared Sisneroz and graduate student Zhou Yang measure and evaluate plants.

Figure 3. Each fall, local horticulturists are invited to evaluate the plants in the irrigation trials. The data is used to develop recommended irrigation rates for each plant species/cultivar in the trial. See the list at: http://ccuh.ucdavis.edu/academia/plant-trials.

RESULTS

All of the data is used to develop a recommended irrigation rate (K_S) and is reported to funding agencies, plant providers, and posted as the "Compendium of Results, Landscape Trials" on the UC Davis California Center for Urban Horticulture (CCUH) website. This list includes the following information for each of the plants tested:

- Botanical Name and the plant patent designation (if applicable).
- Common Name and any marketing names a plant may be listed for sale as in a nursery, i.e. *Lonicera periclymenum* 'Inov 86', will be marketed as *Lonicera periclymenum* 'Peaches and Cream'.
- OAR (overall appearance rating) is the mean of the monthly overall appearance ratings for the recommended irrigation treatment for each species during the deficit period.
- Recommended Treatment ET% corresponds to the treatment where a species performed the highest in our field trial.
- Suggested Irrigation Frequency provides watering guidelines for each species when planted as part of a landscape, when using these guidelines for irrigation scheduling it is important to water deeply, fully saturating the root zone and use a thick layer of mulch.
- Year Trialed is when a selected species was trialed.

For more information about a species please visit: ccuh.ucdavis/academia/plant-trials. This site includes the list described above, reports for funding sources, and descriptions of the work conducted.

CONCLUSIONS

To date, there have been 48 full-sun and 14 shade plants tested. Results are reported as a downloadable PDF file that contains a summary of results of all of the plants tested. More complete reports that include detailed information including: discussion of performance, data analyses, images of the plants, and other issues such as pests, diseases, or other notables are also posted at the CCUH website.

This information can be used by landscape architects and designers who will need to calculate an estimated annual water use by the landscape as required by California regulations. WUCOLS is named in the regulation as a source for landscape plant water use, but the list contains "only" about 3,500 plants. The project described here adds information on plants not in the WUCOLS list using science-based methods. The process is slow, taking two years, but is an effective way to continue adding to this needed information.

ACKNOWLEDGEMENTS

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Overcoming the challenges of endogenous contamination in micropropagation of fruit and nut trees©

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Abstract

Micropropagation has become a successful technique for fruit and nut tree rootstocks. However, success of a laboratory to produce a high rate of healthy shoots during multiplication stage depends on its ability to maintain cultures free from contamination. Although, all labs can easily start a clean culture from an explant, many labs report a flare-up of bacterial contamination after a few cycles of multiplication despite using their best laboratory practices. This flare-up is often blamed on endogenous bacteria within the micro shoots. This assumes that such bacteria were always present in the shoots, but were in quiescent stage and/or were non-culturable and suddenly they became active and grew on culture media. Such theory is believable since presence of endogenous bacteria in plants is well-known in the literature and only 1% of bacteria are culturable. To overcome this challenge of flare-up of contamination (endogenous or introduced), a laboratory should have a protocol in place to index their stock materials on a regular basis. For culturable bacteria, contamination can be detected by culturing samples of tissue in a nutrient broth for bacteria. For non-culturable bacteria, sections of stems can be eluted in water and the eluate observed under the microscope for bacteria. Bacteria-specific PCR tests are now available and are helpful. These procedures along with shoot tip cultures, occasional use of antibiotic and close visual observation have proved successful at Micro Paradox in maintaining our nuclear stock of walnuts, pistachio, and peach × almond hybrids free from contamination.

INTRODUCTION

Micropropagation is becoming a popular technique for commercial production of fruit and nut tree rootstocks. At present seven laboratories in California alone are producing rootstocks of walnuts, pistachio, and peach × almond hybrids. One of the challenge that laboratories face is to keep the cultures free of contamination. Almost every laboratory has a horror story to tell that they could not produce enough quantity of certain rootstocks due to flare up of bacterial contamination in their cultures. In some cases, contamination destroys all cultures and the laboratory has to start over from new explants from mother plants.

All laboratories are familiar with surface sterilization procedures and are able to establish cultures free of bacterial and fungal contamination. The mother plant is often indexed for common viruses and it is assumed that viruses and other fastidious organisms are absent in the mother plant and that one has to only worry about bacterial and fungal organisms. Fungi usually grow on tissue culture media and contaminated cultures can be easily discarded. Some of the culturable bacteria can also be easily seen on the tissue culture media and infected cultures are discarded. A challenge emerges when cultures start declining after a few generations of vigorous growth in the absence of any visible contamination. Often, it is blamed on endogenous bacterial contamination. It is assumed that some bacteria were always present in the cultures, but they were in a quiescent stage and suddenly they become actively growing in culture vessels and are responsible for the decline of cultures (Figure 1). This theory is credible as the presence of endogenous bacteria in plant

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tissue is well supported in the literature and the fact that only 1% of the bacteria are normally culturable. For commercial labs, this theory is easier to believe because they follow all good lab practices, believe that contamination is not introduced from environment and that it must be endogenous. However, a proof of introduced contamination is lacking. One should keep in mind that it is common to have 2-5% visible fungal contamination in growth rooms and that invisible bacterial introduction is highly possible.

Figure 1. Decline in pistachio plants in vitro due to endogenous bacterial contamination.

For this article, we have assumed that contamination could originate from endogenous bacteria in the stock or from introduction of non-culturable bacteria during handling and growing of plants in the lab. We have focused on detection of such contamination and management practices to keep bacteria away from tissue culture stock. We have successfully incorporated some of these techniques in our commercial program of walnut and pistachio rootstock production at Micro Paradox laboratory.

DETECTION TECHNIQUES

The following techniques are available. Each has some advantages and disadvantages.

Visual observation

This technique is the gold standard and is usually practiced in all tissue culture labs. The cultures are closely observed for visible contamination and any contaminated plants/ entire containers are discarded. A flash light can be helpful to see infections that are otherwise hard to see with the naked eye. A major limitation of this visual method is that non-culturable endogenous bacteria will go undetected.

Culture indexing

At Micro Paradox, this technique is routinely used. Samples of cultures are submitted to our in-house laboratory for bacterial counts. In this test, plants are crushed in sterile water to extract bacteria from the tissue and the extract plated onto general purpose bacterial media (KB, PDA, 523, etc). Recovery of bacteria from samples indicates infection by culturable bacteria. This test is very simple and effective. Multiple media should be used because each medium is selective for certain bacteria. Unculturable bacteria are not detected by this technique.

Bright field/phase contrast microscopy

In this method, stem tissue is sharply cut in a drop of water using a scalpel. Bacteria will ooze out from the tissue into the water within a few seconds. A slide (wet mount) is prepared and examined at 400X using a transmission microscope by a trained lab technician. At these magnifications several bacteria that vary in size from 1-3 micrometers can be seen as small particles. Many bacteria fall in this size and include *Pseudomonas*, *Xanthomonas*,

Erwinia, *Clavibacter*, *Bacillus*, and many others. Some highly motile bacteria are easily seen. If bacteria are suspected, then Gram staining can be done and examined further at 1000X magnifications. Microscopy works well if there is a large amount of bacteria in the tissue and enough will ooze out. Phase contrast microscopy is more effective (makes bacteria darker) than bright field microscopy.

Electron microscopy

Samples are submitted to a special facility equipped with an electron microscope. A specialist is needed to prepare and observe samples. Bacteria, phytoplasma, and viruses can be easily observed. The size and shape of the organisms also provides some clues on general identification of organisms. Species identification is not possible as several organisms have a similar shape. This technique is extremely helpful and is often used to identify unknown diseases in field samples. However, there are few facilities equipped with an electron microscope and testing is expensive for routine use.

Polymerase chain reaction (PCR)

Recently polymerase chain reaction (PCR) tests have become available for several organisms. Universal primers that detect all bacteria are also available and amplify 1500 nucleotide base pairs. The samples of tissue culture material are submitted to a PCR lab. At Micro Paradox, we routinely submit samples to our in-house lab (CSP Labs) for universal PCR. Unfortunately, primers available today also react with chloroplast and mitochondria of some plants and result in a false PCR product. Therefore, if a PCR product is obtained from a sample, it must be sequenced. The sequence is then compared with GenBank database to confirm if it matches to a bacterium. A complete match of 1500 base pairs to a bacterium is highly reliable. A trained molecular biologist is required to conclude results of sequencing and to rule out any false results.

Next generation sequencing (future)

In this test, all DNA and RNA sequences in a sample are obtained and analyzed. The sequences that do not belong to plants are further analyzed if they match to microorganisms (bacteria, viruses, etc). By this technique, new pathogens in plants have been discovered. A classic example is discovery of red blotch virus in grapevines. At this time, only a few labs are capable of conducting this test and the test is very expensive. It is not routinely used today, but has great promise to detect and identify endogenous bacteria in the future.

MANAGEMENT OF ENDOGENOUS CONTAMINATION

There are several things that can be done to overcome issues of "so called" endogenous contamination in a commercial tissue culture lab.

Culture establishment

This is the most significant step to detect and exclude endogenous contamination. New cultures should be closely watched for contamination. Any cultures where bacterial streaking into the media from callus can be seen must be discarded. One must not attempt to save upper nodes and tips to save the cultures. Callus is most likely to have more concentration of bacteria if there is an infection. Callus samples can be tested by culture indexing. If cultures stay clean during a few transfers, they are likely to be free from any culturable bacteria. If vigor is high during these few transfers, they are likely to be free from non-culturable harmful bacteria. If cultures start declining after several generations, one should look at reasons other than endogenous bacteria arising from initial starting material.

Air flow in the tissue culture facility

Tissue culture laboratory design is important to keep contamination away from entering from outside. Clean areas (transfer hood area, media pouring area, and growth rooms) should be under positive pressure of HEPA-filtered air. Other rooms (worker entry, autoclave room, receiving area and any R&D areas should be under negative air pressure).

Air flow should be gentle that it does not create turbulence.

Good lab practices

Good lab practices are a part of continuous training of workers. Every lab has their own way to implement these practices and they work well if technicians and managers believe in the practices. These include dress code, hand sanitation, work surface sanitation, floor cleaning, and trash removal. Only authorized people should be allowed in clean work areas. Check lists can be used to audit compliance to good lab practices. Good lab practices will reduce workers introducing bacteria especially non-culturable bacteria that can go unnoticed.

On-site management

Many managers record a lot of data to associate general contamination to specific workers. Such contamination analysis requires a few weeks of observation and data analysis before pointing out to the technician for corrections. A proactive approach is to have strong, on-site management. A crew leader should closely supervise laminar flow hood workers and provide on-site corrections and certify technicians. More supervision is needed for the new workers during their first 2-3 weeks. At Micro Paradox, we have concluded that there is no significant relationship between a worker and degree of contamination if workers have been fully trained and supervised. Contamination should not be related to a worker if crew leader is happy with performance of the worker. If contamination rates remain higher in spite of good work by workers, a manager should look for reasons of such contamination other than workers.

Lab testing

Samples of tissue culture stock should be tested on a routine basis for contamination by methods such as culture index, microscopy and PCR. If contamination is detected, infected materials can be quarantined and not used in the multiplication program.

Nuclear program

Establishment of nuclear stock is very important. Establishment of new cultures from mother trees is considered nuclear stock. Since this stock is used for the multiplication phase, it must stay free from contamination. For maintenance, nuclear stock goes through unlimited generations until new materials are again introduced from the mother tree. Therefore, the chance of contamination from endogenous bacteria or introduced bacteria are most likely to be seen in the nuclear stock. For this reason, nuclear stock should be clearly marked, handled by most experienced technicians and frequently tested. It is also good idea to treat nuclear stock with an antibiotic on an annual basis as a preventive measure for contamination control. Finally, use of tips of cuttings to maintain nuclear stock avoids any contamination that could have been introduced recently and not have yet travelled to the tip of the cutting. By use of these practices, Micro Paradox has not seen any contamination for the last 5 years in its nuclear stock.

One-way multiplication system

Micropropagation when compared to conventional propagation provides a one-way system where propagation always starts from clean nuclear stock. Any infections are flushed out as materials are released from the laboratory. However, in practice, cuttings from the multiplication stage may be further multiplied for a few cycles. This can result in the spread of endogenous bacteria in the entire stock in the multiplication phase. To prevent this spread of endogenous bacteria, each generation in the multiplication should be tracked as it is moving to the next multiplication generation. For example, the first generation (M1) should be labeled as second generation (M2) upon transfer. By this method, any contamination will have limited distribution and not spread to the entire stock.

Removal of contaminated containers from the growth rooms

Some contamination (2-5%), usually fungal in nature, develops in growth rooms in spite of best practices followed. This contamination can increase significantly within a growth room if contaminated containers are not discarded quickly. Any contaminated containers should be carefully removed from the growth room, bagged and discarded as soon as possible. A common mistake done in the labs is to rescue uncontaminated plants in a container to another container. This involves opening a container in a laminar flow hood that increases chances for the contaminant to be airborne and contaminate other containers.

CONCLUSION

Controlling endogenous or introduced contamination can be a real challenge in a tissue culture laboratory. Above described detection and management techniques can be very helpful in overcoming this challenge. These techniques have worked well to eliminate or quarantine contamination at Micro Paradox.

Monitoring pathogens and preventative control programs at a nursery producing container-grown plants©

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INTRODUCTION

Like any living organism, plants are susceptible to infection by environmental pathogens at all stages of development. The environmental conditions that nurseries must maintain to achieve plant growth coincide with the conditions necessary for pathogen growth and development. Although the chemical treatment of plants after a pathogen has infected its tissues is appropriate, the prevention of that initial pathogen/host interaction is more important to the long-term health and production of nursery plants. Thus, it is essential that nurseries have, in place, a system of prophylactic measures and monitoring that is designed to minimize this interaction. Such a system must be multi-layered and adaptable. By employing multiple prophylactic measures followed by close monitoring and laboratory testing of plants for potential pathogens, a high degree of success can be achieved.

ENSURING THE CLEANLINESS OF NEW TISSUE CULTURE INTRODUCTIONS

At Duarte nursery, our clean-plant protocol begins with every new plant species or clone we wish to introduce into propagation. These new introductions are put through a three-step virus and phytoplasma elimination protocol. This process begins with the harvest of apical meristems from new plants that are then cultured over a 1-week period. These meristem cultures then undergo thermotherapy, during which they are exposed to the highest temperature that the plant cells can tolerate and still grow. The high temperature thermotherapy lasts for 6 weeks. This will destroy the heat labile viruses only. However, it will also cause most other viruses to stop replicating and restrict the spread of viruses to the newly developed meristematic cells. The new meristematic tissue is then harvested and put through cryotherapy. During this process, meristematic cells are first dehydrated to reduce ice nucleation within the cells. They are then quick frozen in liquid nitrogen to kill any remaining viruses or phytoplasmas. The cells are then rehydrated to preserve the cells and then the surviving meristematic tissue is cultured to produce new plantlets.

PRODUCTION OF PATHOGEN-FREE MOTHER BLOCKS

The origin of plant source material and its previous exposure to pathogens is usually difficult to ascertain with certainty. However, it is not impossible. All properties utilized for our mother blocks were purchased as either uncultivated rangeland or farmland that was not growing the species we intended on planting. In theory, this limits the new plants exposure to a species specific pathogen. All plants designated to be used in our mother blocks start as micropropagated, “pathogen-free”, clones. These micropropagated clones are tested and are only used if they are free of pathogens at the time of planting. This helps reduce the potential pathogen load on a mother block, but does not prohibit the influx of pathogens in the future. For that reason, all scion and rootstock material is screened before the time of harvest and quality tested when it arrives at the nursery.

UTILIZATION OF TISSUE CULTURE TECHNOLOGY FOR PLANT PROPAGATION

At Duarte Nursery, tissue culture technology is used, not only to produce clonal mother stock, but is also used for propagating our fruit and nut trees. The process we use for establishing plants in vitro is multifaceted. First, the cuttings are excised from mother stock

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or other known pathogen-free sources and surface-sterilized with a dilute bleach solution and 70% alcohol to kill any pathogens that may be on the cuttings. These cuttings are then introduced to the proper medium and placed into culture. When suitable growth has occurred, sterile cuttings are made from the cultured mother stock and are then placed in the right nutrient medium. Plants produced from sterile cuttings are grown on sterile artificial media, under sterile growing conditions and controlled environments to multiply the number of stock plants and establish the line in vitro. The nutrient medium the plants are grown on contains sufficient nutrients to support the plant for about 4-6 weeks. At the end of this time the plants undergo the multiplication phase of our tissue culture process. During the first and each successive multiplication step, enough cuttings are taken from each plant to double or triple the amount of plant material in culture. We continue this multiplication until a critical mass of between 20,000-50,000 plants is achieved. This number of plants allows us to take the plants into the commercial production and rooting phase. At this point the extra cuttings from each successive multiplication are siphoned off and placed on an auxin-containing rooting medium to become a production run. Finally, plants cultured for production are grown and callused in the laboratory, then extracted from the rooting medium and planted in plug trays to develop roots. When sufficiently rooted and acclimated to the greenhouse environment, the rooted plugs are transplanted into pots containing a soilless medium. Four separate plant lines are created from each new introduction. Production run lots are kept segregated according to these plant lines throughout the production cycle. This ensures that if any somaclonal variation occurs, the line exhibiting an off-type can be omitted from the production run in the future and replaced by another genetically superior line. Additionally, all original lines are re-introduced every 4-5 yr to reduce the line's exposure to epigenetic effects.

TREATMENT AND SANITATION OF ROOTSTOCK AND BUDWOOD FROM THE FIELD

Grape rootstock and scion cuttings harvested from our mother blocks are all hot water treated to kill overwintering grape vine mealy bugs, if present. We have adapted this hot water treatment system to also clean the wood of any epiphytic fungal spores and bacteria. We accomplish this by filtering and recycling the water in the three hot water baths. We originally trialed commercial pool filters in 2013 and found them to be lacking (Figure 1). Fungal and bacterial contaminates were continually present in the water being recirculated into the bath after filtering. In 2014, we upgraded to industrial sand filters, filled with small pore size glass filtering media and added industrial UV filters to the water line. Testing indicated that the entire microorganism load on the wood was actively filtered out by the upgraded filtration system (Figure 2).

Figure 1. (A) Old pool filtering apparatus used to filter contaminates from the sanitation tanks used to clean rootstock and budwood from the field; (B) Culture plate from tank water containing fungal and bacterial contaminates prior to filtration with the old apparatus; and (C) Culture plate containing tank water contaminates after an 8-h day of filtration utilizing the old apparatus.

Figure 2. (A) New industrial filtering apparatus used to clean tank water; (B) Culture plate containing fungal and bacterial tank water contaminates prior to filtration with the new apparatus; and (C) Culture plate containing tank water contaminates after an 8-h day of filtration utilizing the new apparatus.

SEASONING ROOM SANITATION WITH OZONE

The simple movement of plant material in the external environment can expose that material to potential air-borne pathogens. Therefore, we continue to treat rootstock and scion budwood both during and after processing to inhibit pathogen inoculation. Grape wood that is retrieved from cold storage for seasoning is treated prophylactically in the seasoning room with ozone. Ozone is produced by a corona-discharge ozone generator and pumped into the room by a series of manifolds and jet fans to produce an even dispersal throughout the room. The efficacy of the treatment is periodically tested and has been found to be efficient in the prevention of bacterial and fungal development on the seasoning wood (Figure 3). In addition to the nightly treatment of the room with ozone, the entire room is sterilized every week with bleach.

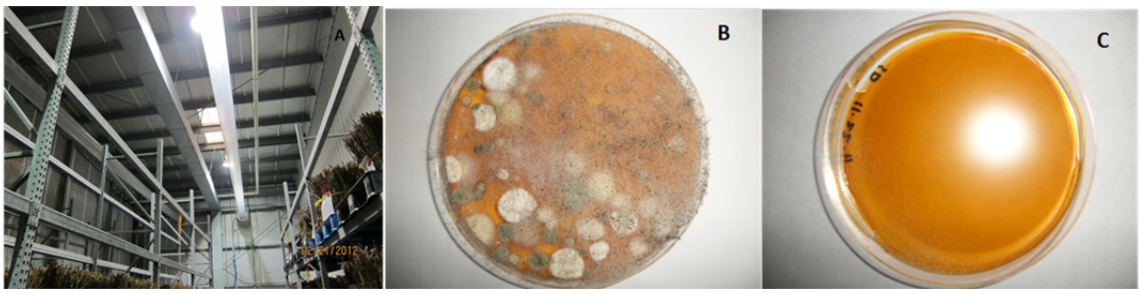


Figure 3. (A) Seasoning room with jet fan for distributing ozone. (B) A culture plate left open in the room for 15 h, without ozone. (C) The same test was done while ozone was actively being pumped through the air into the room.

GRAFTING SANITATION

The cuts made to the rootstock and budwood during grafting provide a direct route for pathogen inoculation into the plant. The sanitation procedures used during grafting are aimed at preventing possible infections. The procedure includes the flame sterilization of both clippers and grafting blades every 30 min during the grafting cycle, disinfecting the entire room with chlorine foam and changing out the polyethylene covering the grafting tables every 2 weeks. Additionally, the pallets and flats used to transport the wood are sterilized with bleach at the start of each day.

CALLUSING WATER FILTRATION AND ULTRAVIOLET AIR SANITATION

After grafting, our grape vines are placed in hydroponic callusing baths. The water used to fill the baths is filtered by the same industrial designed filters and in-line UV water sanitation used in the hot water treatment area (Figure 2A). In addition, the air ducting that supplies outside air to the room is equipped with UV radiation emitting lights to clean the air

coming into the callusing area. Testing of the callusing water filtering system has proven that it is capable of removing fungal spores and bacteria from the water supply feeding the baths (Figure 4). Additionally, ultraviolet irradiation of the incoming air is able to kill any air borne fungal spores (Figure 5). These mechanisms aid in protecting our newly grafted grape vines from potential bacterial and fungal pathogens.

Figure 4. (A) Water inflow to callusing filtration system showing high levels of bacterial contamination; (B) Water outflow from the filtering system to the callusing baths with no biologicals present.

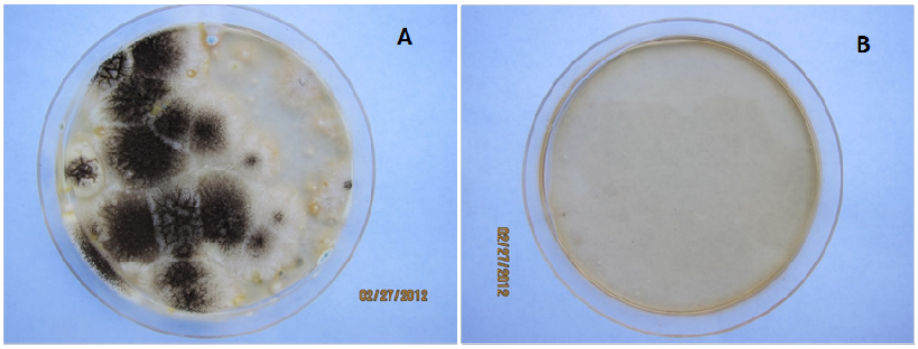


Figure 5. (A) Culture plate exposed to air inflow to the callusing room for 5 h without ultra-violet radiation; (B) Analogous culture plate exposed to ultra violet radiated air inflow for 5 h.

CONTAINERIZED PLANT PRODUCTION IN THE GREENHOUSE

Every plant produced at Duarte Nursery is grown in a container from propagation, through greenhouse growth and until the point of delivery to our customer. The containerization of our plants gives them several benefits with regard to their potential exposure to pathogens. Containerized plants never interface with the soil until they are planted in a customer's field. Since the plant has never been planted in the earth, this minimizes the plants exposure to soil-borne pathogens and nematodes. By being grown in a container, the product is planted with a complete intact root system (Figure 6). This reduces transplant stress, encourages fast establishment and reduces root-wound exposure to crown gall. Using the containerized plant platform also allows us the ability to screen our soilless potting mix periodically for pathogens and add beneficial organisms like mycorrhizae to the mix to increase pathogen resistance to root and soil pathogens.

Figure 6. Fully developed root system on a containerized grafted grape vine.

FACILITY RETROFITS TO IMPROVE SANITATION

The facilities that are used to produce our plants also play a role in the overall sanitation of our nursery. Facilities can either aid or hinder sanitation and pathogen prevention efforts. For example, wooden benches and dirt floors, provide good environments for both bacterial and fungal pathogens to colonize. These older structures, once infected, are very difficult to sanitize. To that end, Duarte Nursery has embarked on a multi-year capital expansion program in which older wood-constructed greenhouses have been removed and 18 acres of new steel-constructed, concrete-floored greenhouses have been built (Figure 7). In addition to the new indoor areas, 35 acres of new outdoor growing space has been added with steel bench construction over concrete pads. At the end of each production cycle excess soil and plant material is collected and discarded and benches and floors are washed thoroughly. After cleaning, benches and floors are treated with oxidizing chlorine foam to kill any pathogens before the next crop occupies that space (Figure 8).

Figure 7. (A) Old type greenhouse with wooden benches over dirt floor; (B) Modernized greenhouse; and (C) modernized outdoor growing area, both constructed with steel benches and concrete floors to minimize pathogen exposure to the plants.

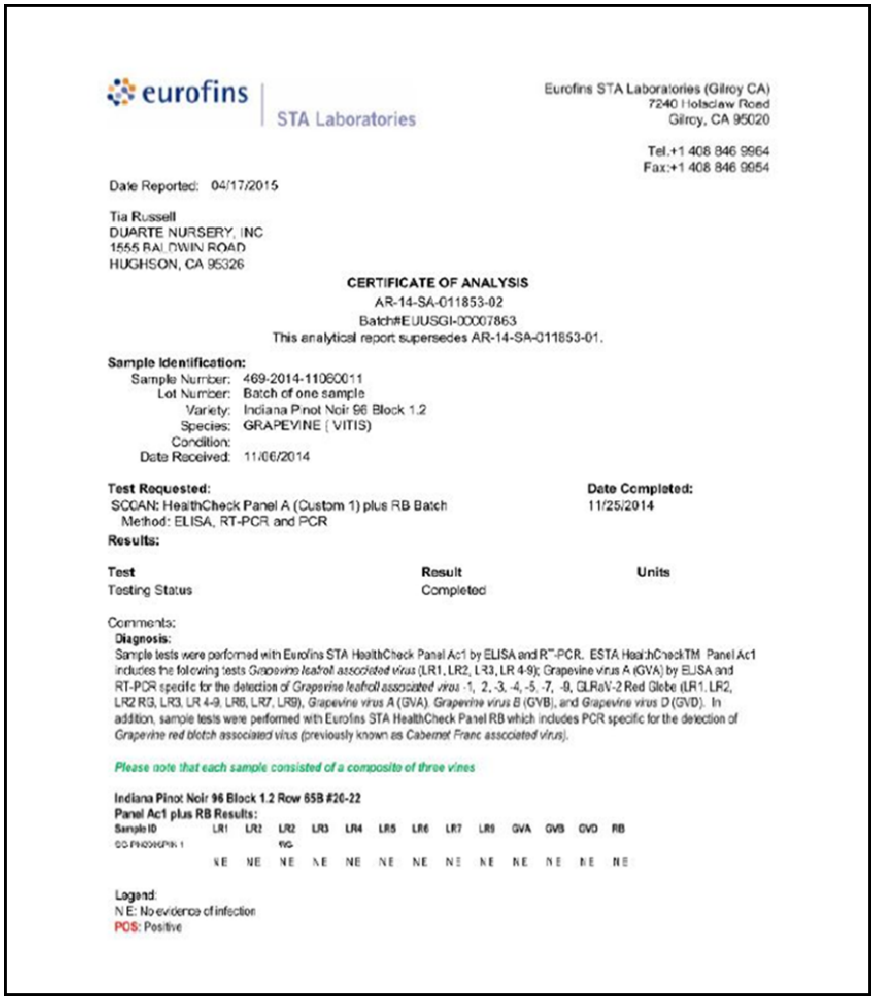
Figure 8. Using chloride foam to sanitize greenhouse and outdoor surfaces used for plant production.

CONTINUED TESTING AND SCREENING OF PLANT PRODUCTS

The prophylactic measures employed by any nursery, no matter how precise or controlled, only limit the ability of pathogens to infect plants. Therefore, programs designed to monitor, test and screen plant material coming into the nursery, plants growing at the nursery and plants ready for sale is essential. Duarte Nursery rootstock and budwood mother blocks are routinely tested every year by state employees through the voluntary CDFA Certification Program. In addition, Duarte Nursery's Quality Assurance Department, samples and screens all Duarte-owned mother blocks and any non-Duarte budwood from outside sources through ELISA and PCR testing at private laboratories (Figure 9). While it is truly impossible to test each individual plant prior to sale, having a thorough screening protocol in place can minimize the number of plants that slip through at the point of sale. At Duarte, we go further than a simple visual screening just prior to shipment. Here, plants are periodically screened for bacterial and fungal pathogens at the tissue culture, acclimation and growth stages of development. This screening process entails plant sampling, sample processing, bacterial and fungal culture, and colorimetric identification of potential pathogens via a GEN III OmniLog Microbial Identification System (BIOLOG, Inc., Hayward, California) (Figure10). Finally, each plant is visually inspected by our shipping department before they are transported to our customers.

SUMMARY

Due to the environmental conditions nurseries must maintain to be productive, pathogen growth and development is inevitable. However, nurseries can minimize their plants' exposure to pathogens through the use of preventive measures implemented in a multi-layered, adaptable sanitation system. Any such system must be combined with a developed integrated pest management program and an appropriate plant monitoring, screening and testing program that ensures a high number of pathogen-free plants reach the customer.



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CERTIFICATE OF ANALYSIS
AR-14-SA-011853-02
Batch#EUUSGI-0C007863
This analytical report supersedes AR-14-SA-011853-01.

Sample Identification:
Sample Number: 469-2014-11060011
Lot Number: Batch of one sample
Variety: Indiana Pinot Noir 96 Block 1.2
Species: GRAPEVINE (VITIS)
Condition:
Date Received: 11/06/2014

Test Requested:
SCGAN: HealthCheck Panel A (Custom 1) plus RB Batch
Method: ELISA, RT-PCR and PCR

Date Completed:
11/25/2014

Results:

Test	Result	Units
Testing Status	Completed	

Comments:
Diagnosis:
Sample tests were performed with Eurofins STA HealthCheck Panel Ac1 by ELISA and R*-PCR. ESTA HealthCheckTM Panel Ac1 includes the following tests *Grapevine leafroll associated virus* (LR1, LR2, LR3, LR 4-9); Grapevine virus A (GVA) by ELISA and RT-PCR specific for the detection of *Grapevine leafroll associated virus* -1, -2, -3, -4, -5, -7, -9, GLRaV-2 Red Globe (LR1, LR2, LR2 R3, LR3, LR 4-9, LR6, LR7, LR9), *Grapevine virus A* (GVA), *Grapevine virus B* (GVB), and *Grapevine virus D* (GVD). In addition, sample tests were performed with Eurofins STA HealthCheck Panel RB which includes PCR specific for the detection of *Grapevine red blotch associated virus (previously known as Cabernet Franc associated virus).*

Please note that each sample consisted of a composite of three vines

Indiana Pinot Noir 96 Block 1.2 Row 65B #20-22
Panel Ac1 plus RB Results:

Sample ID	LR1	LR2	LR2 R3	LR3	LR4	LR5	LR6	LR7	LR9	GVA	GVB	GVD	RB
00-PH000PIK-1	NE	NE	NE	NE	NE	NE	NE	NE	NE	NE	NE	NE	NE

Legend:
N E: No evidence of infection
POS: Positive

Figure 9. Example of test results from ELISA and PCR analysis of grapevine budwood.

Figure 10. (A)Plated samples ready for preparation, (B) sample preparation for colorimetric assay, (C) GEN III OmniLog system, and (D) identification via colorimetric assay showing different color patterns for four different bacteria.

Alternatives to pesticides in controlling pests and diseases©

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Abstract

Many California consumers and government agency regulators increasingly demand agricultural products produced with less, or without, use of synthetic chemical pesticides. As a prime example, soil fumigation with synthetic chemical toxicants now is seen as having decreasing compatibility with public safety and environmental quality. Alternatives are being developed and such methods must be shown to be effective, predictable, and economically viable. Active heat-based treatments are attractive options for soil disinfestation and certain elements of the biogeochemical environment, such as accumulation of passive solar energy and knowledge-based utilization of organic materials and byproducts, may be harnessed to provide economic pest management. Recent research and implementation projects on alternatives including biosolarization, biofumigation, and anaerobic soil disinfestation (ASD) will be discussed.

OVERVIEW

The movement in California agriculture away from use of synthetic chemical soil fumigants is well-known and documented. Over the past 30 years, materials including ethylene dibromide (EDB), dibromochloropropane (DBCP), methyl bromide, and others, have been taken from the marketplace by regulatory action (Stapleton et al., 2000). In addition, newly developed fumigant products touted as useful replacement materials, such as methyl iodide, have yet to pass regulatory scrutiny. At the same time, consumer sentiment toward certified organic agricultural products has skyrocketed. Most large supermarket chains in California now feature sections of certified organic products, both fresh and processed. Rightly or wrongly, synthetic pesticides are seen by large segments of society as being undesirable or harmful and having decreasing compatibility with public safety and environmental quality. As anti-soil fumigant sentiments have grown, interest and economic stimulus in developing alternatives to synthetic soil fumigants has increased accordingly. The arena of soilborne pest management (and in this discussion we will consider disease-causing organisms as pests) has expanded to include not only implementation of fumigant alternatives, but the broader and integrated concept of agricultural soil health stewardship. Apart from soilborne pest management, the integrated concept of soil health requires a focus on optimizing fertility and soil biotic community factors (Simmons et al., 2014).

SYNTHETIC CHEMICAL SOIL FUMIGANT ALTERNATIVES

Planning for soil fumigant alternatives

In considering use of fumigant alternatives, asking a few preliminary questions can assist in the planning process. Obviously, knowledge of field and cropping history is very important. A follow-up question relates to the choice of broad-spectrum or narrow-range strategies. Is elimination of a few specific pests (e.g., weeds) the objective, versus a range of problematic weeds, nematodes, and soil fungi? Or, perhaps even control of unknown/unidentified soilborne pest agents is desired, in order to maximize crop yield? Once these questions are answered, all possible soil treatment options can be evaluated.

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Soil fumigant alternative options

For certain pest problems, specific biological and/or cultural approaches may be sufficient. Such approaches are favored by some agriculturists, and may include inoculation of soil, seeds, or plants with various amendments, inoculants, or competitors. If available, use of crop cultivars with genetic resistance may be used. However, if organic certification is desired, genetically engineered propagative material currently will not be allowed. Some soilborne pests can be effectively controlled by cultural adjustments, such as proper water/nutrient management (e.g., *Phytophthora* diseases), modifying planting date to optimize soil temperature (e.g., certain nematodes), tillage modifications, crop rotation, and/or cover cropping.

Example: sweet potato hot beds, Merced Country, California

In terms of replacing soil fumigant usage, perhaps the least radical option, for non-organic producers, is simply using the above-mentioned questions to replace fumigation with a "softer" synthetic pesticide. One example is that of Stoddard et al. (2011) in Merced County, California. They found that production of sweet potato slips in hotbeds, which were traditionally fumigated with methyl bromide, could be safely done with only an herbicide application. The unique production conditions of early-spring slips, sprouting from healthy mother tubers in cool soil, were seen as prohibitive to activity and development of prevalent nematode and fungal pests.

Another non-pesticide approach to soil disinfestation, which also contributes to overall soil health, is the use of cover crops, green manures, teas, composts, biofumigants (e.g., *Brassica* spp.) and other soil amendments (Stapleton et al., 2000). Depending on the agroecological conditions (e.g. physical, chemical, biological) present in the treated area, as well as the attributes of targeted pest organisms, these practices may or may not provide rapid and effective control of soilborne pests.

Physical soil disinfestation

For fumigant-like biocidal activity in soil, the physical methods of disinfestation may be most useful. Active soil heating, such as with steam, can provide drastic reductions of soil biota; however, this approach is both difficult and expensive to conduct and is used only in very small areas. More recently, two physical methods, passive solar heating of soil by solarization (Stapleton et al. 2000; Dahlquist et al., 2007; Marshall et al., 2013) and deliberate causation of anaerobic conditions in soil by anaerobic soil disinfestation (ASD) (Butler et al., 2011), have received considerable attention for fumigant-like activity. Both of these methods also have some limitations.

"Double-tent" solarization — small soil volumes

The State of California (CDFA) allows a specific protocol of solarization of soil, using a "double-tent" tarping setup (Stapleton et al., 2002), to disinfest soil for containerized nursery production (CDFA, 2009). However, this method is mainly useful for small and/or seasonal operations since solarization is dependent upon atmospheric conditions and high air temperatures. According to CDFA regulations, the "double-tent" protocol dictates treatment "until temperature of all soil reaches a minimum of 158°F (70°C) that is maintained for at least 30 continuous min, or a minimum of 140°F (60°C) that is maintained for at least 60 continuous min. Soil must be either in polyethylene planting bags or in piles not more than 12 in. high. Soil in piles must be placed on a layer of polyethylene film, concrete pad, or other material, that will not allow reinfestation of soil and covered by a sheet of clear polyethylene film. An additional layer of clear polyethylene film must be suspended over the first layer to create a still air chamber over the soil to be treated. Soil moisture content must be near field capacity. Soil temperature at the bottom center of the pile or bag must be monitored" (CDFA, 2009).

Biosolarization and anaerobic soil disinfestation (ASD)

Recent developments in deploying various combinations of plastic film-covered soil, organic materials, moisture, and heat have provided some promising directions in the future of soil disinfestation. These integrated soil treatments can provide more effective and predictable pest management options for operations not wishing to use synthetic chemical products. Although research and implementation efforts are well-underway, the precise effects and modes of action of biosolarization and ASD have yet to be fully elucidated (Butler et al., 2011; Simmons et al., 2013, 2014). Both approaches use various tarped applications of organic residues and/or composts to produce naturally-occurring, biocidal conditions in soil. The ASD process emphasizes reductive and fermentative conditions to inactivate pests, while biosolarization efforts are focused on aerobic processes leading to effective soil disinfestation (Simmons et al., 2013). Further clarification of pesticidal activity mechanisms may be expected in the near future.

ACKNOWLEDGEMENTS

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Development of novel plant phenotypes using plant pigment-associated genes©

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Abstract

A number of flavonoids produced by plants impart specific flower and fruit color. The R2R3-Myb transcription factors are key regulatory genes involved in flavonoid biosynthesis. Such transcription factors can be potentially used in the development of new plant phenotypes via genetic engineering. In the current study, anthocyanin biosynthesis-related genes from *Citrus* (RUBY), grapevine (*VvMybA1*), and maize(leaf color-LC) were isolated and placed along with a NPTII gene under the control of a CaMV35S-derived promoter complex. Embryogenic cultures of *Vitis vinifera* 'Thompson Seedless' were initiated from leaves and floral explants. Somatic embryos at the mid-cotyledonary stage of development were co-cultivated with *Agrobacterium* harboring individual candidate genes to regenerate modified plants. Leaf discs of tobacco cultivar 'Samsun' and petunia cultivar 'Mitcham' were also transformed to produce modified plants. Regenerated plants were transferred to potting mix, hardened under conditions of high humidity and transferred to a greenhouse. Transient anthocyanin expression from various genes was evidenced by bright red spots on explants after 3-5 d of co-cultivation with *Agrobacterium.* Stable gene expression was observed in callus and shoot cultures after 4-8 weeks on regeneration medium. Modified 'Thompson Seedless' plants were recovered after 16 weeks of co-cultivation while 'Samson' and 'Mitcham' produced plants in 4-6 weeks. Regenerated plants exhibited varied patterns and intensity of red pigmentation in mature tissues. While some plant lines exhibited uniform red pigmentation on leaves and shoots, other lines exhibited patchy or interveinal accumulation of the anthocyanin pigment. Normal growth and flowering was observed in all plants. Such plants expressing anthocyanin pigments with varied patterns and intensities could be used as breeding lines for the development of ornamental phenotypes with unique coloration.

INTRODUCTION

Anthocyanins belong to a group of flavonoid compounds that are responsible for a wide array of colors in leaves, flowers and fruit, and function to attract pollinators and seed dispersers (Sakuta, 2014). Anthocyanin pigments also exhibit medicinal properties to mitigate effects of several debilitating diseases such as cancer and cardiovascular disease (de Pascual-Teresa and Sanchez Ballesta, 2008). Anthocyanins are produced from phenylalanine via the phenylpropanoid and flavonoid pathways. The regulation of anthocyanin biosynthesis is regulated by several proteins including the R2R3-Myb transcription factors, basis-helix-loop-helix (bHLH) and WD-repeat (WDR) proteins (Czemmel et al., 2012). The Myb transcription factors are large family of proteins that have key functions in the regulation of the anthocyanin biosynthesis pathway. Several regulatory proteins have been isolated from a number of plant species and expressed in heterologous systems for the constitutive expression of anthocyanin and production of red pigmentation in plant tissues. Anthocyanin-related genes can be potentially used to generate novel colors in plants resulting in the development of new ornamental cultivars. In the current study,

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anthocyanin biosynthesis-related genes were isolated from *Citrus*, *Vitis* and *Zea mays* and inserted in *Vitis vinifera*, *Petunia hybrida* and *Nicotiana tabacum* to produce novel phenotypes of potential value to the ornamental plant industry.

MATERIALS AND METHODS

Plant materials

Embryogenic cultures of *V. vinifera* 'Thompson Seedless' were established as described previously (Dhekney et al., 2009). Briefly, unopened leaves obtained from in vitro micropropagation cultures were placed on NB2 medium (Dhekney et al., 2009) and cultured were placed in the dark for 6-8 weeks. Resulting callus cultures were transferred to light (65 μM m^{-1} s^{-1}) with a 16 h photoperiod at 25°C. Embryogenic callus occurring after 10 weeks of culture was transferred to X6 medium (Dhekney et al., 2009) for the development of somatic embryos. Somatic embryos (SE) at the mid-cotyledonary stage of development were used for gene insertion and plant regeneration.

Seed material for tobacco cultivar Samsun and petunia cultivar Mitcham were surface-sterilized in 50% commercial bleach solution followed by three washes in sterile distilled water. Seeds were then transferred to MS medium and maintained at 25°C and 16-h photoperiod as described above. Leaf discs were obtained from 21-day-old seedlings and used in gene insertion studies.

Vector construction

The coding sequences of the *Citrus* RUBY, *Vitis* MybA1, and maize LC (leaf color) genes were placed along with a npt II gene under the control of a CaMV35S-derived promoter complex. Each gene cassette was placed into a binary vector, which was subsequently transferred to *Agrobacterium* strain 'EHA 105' using the freeze-thaw method (Burow et al., 1990).

Gene insertion and culture development

Agrobacterium cultures harboring the RUBY, MybA1 and LC genes respectively were grown overnight on a rotary shaker at 28°C in MG/L medium (Garfinkle and Nester, 1980). The overnight culture was then centrifuged at 6000 rpm for 8 min to obtain a bacterial pellet. The pellet was resuspended in X2 medium (Li et al., 2006) and grown for an additional 4 h prior to gene insertion experiments.

Co-cultivation of in vitro cultures

Grapevine somatic embryos at the mid-cotyledonary stage of development, and petunia and tobacco leaf discs were used as explants for gene insertion experiments. Explants were transferred to sterile Petri dishes and submerged in *Agrobacterium* solution for 8 minutes. The excess bacterial solution was then removed by pipetting and explants were transferred to a Petri dishes containing two layers of filter paper (Fisherbrand P8) soaked in liquid DM medium (Li et al., 2001). Explants were co-cultivated in the dark for 3 days at 28°C. After 3 days, grapevine somatic embryos were transferred to liquid DMcc medium (DM medium containing 200 mg L^{-1} each of carbenicillin and cefotaxime antibiotics) and transferred to a rotary shaker at 120 rpm to inhibit bacterial growth. After two days of washing in liquid DMcc medium, explants were transferred to Petri dishes containing solid DMcck medium (DM medium containing 200 mg L^{-1} each of carbenicillin and cefotaxime and 100 mg L^{-1} kanamycin) and incubated in the dark at 28°C for 30 days. Resulting callus cultures were then transferred to X6cck70 medium (Li et al., 2006) for development of secondary embryos. Petunia and tobacco explants were blotted on sterile filter paper to remove excess bacterial growth. Explants were then transferred to regeneration medium for shoot proliferation described previously (Dhekney et al., 2011; van der Meer, 2006).

Regeneration of modified plants

Secondary embryos expressing the RUBY, MybA1 and LC genes were identified on the

basis of red pigmentation. Such embryos were transferred to MS1B medium (Li et al., 2006) for embryo germination and plant regeneration.

Modified shoots identified on the basis of red pigmentation were excised from proliferating cultures and transferred to MS medium containing 200 mg L^{-1} each of carbenicillin and cefotaxime and 100 mg L^{-1} kanamycin for rooting. Fully developed plants were transferred to plug trays containing sterile potting mix and maintained under conditions of high humidity for hardening. Fully hardened plants were transferred to a greenhouse.

RESULTS AND DISCUSSION

Transient gene expression in explants was observed as bright red spots after 3 days of co-cultivation (Figure 1A). Transient gene expression decreased in intensity after 8 days of transfer to regeneration medium. Modified callus cultures were evidenced by bright red pigmentation (Figure 1B). Petunia and tobacco explants exhibited multiple shoot proliferation following transfer to regeneration medium Figure 1C) and shoots appeared dark red in color compared to non-transformed proliferating shoot cultures (Figure 1D). Shoot production in petunia and tobacco occurs via indirect organogenesis where co-cultivated leaf discs exhibit callus formation followed by the production of meristemoids. Such meristemoids eventually produce shoots, which may be modified following gene insertion. Modified shoots evidenced by bright red pigmentation produced roots following transfer to MS medium. Similar results were observed with grapevine somatic embryo explants where modified callus cultures produced somatic embryos when transferred to development medium. Secondary embryogenesis is frequently observed in grapevine cultures with secondary embryos arising from surface epidermal cells of the primary embryos (Dhekney et al., 2009; Gray et al., 2005). Thus, grapevine somatic embryos are ideal targets for gene insertion and recovery of stable modified plants via the process of secondary embryogenesis. Plant lines with varying levels and patterns of anthocyanin were observed in the greenhouse (Figure 1E, F). This may be attributed to various factors including copy numbers of inserted genes and their location of insertion in the plant genome. Grapevine plant and tobacco plants exhibiting intense anthocyanin pigmentation showed reduced growth and lack of vigor compared to those with low to moderate levels of anthocyanin pigmentation. This may be attributed to the hyperaccumulation of anthocyanin in the cytoplasm that may cause unintended toxic effects on plant growth and development (Marrs et al., 1995; Marrs, 1996). Varied patterns of anthocyanin pigmentation were observed on the abaxial and adaxial surfaces of plant leaves. Normal flowering was observed in tobacco plant lines expressing the RUBY, MybA1 and LC genes (Figure 1E). The pattern and intensity of floral pigmentation varied among various plant lines.

We are currently studying growth and development of grape and petunia plant lines in the greenhouse. The effects of the constitutive expression of anthocyanin related genes and subsequent anthocyanin pigment accumulation on reproductive development including fruit development and seed viability will be evident as plants mature. Such plants expressing anthocyanin pigments with varied patterns and intensities could be used as breeding lines for the development of ornamental phenotypes with unique coloration. Additionally, they could also serve as a source of red pigment production that could have potential applications in the food industry.

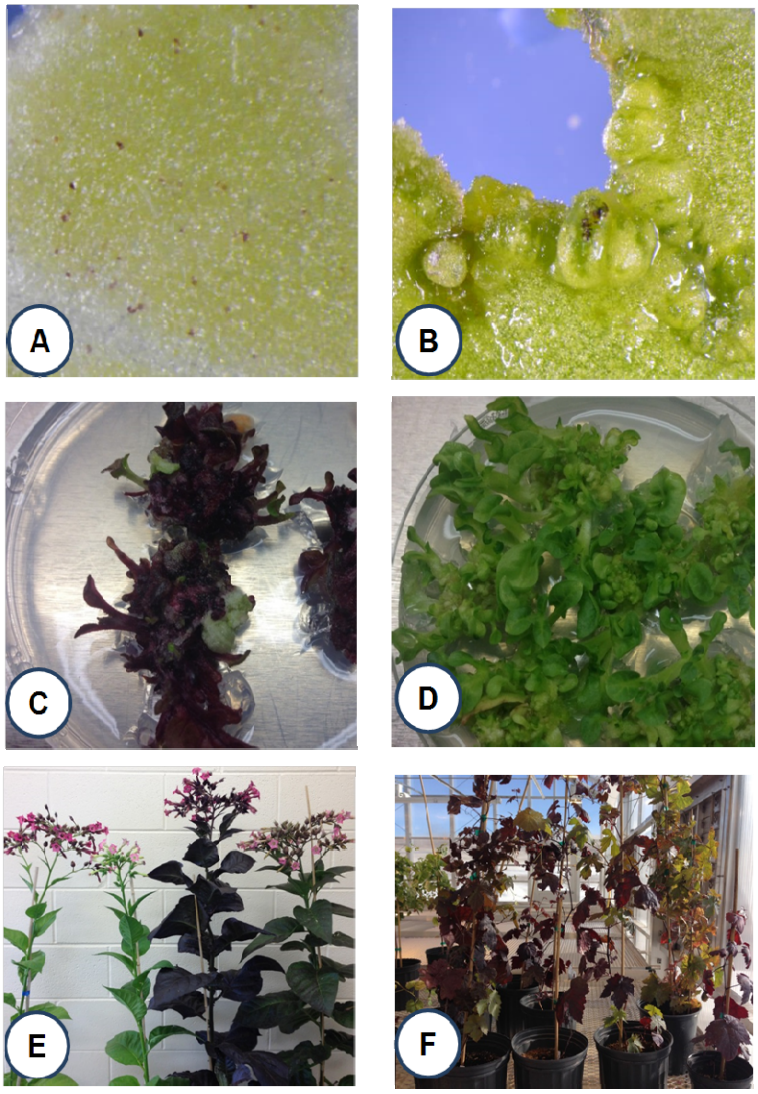

Figure 1. Transient anthocyanin expression (A) in tobacco leaf discs after 3 days of co-cultivation. Stable callus (B) and shoot (C) production on regeneration medium. Note that shoot cultures exhibit dark pigmentation compared to non-transformed control cultures (D). Mature tobacco (E) and grapevine plants (F) exhibiting varied levels and patterns of anthocyanin pigmentation.

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Instant bonsai: successfully rooting very woody grapevine cuttings©

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At the beginning of March, 2014, I was given access to some rather old grape vines that were going to be removed for road work on Highway 152 just west of Gilroy, California. After digging down for 2 ft. before finding any roots at all, I realized I would not be able to dig and move them by hand. I consulted with a friend who had been creating bonsai for many years and, with his advice, I decided to try rooting the picturesque growth at the tips of the vines that had been created by 40 years of pruning for wine grape production.

After pruning off all but two or three leaf nodes from the previous year's growth, I used a bow saw with a new blade to cut off sections with growing tips, from 8 to 10 in. long for the smaller pieces and 12 to 18 in. long for the larger ones. The diameter of the cuts ranged from 2 to 3 in. I dusted the cuts with a little rooting hormone (probably Hormex #3) then stuck these cuttings into qt- or gal-sized containers. I used my regular potting soil lightened with extra perlite. The cuttings were so top-heavy I had to wedge assorted rocks between the trunk and the sides of the pots to keep them upright.

The containers went into a 40% shade house and were kept moist, not wet. Within a month, most of the vines had started to leaf out. Finally, at the end of May I couldn't wait any longer and tipped one of the plants out of the pot. I was delighted to find that it was well-rooted.

All in all, I had about an 85-90% success rate. Some of the plants took considerably longer to root, closer to 4 or even 5 months. I think the success rate was high because I made the cuttings just as the leaf buds were starting to swell. I have spoken with others who tried similar techniques, but in the autumn, and had little to no success rooting their cuttings.

Figure 1. Gallon-sized container, 14 in. above soil, 2.5 years after cutting.

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New plant session: Western region©

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Ribes sanguineum 'Oregon Snowflake'

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Ruellia simplex 'Mayan Pink' (PPP)
Ruellia simplex 'Mayan Purple' (U.S. Patent PP24,422)
Ruellia simplex 'Mayan White' (U.S. Patent PP25,156)

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Lagerstroemia 'Miss Francis'
Lagerstroemia 'Miss Gail'
Lagerstroemia 'Miss Sandra'

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Ilex crenata × *I. maximowicziana* 'RutHol1' Emerald Colonnade® holly PP23,905
Rhaphiolepis umbellata 'RutRhaph1' Summer Moon® Indian hawthorn PP20,730

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Weigela 'Slingco 2,' Maroon Swoon™ weigela (PPAF, CPBRAF)
Weigela 'Velda', Tuxedo™ weigela (PPAF, CPBRAF)

NEW PLANTS

***Ilex crenata* × *I. maximowicziana* 'RutHol1', Emerald Colonnade® holly PP23,905**

Emerald Colonnade® holly is the result of hybridization program at The University of Georgia in an attempt to get a faster growing, upright form of small-leaved holly. This plant is a cross between *I. crenata* 'Sky Pencil' and a male form of *I. maximowicziana*. The resulting plant is faster growing than its female parent (24 months from cutting to a finished #5), is resistant to spider mites and is female sterile so invasiveness is not an issue. Both parents and Emerald Colonnade® have survived -5°F with no foliar or stem damage. Mature size

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after 10 years is 8 ft by 8 ft. Emerald Colonnade® holly is useful for hedging where larger plants are not needed or for topiary work since it is easily shaped and grows vigorously (Figure 1).

Figure 1. *Ilex crenata* × *I. maximowicziana* 'RutHol1', Emerald Colonnade® holly.

***Lagerstroemia* 'Miss Frances' (CM224)**

'Miss Frances' has a round, spreading growth habit with approximate dimensions of 5.5 m (18 ft) tall and 6 m (20 ft) wide after 9 years growing under ambient field conditions in south Mississippi. Crown branching is vigorous and dense with good foliage cover. Good leaf retention has been observed from spring through fall. Flowers are red (RHS Red 46A) and flower panicles average 16 cm (6.4 in.) in length and 8 cm (3.2 in.) in width on the terminal ends of branches. Plants flower from late June into August in south Mississippi. 'Miss Frances' displays a high level of field resistance to "rabbit tracks", bacterial leaf spot and powdery mildew, with moderate resistance to *Cercospora* leaf spot. Disease resistance is combined with other desirable horticultural traits including a large growth habit (5 to 7 m), dark red flowers over an extended bloom season and attractive persistent green foliage. Plants are more vigorous than many dark red-flowered cultivars such as its male parent, 'Arapaho', and its female parent, 'Gamad I' (Figure 2).

Figure 2. *Lagerstroemia* 'Miss Frances'.

***Lagerstroemia* 'Miss Gail' (CM223)**

'Miss Gail' has an upright, tight, vase-shaped growth habit with approximate dimensions of 6.5 m (21.5 ft) high and 3 m (10 ft) wide at 9 years of age under ambient field conditions in south Mississippi. Plants develop thick crown branching with good foliage cover of large dark-green leaves. Foliage retention is excellent from spring through fall. Inflorescences average 14 cm (5.6 in.) in length and 8 cm (3.2 in.) in width on the terminal ends of branches. Flowers are colored dark-purple (RHS Purple Violet N80A). Yellow

stamens contrast nicely with the purple petal color. Flowering occurs from late June into August. 'Miss Gail' displays a high level of field resistance to *Cercospora* leaf spot, powdery mildew, and "rabbit tracks" and moderate resistance to bacterial spot. In addition to disease resistance, 'Miss Gail' has a combination of other desirable horticultural traits including a large growth habit [7 m (23 ft)], dark-purple flowers over an extended bloom season and attractive persistent green foliage. 'Miss Gail' resulted from a cross-pollination between 'Catawba' as the female parent and 'Arapaho' as the male parent (Figure 3).

Figure 3. *Lagerstroemia* 'Miss Gail'.

***Lagerstroemia* 'Miss Sandra' (CM078)**

'Miss Sandra' has an upright spreading growth habit with approximate dimensions of 6 m (20 ft) high and 3 m (10 ft) wide at 9 years of age in south Mississippi under ambient field conditions. Plants develop thick crown branching with good foliage cover. Foliage retention is excellent throughout the summer. Flowering occurs from late June into August. Inflorescences average 14 cm (5.6 in.) in length and 7 cm (2.8 in.) in width on the terminal ends of branches. Flowers are dark-purple (RHS Purple Violet N81A) and measure 4 cm (1.6 in.) in width. Petals are fan-shaped with a ruffled apex and ruffled margins. 'Miss Sandra' displays a high level of field resistance to bacterial spot, powdery mildew, *Cercospora* leaf spot, and "rabbit tracks", combined with other desirable horticultural traits including a large growth habit [6 to 8 m (20 to 26.5 ft)], dark-purple flowers over an extended bloom season and attractive persistent green foliage. 'Miss Sandra' resulted from a cross-pollination between an unregistered, purple-flowered *L. indica* seedling collected in San Antonio, Texas as the female parent and 'Tonto' as the male parent (Figure 4).

Figure 4. *Lagerstroemia* 'Miss Sandra'.

***Rhaphiolepis umbellata* 'RutRhaph1' PP20,730 Summer Moon® Indian hawthorn**

Summer Moon® Indian hawthorn (Figure 5) was selected from a group of

approximately 1000 seedlings growing at Wight Nurseries in South Georgia in the late 1990s. It was selected for its excellent disease resistance to entomosporium leaf spot under nursery conditions and its wavy, dark-green, waxy foliage. Summer Moon® Indian hawthorn is ideal for the lower south and along the Pacific Ocean in USDA hardiness Zone 8. Plants in Athens, Georgia have survived 6°F with minimal leaf burn. This is a great landscape plant for foundation and/or mass plantings. After 10 years, the mature size is roughly 4 ft tall by 6 ft wide.

Figure 5. *Rhaphiolepis umbellata* 'RutRhaph1' Summer Moon® Indian hawthorn.

***Ribes sanguineum* 'Oregon Snowflake' PP26763**

'Oregon Snowflake' is a new and distinct cultivar of flowering currant being released as an alternative to White Icicle™ currant, the most popular white flowering cultivar in the trade. 'Oregon Snowflake' was selected for its dissected foliage and compact, mounded, and semi-dwarf growth habit that is novel and superior to other available cultivars. A morphological comparison of 'Oregon Snowflake' to White Icicle™ currant for leaf and growth habit characteristics has demonstrated the distinctness of 'Oregon Snowflake'. After 3 year, field-grown plants of 'Oregon Snowflake' were more than 30 cm shorter than White Icicle, but 15 cm wider, demonstrating its semi-dwarf, mounding habit (Figure 6). More details on this cultivar may be found by reading its release (Contreras and Friddle, 2015).

Figure 6. *Ribes sanguineum* 'Oregon Snowflake'.

***Ruellia simplex* 'Mayan Purple', 'Mayan White', and 'Mayan Pink' Mexican petunias**

Wild *Ruellia simplex* was introduced to Florida from Mexico in the 1940s and is now the third most important herbaceous perennial landscape plant in the state (after pentas

and lantana). However, *Ruellia simplex* is very fertile and has become invasive in natural areas in seven Southern USA states, Hawaii, Puerto Rico, and the Virgin Islands. The *Ruellia* breeding program at the University of Florida was started in 2007 with the objective of developing new, sterile cultivars with a range of flower colors and growth habits.

The first two sterile cultivars released at the University of Florida were *Ruellia* 'Mayan Purple' ('R10-102', U.S. Patent PP24,422) (Figure 7) and 'Mayan White' ('R10-108', U.S. Patent PP25,156) (Figure 8), which have great landscape performance and profuse flowering. 'Mayan Purple' has large purple flowers and a more full growth habit than 'Purple Showers', which is also sterile but grows very tall and tends to lodge. 'Mayan White' has large white flowers profuse flowering, and a fuller growth habit then 'Snow White'. These two cultivars are patented and unrooted cuttings are available from Horticultural Marketing Associates.

Figure 7. *Ruellia simplex* 'Mayan Purple'.

Figure 8. *Ruellia simplex* 'Mayan White'.

Ruellia 'Mayan Pink' ('R10-105Q54', PPP) has medium-sized pink flowers, has a compact growth habit and is shorter than 'Mayan Purple' and 'Mayan White'. In some environments it may produce a very few fruits, which usually abort before maturing. 'Mayan Pink' is a good replacement for the very fertile and invasive 'Chi Chi'. The patent for 'Mayan Pink' is pending and this cultivar will be commercially available later this year (Figure 9).

These *Ruellia* cultivars can also be grown in containers and we are working on developing blueprints to grow them using plant growth regulators.

Figure 9. *Ruellia simplex* 'Mayan Pink'.

***Weigela* 'Velda' (PPAF, CPBRAF) Tuxedo™ weigela**

Tuxedo™ weigela is the only dark-leaved, white-flowered weigela on the market (Figure 10). Plants add an upscale, refined tone to the landscape and are best grown in full sun so that the leaves will be dark green. Tuxedo™ weigela grows 2-3 ft tall and 3-4 ft wide and the plants can handle temperatures down to -34°C (Zone 4).

Figure 10. *Weigela* 'Velda', Tuxedo™ weigela.

***Weigela* 'Slingco 2' (PPAF, CPBRAF) Maroon Swoon™ weigela**

Maroon Swoon™ weigela is an improvement over 'Red Prince' weigela, with Maroon Swoon™ weigela exhibiting a deeper flower color, heavier flowering and a more compact growth habit (Figure 11). Plants may be grown in full or part-sun. Maroon Swoon™ weigela grows 4-5 ft tall and 2-3 ft wide and the plants can handle temperatures down to -34°C (Zone 4).

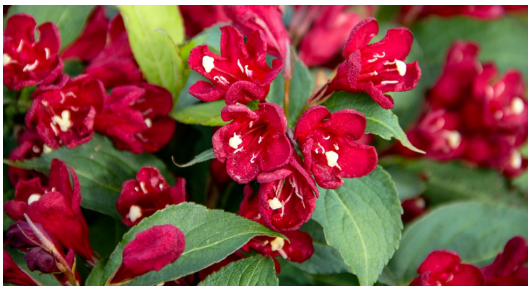

Figure 11. *Weigela* 'Slingco 2' Maroon Swoon™ weigela.

Literature cited

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All-America Selections winners for 2015: ornamentals and edibles with proven national and regional garden performance©

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INTRODUCTION

Seventeen selections became All-America Selections (AAS) National Award Winners for 2015. AAS includes a network of over 80 trial grounds all over North America where new, never-before-sold cultivars are "Tested Nationally and Proven Locally®" by skilled, impartial AAS Judges. Only the best performers are declared AAS Winners. Once these new cultivars are announced as AAS Winners, they are available for immediate sale and distribution.

An additional eight cultivars were selected as All-America Selections (AAS) Regional Award Winners for 2015. Regional winners undergo the same trialing process as national winners, but are recognized as cultivars that exhibit outstanding performance in specific regional climates.

AAS NATIONAL WINNERS FOR 2015

Allium tuberosum 'Geisha' (garlic chives)

'Geisha' is a vigorous grower with a pleasant garlic flavor. Wider, flatter and more refined leaves topped by pretty white flower stalks late in the season allow this edible crop to serve a dual purpose as an ornamental. Bred by Terra Organics.

Beta vulgaris 'Avalanche' (beet)

This beet exhibits a mild, sweet taste with a uniform root shape and no reddish tinge, making for more attractive produce. This beet is excellent for eating raw, without an earthy/beety taste, nor any bitter aftertaste. Bred by Bejo Seeds Inc.

Brassica oleracea var. *italica* 'Artwork' (F_1 broccoli)

This stem (or baby) broccoli was previously available only in gourmet markets and up-scale restaurants, but is now available for home gardeners. After harvest of the first crown, easy-to-harvest tender and tasty side shoots continue to appear long into the season. Plants resist bolting in warm weather better than other stem broccolis currently on the market. Bred by Seminis Vegetable Seeds.

Capsicum annuum 'Emerald Fire' (F_1 jalapeño pepper)

'Emerald Fire' produces attractive, glossy green peppers with thick walls that have very little cracking, even after maturing to red. Plants are high-yielding, with fruits rated at 2,500 Scoville units. Bred by Seminis Vegetable Seeds.

Capsicum annuum 'Flaming Flare' (F_1 chili pepper)

Unlike many Fresno chili peppers that typically grow better in warm and dry climates, 'Flaming Flare' performed well at all AAS trial sites. Fruits are sweeter tasting than similar Fresno chili peppers and consistently produce larger fruits and more peppers per plant. Bred by Seminis Vegetable Seeds.

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***Capsicum annuum* 'Pretty N Sweet' (F_1 ornamental pepper)**

Plants of this cultivar are multipurpose, producing multi-colored peppers that are both ornamental and edible. Fruits are sweet and are produced prolifically on 18-inch plants that may be grown in the ground or in containers. Bred by Seeds by Design.

***Cucurbita moschata* 'Butterscotch' (F_1 butternut squash)**

This attractive, small-fruited butternut squash has an exceptionally sweet taste and is perfect for just one or two servings. Compact vines are space-saving for smaller gardens and containers and are resistant to powdery mildew later in the season. Bred by Johnny's Selected Seeds.

***Cucurbita pepo* 'Bossa Nova' (F_1 zucchini squash)**

The beautiful dark and light-green mottled exterior of this zucchini is more pronounced than other cultivars on the market. Compact plants produce fruits earlier in the season and continue producing for three weeks longer than comparable cultivars. Bred by Seminis Vegetable Seeds.

***Dianthus* Jolt™ Pink (F_1 dianthus)**

This interspecific dianthus features large, showy flower heads with bright, pink, fringed flowers. This dianthus is heat tolerant, continues to flower through the season without setting seed and is ideal for low-maintenance gardens. Bred by PanAmerican Seed.

***Impatiens* 'Balboufink' Bounce™ Pink Flame impatiens**

This hybrid impatiens looks like an *I. walleriana* in habit, flower form, and flower count, but is downy mildew resistant. Plants produce bright pink bicolor flowers in sun or shade. This cultivar is available in plant form only. Bred by Selecta.

***Impatiens* SunPatiens®, Spreading Shell Pink impatiens**

This hybrid impatiens produces soft-pink flowers throughout the season on vigorous, spreading plants and is resistant to downy mildew. Plants thrive under high heat, rain, and humidity and may be grown in sun or shade. This cultivar is available in plant form only. Bred by Sakata Ornamentals.

***Lactuca sativa* 'Sandy' (lettuce)**

The first AAS winning lettuce since 1985, 'Sandy' is an attractive oakleaf-type lettuce with a multitude of sweet tasting, frilly, dark-green leaves. 'Sandy' also has exceptional disease resistance (especially to powdery mildew) and is slow to bolt. Bred by Terra Organics.

***Ocimum basilicum* 'Dolce Fresca' (basil)**

Plants produce sweet, tender leaves while maintaining an attractive, compact shape. Plants regrow quickly after a harvest. 'Dolce Fresca' may be used as both an herb and an ornamental. Bred by PanAmerican Seed.

***Petunia ×hybrida* Tidal Wave®, Red Velour (F_1 petunia)**

Large, deep-red, velvety flowers cover vigorously spreading plants of this petunia. New flowers are produced continuously to cover the old, spent blooms. Plants recover quickly after a hard rain. Bred by PanAmerican Seed.

***Petunia ×hybrida* 'Trilogy Red' (F_1 petunia)**

Compact, dome-shaped plants cover and recover themselves with non-fading, upright, vibrant-red flowers. Growers will appreciate the controlled growth habit of this cultivar, thus less need for plant growth regulators and greater ease in separating and shipping. Bred by Takii & Co., Ltd.

***Raphanus sativus* 'Roxanne' (F_1 radish)**

Roots have a uniform, bright-red color and a creamy-white interior, along with a great flavor and no pithiness. This radish stays firm and solid even when oversized and grows well in a wide range of climates. Bred by Bejo Seeds, Inc.

***Salvia coccinea* 'Summer Jewel White' (Texas sage)**

Dwarf-sized, compact plant features prolific production of creamy-white flowers throughout the summer. Flowers appear almost two weeks earlier than other white salvias and are useful for attracting pollinators to the garden. Bred by Takii & Co., Ltd.

AAS REGIONAL WINNERS FOR 2015

***Brassica oleracea* var. *gemmifera* 'Hestia' (F_1 brussels sprouts) (Regions: Southeast, Mountain/Southwest)**

This cultivar produces sprouts with a bright-green exterior and smooth, dense, yellow interior. Flavor improves when temperatures dip below 40°F; however, this cultivar tolerates both warm and cool temperatures, providing the potential for a second crop. Bred by Bejo Seeds, Inc.

***Brassica rapa* subsp. *chinensis* 'Bopak' (pak choi) (Regions: Northeast, Great Lakes, Mountain/Southwest)**

This is the first pak choi to become an AAS Winner. The upright, uniform, and dense plants mature early with tender leaves and crisp, sweet stalks that can be eaten raw in salads and sandwiches. Bred by Bejo Seeds, Inc.

***Capsicum annuum* 'Hot Sunset' (F_1 banana pepper) (Regions: Southeast, Heartland, Great Lakes)**

Large, healthy, vigorous plants are disease-free and produce tasty and attractive fruits all season long. Fruits (650 Scoville units) are thick-walled with a tasty flavor and may be used fresh, grilled, roasted, or pickled. Bred by Seminis Vegetable Seeds.

***Capsicum annuum* 'Sweet Sunset' (F_1 banana pepper) (Regions: Southeast, Heartland, West/Northwest)**

'Sweet Sunset' is a compact banana pepper that is vigorous and produces a high numbers of concentrated fruit. The compact, upright plants do not require staking and can be grown in a container. Bred by Seminis Vegetable Seeds.

***Cucumis sativus* 'Parisian Gherkin' (F_1 cucumber) (Regions: Northeast, Mountain/Southwest)**

'Parisian Gherkin' is an excellent mini (or gherkin) pickling cucumber which can be picked either at the midget size or small pickle stage and processed. The disease-resistant, semi-vining plants can be planted in the garden or staked in patio containers. Bred by Terra Organics.

***Ocimum basilicum* 'Persian' (basil) (Regions: Heartland, Mountain/Southwest, West/Northwest)**

'Persian' basil is a large, vigorous plant and a prolific producer of pleasant tasting leaves for culinary use. The green foliage, sturdy branches and large leaves also make this cultivar useful as an ornamental. Bred by Terra Organics.

***Origanum syriaca* 'Cleopatra' (Syrian oregano) (Regions: Northeast, West/Northwest)**

Attractive silver-gray foliage on a compact, trailing plant makes this oregano useful as an herb and an ornamental. Different from Greek and Italian oreganos, 'Cleopatra' has a mildly spicy, peppermint-like flavor. Bred by Genesis Seeds and offered by Terra Organics.

***Solanum lycopersicum* 'Chef's Choice Pink' (F_1 beefsteak tomato) (Regions: Southeast, Great Lakes)**

Plants produce large numbers of 12- to 14-oz. pink, beefsteak tomatoes with a satisfying acid-to-sugar balance. The indeterminate, potato-leaved plants have resistance to multiple diseases. Bred by Seeds By Design.

In summer 2015, the first three AAS National and Regional Winners for 2016 were announced:

***Allium fistulosum* 'Warrior' (bunching onion or green onion) (Regional Winner: Southeast, Mountain/Southwest)**

This bunching onion grows quickly and matures early, producing a very uniform crop of slender, crisp onion stalks that are easy to harvest and clean. Bred by Seeds by Design.

***Brassica juncea* 'Red Kingdom' (F_1 mizuna or Japanese mustard) (National Winner)**

Foliage color of this high-yielding mizuna was a vibrant reddish-purple throughout the trial season and slower to bolt than other cultivars. This flavorful, mild-tasting green may be used as a vegetable or as an ornamental in containers and in the landscape. Bred by Asia Seed Co. Ltd.

***Raphanus sativus* 'Sweet Baby' (F_1 radish) (Regional Winner: Southeast, Great Lakes)**

This purple, white and rose-colored radish produces crops of uniform size with a crispy, crunchy and slightly spicy taste. Bred by Asia Seed Co., Ltd.

More information on AAS and AAS winners is available at: www.all-americaselections.org or www.aaswinners.com

Grafted watermelon transplants: a new business opportunity©

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Grafting vegetable plants onto specific rootstocks that are resistant to soilborne diseases is a unique horticultural technology attracting interest among intensive vegetable crop producers as well as organic growers in many parts of the world. Grafting often represents the only feasible measure to control a diversity of problems such as soilborne disease and saline soil conditions. In this poster presentation we provide an overview of the steps for grafting cucurbit plants, particularly watermelon, using the one-cotyledon method. The optimal stage of growth for grafting watermelon is the 1- to 2-true-leaf stage for the scion and the 1-true-leaf stage for the rootstock. Also included is a 9-day healing regimen which is appropriate for watermelon in western Washington conditions and has 90% survival for grafted watermelon transplants. Our future goal is to conduct more research to further optimize the success rate for grafting watermelon transplants, such as applying antitranspirants to reduce water loss and utilizing the splice grafting method to eliminate rootstock regrowth. Additionally, we will test grafted plants to control Verticillium wilt caused by *Verticillium dahliae* in Washington.

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Non-grafted and grafted seedless watermelon transplants: a comparative economic feasibility analysis©

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SUMMARY

Most commercial watermelon producers purchase transplants from commercial greenhouse plant propagators. This study evaluated the feasibility of producing greenhouse, seedless-watermelon transplants, both non-grafted and grafted, as well as using grafted transplants to produce seedless watermelon in Washington State. Results suggest that the production of grafted watermelon transplants can be economically feasible for commercial greenhouse propagators if the transplants can be sold at more than $0.20 per plant. This break-even price is five times greater than the break-even price of non-grafted transplants. The higher price for grafted transplants is due to the additional capital investments needed for grafting as well as the additional labor needed for grafting transplants. The extra cost of grafted transplants can be acceptable to watermelon producers if using these transplants would provide a viable alternative to field fumigation and improve crop yield. From the watermelon producer's perspective, the field utilization of grafted watermelon transplants can be economically feasible. The producer breaks even if the price of grafted transplants is about $1.38 per plant, but if the goal earn a profit that is at least equal to the profit of utilizing non-grafted transplants ($1,473 per acre), the producer would be willing to pay no more than $0.92 per plant. Watermelon producers will choose to use grafted over non-grafted transplants primarily based on the benefits gained from the effectiveness of grafted transplants as an alternative to chemical use in managing soil-borne disease. Benefits include reduced overall costs, improved yield and maintained or augmented profit relative to using non-grafted transplants.

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Effect of four root-pruning nursery containers on biomass, root architecture and media temperature©

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The nursery industry is transitioning away from standard plastic containers in attempts to reduce production costs and use of petroleum-based products and to improve plant health, root architecture, and transplant success. This study evaluated the effect of four root-pruning containers: Air-Pots®, Light Pots™, Root Pouch Pots, and Smart Pots on plant biomass, root architecture, and medium temperature, relative to standard plastic containers. Two deciduous woody shrubs were used: *Amelanchier* × *grandiflora* 'Cole's Select' and *Rhus aromatica* 'Gro Low', with six replicates of each container/species combination randomized in a complete-block design. All four root-pruning containers promoted root branching that limited circling and produced more fine roots than the standard plastic containers. The two porous-fabric containers, Smart Pot and Root Pouch Pots, displayed little to no root growth along the sides of the containers while Air-Pots and Light Pots demonstrated somewhat more, but still significantly less than the standard plastic containers. In the Air-Pots roots growing along the container sides were much shorter than those in the standard plastic container, generally terminating at one of the side holes and, thus, would not be considered detrimental to plant landscape establishment and growth. Serviceberry roots in the Light Pots displayed a strong gravitropic response, growing downwards once they were exposed to light at the container sides. This root architecture could also be considered less detrimental to long-term growth relative to the root circling typical in standard containers. An important observation of this study that was first reported by J. Altland in 2007 is that primary roots touching the side of the root-pruning containers at the time of planting often produced inward-growing roots, resulting in root structures that could lead to girdling roots later in the plant's life. Thus, at the time of planting it is important to prune roots so they are several inches away from the side of the container. For both species, dry weights of shoots were highest in the Air-Pots and lowest in the Smart Pot containers. While shoot weights ranged from 72 to 90 g in sumac, greater differences were observed in serviceberry shoots, which were nearly 66% greater in the Air-Pots (494 g) relative to the Smart Pot (298 g). The root-pruning containers also had significantly lower media temperatures measured on the sun-exposed side, with the lowest temperatures observed in Smart Pots, where medium temperatures were as much as 35°F lower than in plastic containers (121.4 vs. 86.3°F). The low medium temperatures and dry weights of shoots in Smart Pots were likely due to evaporative water loss through the highly-permeable fabric. Because all containers received the same amount of water each morning via drip irrigation, greater water loss through the highly-permeable Smart Pot containers could have resulted in mild water stress during mid and late day, leading to lower shoot biomass. This supports the findings of other studies that observed greater water requirements for plants grown in porous containers. Dry weights of roots were not reported, as fine roots of both species grew into porous medium components such as composted bark and perlite, making it unfeasible to separate the medium from the roots without removing large amounts of fine-root mass. Therefore, when media typical to nursery container production are used in such studies, root biomass data will be unreliable and, if reported, should probably be viewed with skepticism.

Literature cited

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Light source effects on hydroponically grown compact 'Winter Density' bibb lettuce©

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There is growing concern about food safety, environmental impact, and efficient energy usage in horticultural production systems. Producing lettuce under different kinds of artificial lighting can be a solution addressing these concerns. Light-emitting diodes (LED) offer the advantages of a narrow light spectrum, low power consumption, and little heat production. The objective of this study was to determine the effects of different light sources and crop phenology (growth stage) on the growth of compact 'Winter Density' Bibb lettuce in a noncirculating hydroponic system. *Lactuca sativa* 'Winter Density' bibb lettuce seedlings were started in Oasis® cubes. Seedlings were transferred to 5.1-cm net pots and put in 1.9-L containers containing a hydroponic nutrient solution. The solution was composed of Hydro-Gardens' Hobby Formula 10N-$8P_2O_5$-$22K_2O$ hydroponic fertilizer with added magnesium sulfate (9.8% Mg). The lettuce was grown in a lab under high output T-5 fluorescent lights. The light level was 119.5 µmol m^{-2} s^{-1} with an air temperature of 22.6°C. The photoperiod was 16 h. After 10 d, half the plants in the containers were moved under red+blue+white LEDs for 10 more d. At the end of the study, plant height, shoot-root ratio, percent dry weight partitioned to shoots, nutrient solution used and electrical conductivity of the remaining nutrient solution were greater under fluorescent lighting. Root dry weight, percent dry weight partitioned to roots, and shoot dry weight per nutrient solution used were greater under LED lighting. There were no significant differences in shoot dry weight, total plant dry weight, SPAD readings, or pH of the remaining nutrient solution. In conclusion, moving lettuce plants from initial fluorescent lighting to LED lighting showed that crop phenology (growth stage) enhanced certain attributes of hydroponically grown compact lettuce.

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Non-targeted mutagenesis of *Ornithogalum candicans* through exposure to ethyl methanesulfonate©

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Ornithogalum candicans [syn. *Galtonia candicans* (Decne.)] (Baker) J.C. Manning & Goldblatt, cape hyacinth, is a white flowering bulbous species native to South Africa. The large, white flowers attract a diverse set of pollinators providing pollen and nectar throughout the growing season. While it blooms profusely from early June until frost in the Willamette Valley of Oregon, there has been only one cultivar, 'Moonbeam' (Hammett and Murray, 1993), introduced to the market. Typically, the species is seed-propagated for sale in nurseries.

One issue that has been noted is the tendency for cape hyacinth to lodge once 4-5 ft.-tall inflorescences are in full flower and begin to fruit (Armitage, 2008). Additionally, the plant is too large to containerize and fit onto a nursery shipping cart when in flower. Another concern is the potential for weediness. Individual plants produce thousands of seed and, in the field, they readily germinate (pers. observ.).

Ethyl methanesulfonate (EMS) is a chemical mutagen used by breeders to induce variation in relatively homogeneous populations. Other reported effects of EMS include reduced height and fertility. Ethyl methanesulfonate can be applied to seed through imbibition (Alcantara et al., 1996; Froese-Gertzen et al., 1964; Talebi et al., 2012).

In this poster, we present the results of EMS application on *O. candicans* seeds. Seeds were collected from a single plant at the OSU North Willamette Research and Extension Center in September, 2011. Seeds were sorted into experimental units of 300 seed each. A factorial arrangement of treatments was applied to the seed. The first factor was a 24-h imbibition treatment in water (no soak or soak). The second factor was concentration of EMS (0, 0.2, 0.4, 0.6, 0.8 and 1%). Seeds were germinated and planted in the field.

Here we report the results of seed germination experiments, pollen viability, and phenotype data of the M_1 population and we report preliminary results of seed germination of the M_2 population. The general trend observed is a reduction in height and fertility as the concentration of EMS increases.

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Propagation and growth parameters of preselected pomegranate (*Punica granatum*) cuttings from the USDA-ARS National Clonal Germplasm Repository©

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Abstract

Pomegranate (*Punica granatum*) cultivars from the USDA-ARS National Clonal Germplasm Repository, Davis, California were evaluated for propagation success and early plant growth. Twelve cultivars: 'Ki Zakuro', 'Phoenecia', 'Nochi Shibori', 'Golden Globe', 'Green Globe', 'Loffani', 'Wonderful', 'Eversweet', 'Haku Botan', 'Parfianka', 'Desertnyi', and 'Ambrosia' were included in the trial. Stem cuttings were harvested from basal suckers, cut to a length of 10.5±1.0 cm long and treated with 3 g L^{-1} of indolebutyric acid and planted in a Sunshine potting mix and perlite (1:1, v/v) medium in 2.5×2.5-cm potting containers, separated by block in plastic flats irrigated with deionized water. A randomized-complete-block design was used with eight blocks and four pseudoreplicates per block, totaling 32 trees for each accession. 'Green Globe' was found to root the poorest compared to the other cultivars, with 'Ambrosia' having the second poorest success rate. All other accessions had rooting success rates above 80%. 'Parfianka' had greater branching than 'Eversweet', 'Nochi Shibori', 'Haku Botan', 'Desertnyi', 'Loffani', 'Ki Zakuro', and 'Golden Globe'. Apical shoot length was also different between two groups, with 'Golden Globe', 'Phoenicia', and 'Wonderful' with greater apical shoot growth and 'Ki Zakuro', and 'Haku Botan' growing slower. 'Golden Globe' and 'Phoenicia' were taller than 'Eversweet' and 'Haku Botan'. Leaf chlorophyll was measured with a SPAD-502 chlorophyll meter and 'Haku Botan' and 'Loffani' had greener leaves than 'Eversweet', 'Ambrosia' and 'Desertnyi'. The results of this study indicate that pomegranate cultivars vary significantly in a range of pomegranate propagation parameters and that not all pomegranate cultivars are readily propagated by vegetative cuttings treated with exogenous rooting hormone.

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Effect of ploidy level on vegetative propagation of two *Prunus laurocerasus* 'Schipkaensis' cytotypes©

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Chromosome doubling (ploidy manipulation) is a useful tool in ornamental plant breeding. One application is reducing fertility of weedy species. While many studies have compared morphological variability between cytotypes (ploidy levels), we have found none that focused on rooting potential. A plant's ability to root at a high percentage is an important consideration in determining its potential for large-scale production. *Prunus laurocerasus* L. 'Schipkaensis' is a common ornamental shrub widely used in the landscape. This study was conducted to determine if rooting potential varied between natural (2*n*=22*x*=176) and chromosome doubled (2*n*=44*x*=352) cytotypes of this cultivar.

Polyploid plants previously developed were confirmed using flow cytometry the spring before the current study was initiated. Cuttings of polyploids (44x) and standard cytotype (22x) were collected from Blue Heron Farm in Corvallis, Oregon, and from container-grown plants maintained in our program, respectively. In late July, 24 cuttings of each cytotype were collected and arranged in a randomized-block design with three blocks. All cuttings were dipped for 10 sec in a solution of 1000 ppm indole-3-butyric acid and 500 ppm 1-naphthaleneacetic acid (Woods Rooting Compound, Earth Science Products, Wilsonville, Oregon), and set in a perlite and Metro-Mix 840PC (SunGro Horticulture, Agawam, Massachusetts) (2:1, v/v) under intermittent mist. Cuttings were between 7.5 cm and 10 cm in length with 4 to 5 nodes. Three leaves were left on each cutting and these were bisected to reduce water loss.

After 1 month, the polyploid and the standard cytotype showed rooting percentages of 87.5% and 62.5%, respectively. Average root length was not different between cytotypes, but average number of roots per rooted cutting was 16.2±1.8 and 27.1±3.6, respectively. The polyploid, while having fewer roots per cutting, rooted at a higher percentage with similar average root lengths. It is likely that large-scale propagation of this alternative cytotype is a viable option. Interestingly, block 1 showed evidence of an unknown pathogen that reduced the rooting percentages of both cytotypes. However, the polyploid showed fewer signs of disease while rooting at a higher percentage. This may indicate that the polyploid has increased disease resistance and, if so, represents an option to develop cultivars with improved resistance or tolerance to shothole disease.

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The effects of auxin and substrate on rooting blueberry softwood cuttings©

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Commercial blueberry cultivars can be propagated by a range of methods including softwood and hardwood stem cuttings and micropropagation. Softwood cuttings are commonly used due to a high rooting percentage and rapid rooting period of 6-8 weeks. Rooting success varies among blueberry cultivars, but this may be due to a number of factors including timing, cultural practices, and inherent genetic rooting potential. The objective of this study was to determine the effects of rooting substrate and auxin on softwood cuttings of *Vaccinium* 'Jewel', *V*. 'Powderblue', *V*. 'Tifblue', and *V. corymbosum* 'Hodnett'.

Softwood cuttings (4.5 to 5 in.) were collected in late May or early June and randomly assigned an auxin treatment (with or without a basal dip of Dip'N Grow at 500 ppm indole-3-butyric acid and 250 ppm 1-naphthaleneacetic acid). Cuttings were inserted into individual cells (cut from 72-cell sheets) filled with substrate [pine bark, pine bark and peatmoss (3:1, v/v), or peatmoss and perlite (1:1, v/v)] and placed under intermittent mist in a greenhouse. In mid-October, roots were washed and data were collected (rooting percentage and root dry weight).

Substrate pH ranged from 5.0 [peatmoss and perlite (1:1, v/v)] to 5.7 (pine bark). Overall, rooting percentage varied among cultivars and ranged from 25% ('Jewel') to 100% ('Hodnett'). 'Tifblue', 'Jewel', and 'Hodnett' rooting percentage was greatest in pine bark and peatmoss (3:1, v/v), while root dry weight was greatest in peatmoss and perlite (1:1, v/v) for 'Tifblue', 'Jewel', and 'Powderblue'. 'Jewel' rooting percentage was 10% [peatmoss and perlite (1:1, v/v)] and 20% [pine bark and peatmoss (3:1, v/v)] greater for the auxin basal dip compared with no auxin, yet auxin did not significantly improve rooting percentage for the remaining cultivars. Blueberry softwood cuttings can be rooted in a pine-bark substrate, yet peat moss-based substrates should be considered for improved rooting. Additionally, an auxin-basal dip may improve rooting in difficult-to-root cultivars.

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Propagation of selected Kentucky natives©

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INTRODUCTION

The University of Kentucky (UK) Native Plants Program at the UK Research and Education Center in Princeton, Kentucky was started when a stunning plant, *Spigelia marilandica,* of native provenance was found to be relatively easy to propagate in spite of being described in literature as difficult to propagate. The program has been continued in support of the Kentucky native plant production economics (Ingram et al., 2015) that indicate there has been an increase in native plant production since 2003.

Propagating Kentucky native plants from known provenances has been investigated. This presentation will discuss the successes and those yet to be successfully propagated. The propagation methods were to increase numbers of plants of a given provenance for distribution and landscape evaluation; therefore, they may not be efficient production protocols.

HERBACEOUS PERENNIALS AND BIENNIALS

Amsonia tabernaemontana Walt. eastern bluestar

It has early-season blue flowers, clear green summer foliage, and an upright habit. The golden-yellow late-summer/autumn is not as pronounced as the non-Kentucky native *A. hubrichtii*. Plants at the edge of a west Kentucky wooded area were divided.

1. Seed.

The fruits are paired tan cylindrical pods borne upright within the foliage. The brown cylinder-shaped seed was broken out of the pods. The seed were planted in a germination medium. Germination was irregular and can take several weeks requiring waiting to transplant if broadcast in mass. Commercially it is recommended two to three seeds be placed directly in individual pots or cells (Pilon, 2011).

2. Division.

Division of the woody crown is relatively easy with small plants, but can be quite difficult with large long-term established plants.

3. Cuttings.

Cutting propagation has been successful with *A. tabernaemontana* 'Big Jon', a selection of the late University of Arkansas Professor Jon Lindstrom. June taken two-three inch cuttings were stuck in perlite without hormone in an outdoor mist bed; 100% rooted.

Arisaema species L.

Arisaema dracontium, green dragon, and *A. triphyllum*, Jack-in-the-pulpit, are attractive shade plants. The west Kentucky provenance plants are green leaved and green flowered. Green dragon has a unique semi-circle leaf of 7-15 leaflets depending on the vigor of the plant and environment. Green dragon gets its name from the flower; a long narrow spadix that exceeds the leaf in height. We have propagated green dragon and Jack-in-the-pulpit by seed and division. The seed were collected in west Kentucky, placed in a plastic bag, crushed to separate the fruit from the seed and washed. The seed was stored in a plastic storage bag in moist perlite and stored at 40°F. The seed was planted in 1-qt containers when radicals were observed in cold storage.

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Division is recommended for fall after the plants dry down in order to collect the seed and corms that develop over the course of the growing season. We have performed division at or after bloom. The plants were washed to separate the large corms from the parent plant and immediately potted. In this way viable seed was still produced. At this time the newly developing corms are not mature enough to harvest.

***Asarum canadense* L., Canadian wild ginger**

Wild ginger is a ground cover that grows in shade environs. Initial plantings will seem thin as wild ginger puts out just one flush of growth per season. Then it will spread by rhizomes creating a dense cover. In the wild and in landscapes near natural areas rodents dig up rhizomes and help spread the plant to neighboring areas.

We have used division as the sole means of propagation. Seed can be collected, cleaned and direct sown (Cullina, 2000).

***Cunila origanoides* (L.) Britt., dittany**

Dittany an obscure infrequent Kentucky native with a wide north central southeastern United States native range that has potential as a full sun landscape plant. In the wild it is a woody-stemmed, thin, late-summer blooming plant frequently mixed in with *Eupatorium capillifolium, Conoclinium coelestinum* (syn. *E. coelestinum*), *Spigelia marilandica*, and *Desmodium cuspidatum* that is not noticeable. Once placed in full sun it makes a rounded ground cover 12-18 in. tall by equal width. The many small blue-lavender flowers invisible in the wild, densely cover the plants in full-sun landscape sites. A member of the mint family the foliage has a pleasant aroma and is not known to be a preferred food of rodents or deer.

Little literature is available on propagation of this plant. Division has been the primary method of propagation to increase numbers of west Kentucky provenance plants. The seed is a very small brown nutlet covered with short thin hairs. The nutlet width is about the size of a line on a ruled piece of paper with slightly greater length. The seed heads remain green until mid-late fall. For easier seed cleaning wait until the heads have turned black.

***Echinacea tennesseensis* (Beadle) Small, Tennessee coneflower**

A wonderful native once endangered makes a great landscape plant. The purple-lavender petals are very attractive and once planted this plant will slowly spread filling in an area. Deadheading following bloom can lead to repeat fall blooms in Kentucky. Seed is collected from the dried seed heads in September-late fall. Small birds do eat the seeds so it is best to not wait too long to harvest seed once the heads turn black. The light tan-beige seed are broadly flat on one end tapering to a point on the opposite end. They readily germinate following dry storage and have been grown in a well-drained germination media.

***Pachysandra procumbens* Michx., Allegheny spurge**

Allegheny spurge is an attractive ornamental plant that grows well in shade and forms a dense ground cover. The primary landscape characteristic is its dark green foliage. We have yet to collect seed even though the stock plants have flowered. The primary propagation method used is division.

***Sabatia angularis* (L.) Pursh, rose gentian or rose pink**

Rose gentian is a beautiful mid-late summer blooming biennial. The pink petals with the yellow centers separated by a red line are numerous and very attractive. Regrettably, the biennial nature limits its commercial value to specialty markets. Also, seed germination occurs sporadically over time leading to trays with large plants, small plants and seed that has not germinated further limiting its potential profitability.

***Spigelia marilandica* (L.) L, Indian pink**

Indian pink or woodland pinkroot is a very showy native to the entire southeastern USA and Illinois, Indiana, Maryland, and Delaware. The tubular pink-red blooms with yellow-green throat opens to a star-like appearing yellow bloom on the red tube. The blooms

occur on an arching cyme. It was once thought to be hard to propagate based on the difficulty collecting seed. *Spigelia marilandica* seeds can be collected but it requires daily observation to avoid loss of the shiny black seeds to its explosive dehiscing seed dispersal characteristic (Bush, 2015). This characteristic leads to small seedlings in the garden area where the original plant is placed. The seeds form in bi-pods from which the seed can be collected when black on the top and black-green on the bottom. We have found that allowing the seed to dry (loss of shine) reduces germination success. Seed is directly sown or placed in washed moistened perlite in plastic storage bags. It is then stored in 40 °F and constantly observed. Once germination occurs it is planted. The author prepared a review of *Spigelia* propagation for the Eastern Region International Plant Propagator's in 2003 (Dunwell, 2003) that since has been updated and posted online (Dunwell, 2015) with Dr. Amanda Hershberger's (Hershberger, 2012) recent research in which it was reported that *S. marilandica* and *S. gentianoides* (syn. *S. gentianoides* var. *alabamensis*) may be successfully propagated by treating stem cuttings taken in May, June, July, or August with 0.3% IBA. Cuttings of *S. marilandica* × *S. gentianoides* hybrids can be taken through September. These protocols provide a basis for rapid propagation of *Spigelia*. Dr. Sherry Kitto, University of Delaware, developed a tissue culture propagation protocol (Kitto, pers. commun.). We have had AgriStarts III, (http://www.agristarts.com), Eustis, Florida, propagate *Spigelia* using Kitto's protocol and the plants from them have been uniform and have grown well.

WOODY PLANTS

Aesculus pavia L., red buckeye

Red buckeye is a beautiful plant with 8-in. panicles made up of 1-1½ in. red tubular flowers. Red buckeye is easy to propagate from seed removed as the capsule starts to split or when the seed falls to the ground and is still smooth without starting to dry and wrinkle. The seed should be planted immediately following collection. The only limitation to propagation is the amount of seed one is able to collect. Red buckeye will flower in the production systems after 2-3 years growth from seed; tailoring growth to sell a flowering plant should be possible.

Chionanthus virginicus L., white fringetree

Fringe tree is a stunning native shrub covered with fine panicles of white blooms in the spring. Followed by attractive foliage and fall dark purple fruit. Fringe tree has been very difficult to propagate. Our attempts at cutting propagation have not produced a single plant. Seed propagation has been equally frustrating with prolonged cycles of warm and cold required to get germination sometimes more than the typically described warm/cold/warm/cold cycles have been required. Bill Hendricks has stated that he places the seed in flats of sand and sets them in the corner of an overwintering house and lets them go through natural cycles of heat and cold until they germinate. It has been reported that the emerald ash borer attacks fringe tree leading to less interest in its use or the need for rapid propagation protocols (Entomological Society of America, 2015).

Clematis glaucophylla Small, whiteleaf leather flower

Clematis glaucophylla is a beautiful vine with petite pink urn-shape flowers. Very few seeds germinated from the first seed collection. In 2014 seed were collected and stored dry or stratified in moist perlite at 40°F or with the seed coat removed and stratified in moist perlite at 40°F. The stratified seed germinated while in cold storage whether the seed coat was removed or not. The germinated seed was planted in 36 cell trays filled with pine bark and peat medium (2:1, v/v). The dry seed was planted in 36-cell trays filled with peat and perlite medium (2:1, v/v). All the stratified germinated seeds grew on to produce usable plants. The dry stored seed did not germinate.

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Developing a LEAN culture at your workplace: fueling your bottom line©

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INTRODUCTION

In honor of the passing of Yogi Berra this week I thought I would begin my talk by quoting a few "Yogism's" that are appropriate for this talk.

"It's tough to make predictions, especially about the future"

"You can observe a lot just by watching"

"If you don't know where you are going, you might end up somewhere else"

All of the above quotes are relevant to understanding and embracing business in general, but can especially be applied to LEAN culture. My favorite is: "You can observe a lot just by watching." LEAN is a process that begins with observation.

I want to remind everyone that I was here last year and discussed how North Creek is using the Working Smarter Training Challenge™ to teach LEAN culture in our workplace. Similar to LEAN, the Working Smarter program teaches easy ways to take action that drive waste out of our processes. This enables our business to score a series of wins while we as individuals get to be rewarded like champions. Eventually everyone in the company develops the culture of seeking continuous "wins" or continuous improvement. These actions resulted in the reduction of lost time, decreasing unnecessary costs, and ultimately allowing us to find better ways to service our customers.

REVIEW

As a review, there are seven types of waste we recognize in a LEAN environment:

1) Transport: Unnecessary movement of materials, equipment or people.
2) Inventory: Too many materials delivered to a site; wasted materials or resources; overstocked parts and supplies.
3) Motion: Unnecessary steps taken by employees or equipment because of inadequate planning, poor communication, using the wrong equipment or tool.
4) Waiting: People standing because there is a lack of information, insufficient organization, unprepared foreman, or problematic site conditions.
5) Overproduction: Too many people on the job, providing more quantity or overproduction than needed, doing work that is not on the work order.
6) Over-processing: Reworking due to faulty information, materials, equipment, or not having standard work or following standard procedures.
7) Defects: Machine breakdowns, poor quality of materials, and ultimately service calls and replacements.

We like to add one more waste – Lost opportunity: A company cannot capitalize on the prospect of new opportunities if they can't manage their efficiency.

Shigeo Shingo, who is considered as the world's leading expert on manufacturing practices and the Toyota Production System states: "The most dangerous kind of waste is the waste we do not recognize."

GOAL SETTING

Goal setting: know what you expect to achieve. Goals should be: Specific, Measurable, Attainable, Relevant, Time based, or also known as S.M.A.R.T. goals. The following are the five basic concepts that your team needs to be aware of and use to develop and monitor their progress.

1) Specific: Create a specific goal. It has a much greater chance of being attained than a

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general goal. Know what you wish to accomplish, who will be involved, identify the location, establish a timeframe, and specify the purpose and benefit of accomplishing the goal.

2) Measurable: Establish criteria to measure progress of each goal you set. When you measure your progress, you stay on track, reach your target dates, and experience the exhilaration of achievement that spurs you on to continue the effort required to reach your goal.
3) Attainable: Identify and prioritize the goals that are the most important. Begin by figuring out ways to accomplish them so they become reality. Make sure you develop the attitudes, abilities, skills, and financial capacity to reach them.
4) Realistic: A goal must represent an objective that you are both willing and able to work toward. Be sure that every goal will realize substantial progress.
5) Timely: A goal should have a timeframe associated with it. Without this timeframe there is no sense of urgency.

It's important to remember not to take on too much at once. Continuous, smaller wins are better than big disruptive changes.

The LEAN and Working Smarter programs propose that change and implementation is: "Soft on the people and hard on the process." Companies should direct their frustration towards processes rather than the individuals doing the processes.

We use a visual tool that was developed by the Working Smarter folks. It's based on a couple facial cartoons that are known as "Builders" versus "Destroyers." We have all seen these folks at our workplace. Builders are those that are wide eyed and engaged, always interested and asking questions. They are eager to learn and do a good job. They are respectful and polite.

Destroyers, on the other hand, do not look you in the eye, are not engaged. They are often talking rather than listening, hold a negative attitude, don't ask question, shirk responsibility, and are often disruptive. When asked about Builders vs. Destroyers, Jim Paluch, the founder of the Working Smarter stated: "My attitude can color any situation, and the great thing is I get to choose the color." Enlist the builders in your workplace and weed out the destroyers. Your better employees will thank you.

TRAINING AND IMPLEMENTATION

Training is an investment in time. To get the results you expect a company must realize that they need to invest in training. This training is not a onetime event, but is an ongoing process. Recurring training sessions are how we forge our foundation lessons and facilitate continuous improvement. Along with the training lessons, we try to create enthusiasm in the workplace. This approach engages people and improves employee morale and retention. Enthusiasm fosters proper work habits and advances positive attitudes. It is important to prioritize project areas and procedures within the company that need improvement. Concentrate on these until positive changes take place. Care needs to be taken not to tackle too many improvements at one time. This dilutes the results, requiring a lot of rework because you are not able to spend the time necessary to see sustained results. An important lesson to understand is that success is built upon smaller, manageable goals. Try not to tackle too many ideas or improvement at once.

5-S pods — every place that has a thing and everything is in its place

Applying 5-S pods to work and storage areas is simple and easy. It's were we start with a lot of our projects. These are often easily accomplished and provide for easy and early wins.

5-S improves organization and efficiency. The five steps in 5-S are as follows:

1) SORT: Reducing the number of items in a work area to just those things that we really need.
2) SHINE: Cleaning and "shining" your workplace, desk, office, truck, bay, or wherever you perform your work.
3) SET IN ORDER: Evaluating and taking actions to improve workflow, reduce motion,

and increase efficiency in the setup of your workspace.

4) STANDARDIZE: Making sure the key steps are understood by everyone on how to keep the workspace looking like we used the first 3 S's.
5) SUSTAIN: Making sure all employees are trained on the standard procedure to keep an area clean and clutter free while also using visuals like charts and graphs to measure and audit current conditions.

The standard pig

I found a very good example of how a company sets out to make a product and how that product might be perceived by the workforce. It's known as the "Standard Pig."

The story goes like this: your customer asks for a drawing of a pig. You ask each member of your team to produce a drawing for your customer. You receive back a different looking pig from every team member you ask to draw one.

This story reinforces the need for standard work and standard training. You need to make certain everyone knows what type of pig the customers expects (communication with your customer is essential) and you need to train and make certain all pigs are produced to an acceptable standard. At North Creek this example flows through our processes. Offering a consistent reliable, high quality, fully rooted, propagated liner that meets our customer's expectation is what we strive for. We are developing examples of standard work for all processes. One example is our cutting standard. We have created a photo book of every crop we do from cuttings. A photo is taken of the ideal cutting. All employees refer to this standard when we go about producing that crop. We also just purchased a trimming machine. Now all flats have a standard trim height that eliminates guesswork. It also has increased production output while reducing worker fatigue.

4P training — creating the standard pig:

- PREPARE — both parties prepare to be engaged in training
- PRESENT — trainer presents the training
- PRACTICE — trainee practices
- PERFECT — trainer follows up with trainee

It's very important to document the current conditions in every process you plan to evaluate and improve upon. Tasks need to be process mapped to understand the problem and identify waste. If you can name it, you can understand it, and waste can be eliminated. Make the changes that need to be made. This will allow you to set a new standard. Train those who need to be trained on the new process. Proper training will build trust in the system.

Track the results of your efforts; this helps sustain them. Find ways to celebrate your wins. Celebrate accomplishments, acknowledge everyone who contributed.

Every employer must be aware that some employees may not adhere to the new protocol and may revert to old ways. It might be appropriate for them to leave the company or to be reassigned to a new department or task.

It's also important to implement sustaining "audits" of 5S areas or walk-through observations to see if processes are being done in the improved ways.

Remember, none of this matters and it all becomes a waste if you don't SUSTAIN the improvements. The Working Smarter Training Program is about creating a culture of continuous wins, so you are always moving your company forward.

Franklin Park Conservatory & Botanical Gardens: 2020 vision©

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Franklin Park Conservatory and Botanical Gardens is an architectural landmark, a work of art, a place of discovery. People stroll through the gardens to appreciate beauty and celebrate nature's diversity. They come to see butterflies and they learn about our world and themselves.

Since 1896, the Franklin Park Conservatory's Palm House has been a centerpiece of the 88-acre Franklin Park. In the 1990s, after hosting the AmeriFlora '92 exposition, legislative action conferred ownership of the Conservatory and 28 acres around it to a Board of Trustees.

In the years since, the Conservatory has dedicated itself to the following mission and vision:

- MISSION: inspired by horticulture, Franklin Park Conservatory and Botanical Gardens elevates quality of life and connects the community through educational, cultural and social experiences.
- VISION: to establish a new paradigm for the role, community contribution, and performance of a mission-driven organization.

The Franklin Park Conservatory and Botanical Gardens opened to the public in 1993. In its first decade, it saw the debut of much of its signature programming in exhibitions, community outreach and education. In 1994, the first exhibition of *Blooms & Butterflies* was held, becoming an annual tradition now celebrating its 21st year. In 2000, the community-gardening program *Growing to Green* was established. Beginning with an exhibition in 2003-2004 and continuing through additional exhibitions in 2006-07 and 2009-10, the Conservatory amassed the largest collection of Chihuly artwork owned by any botanical garden in the world.

In 1999-2000, work began on Phase One of the Master Plan. When completed, the Conservatory was a place of pride for the community and set a new standard for public gardens and community education. Ground was broken in 2007 for several "Growth By Design" projects: additions to the Palm House, the Scotts Miracle-Gro Community Garden Campus, and the new production greenhouse. Master Plan Phase One was completed in August, 2011.

These new additions to the Conservatory allowed for further expansion in exhibitions, programming, and community outreach. Cutting-edge exhibitions included:

- Aurora Robson; Sacrifice & Bliss (2013).
- The award-winning Bruce Munro: Light at Franklin Park Conservatory (2013-14).
- Most recently David Rogers's Big Bugs and Samuel Jaffe: Life on the Leaf Edge (2015).

The Conservatory also established its horticulture-training program, Green Corps, in 2010 that prepares participants for careers in green, environmental, and agricultural industries. A neighborhood farmers' market was initiated in 2012. The Conservatory was also the host of the American Public Garden Association (APGA) Conference in 2012 as well as the Directors of Large Public Gardens Conference (DLG) in 2015.

As a result of these new facilities, programs and exhibitions, the Conservatory was able to grow earned revenue from projects from 48% in 2008 to 60% in 2014.

Energized by the completion of the first Master Plan, Conservatory leadership undertook a comprehensive review of the original Master Plan in 2012 and concluded that several projects needed to be revised, updated or removed, due to new priorities and the current economic climate. Master Plan 2.0 retains the key components of the original master

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plan, preserving historical structures, water features and trees, pastoral green space and passive recreation, as well as paths and playgrounds. It will offer visitors a transformational experience through world-class botanical gardens with an emphasis on children, families, and STEM education. It will also ensure the Conservatory's long-term sustainability and continued impact on economic development in Columbus.

Projects for Phase One of MP 2.0 are as follows:

- An 11,000 square-foot, 200-year-old Ohio barn, containing space for the Conservatory's community outreach and education programming and Conservatory events (completed October, 2015);
- An aesthetic and functional "refresh" of the Conservatory's Grand Atrium, including replacing the roof lining and updating 20-year-old design elements;
- A new entrance and lower lobby remodel;
- A dedicated butterfly biome;
- A children's discovery place connected to the butterfly biome;
- A children's garden;
- World class display horticulture, established in a defined Conservatory footprint. Exhibitions such as the current Fall Mum Display and upcoming 2016 Spring Bulb Show demonstrate the potential for the Conservatory, via this expanded horticulture experience, to make Franklin Park Conservatory a destination garden.

Perennial plant breeding at Chicago Botanic Garden©

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INTRODUCTION

The Chicagoland Grows®, Inc. plant introduction program was founded in 1986 by the Chicago Botanic Garden, The Morton Arboretum, and the nursery consortium Ornamental Growers' Association of Northern Illinois (OGA). From its inception, the program has been dedicated to the development and introduction of superior landscape plants to the Midwestern USA and comparable climates in the USA, Canada, and Europe. Initially the program focused on the introduction of woody landscape plants, including numerous trees and shrubs from The Morton Arboretum's breeding research and historic landscape collections and selections from several regional nurseries. More recently, the program has also introduced several herbaceous perennials developed by regional nurseries and a garden center. In support of Chicagoland Grows, the Chicago Botanic Garden initiated a perennial plant breeding program in 1995, with its first introduction in 2004. Plant propagation for the Chicago Botanic Garden's breeding program was previously reported (Ault and Thomas, 2013). This report will focus on the breeding efforts of the program.

The breeding program's parameters are as follows. We have, for the most part, utilized taxa indigenous to North America, drawing on cultivated forms as well as wild-collected germplasm for breeding stock. Parent plants are selected based on their respective traits, crossed under controlled conditions, and their progeny assessed for continued breeding or potential introduction. Breeding has continued for as many as six generations beyond the original breeding stock. Most of the breeding projects have focused on developing interspecific hybrids, as advanced generation hybrids often exhibit novel flower colors and fragrances, plant habits, leaf and flower shapes, bloom times, etc., as well as broader environmental adaptability to temperature extremes, drought, soil pH, etc., than the original parents. When individual plants with introduction potential are selected out of the seedling blocks, they are clonally propagated (cuttings, division, tissue culture), and then trial blocks are evaluated for a minimum of 2 years at Chicago Botanic Garden. The best-performing plants are then propagated again and distributed for evaluation and production by a network of licensed nurseries. The timeline from the first interspecific cross attempted for a given genus to a plant selection becoming available at a retail garden center has been at least 7 to 10 years, longer for slower to mature taxa or for advanced generation selections to be developed and introduced.

SELECTED GENERA IN THE BREEDING PROGRAM

Baptisia, false indigo, breeding

The program has introduced four hybrid false indigos with eight more selections in nursery production, which are all being marketed under the Prairieblues™ false Indigo series. These are all interspecific hybrids with one exception. The program utilized the following species as its initial breeding stock: *Baptisia alba* (syns. *B. alba* var. *pendula, Baptisia alba* var. *alba*), *B. alba* var. *macrophylla* (syn. *B. leucantha*), *B. australis, B. australis* var. *minor, B. bracteata* (syn. *B. leucophaea*), *B. sphaerocarpa*, and *B. tinctoria*. In short, basically every interspecific hybrid combination attempted between the above species produced seed, and nearly all hybrid combinations developed proved to also be fertile. The most complex hybrid we developed combined four species and was still fertile. Therefore, the potential for combining these and other *Baptisia* species into a myriad of complex interspecific hybrids seems almost unlimited. Our selections and their parentage are found in Table 1.

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Table 1. Our selections and their parentage.

Introduction	Combination
Baptisia ×*variicolor* 'Twilite' PP#19,011 (2006 introduction)	*Baptisia australis* × *B. sphaerocarpa*
Baptisia ×*bicolor* 'Starlite' PP#19,971 (2007 introduction)	*Baptisia australis* × *B. bracteata*
Baptisia 'Midnight' PP#20,432 (2008 introduction)	(*Baptisia tinctoria* × *B. alba*) × *B. australis.*
Baptisia 'Solar Flare' PP#20,408 (2008 introduction)	[(*Baptisia tinctoria* × *B. alba*) × *B. australis*] × *B. sphaerocarpa*(?) or open-pollinated
Baptisia australis 'Blue Mound' PP#25,902	*Baptisia australis* (syn. *B. australis* var. *australis*) × *B. australis* var. *minor*
Baptisia 'Lavender Rose' PP#25,876	Advanced generation hybrid from *B. australis* and *B. bracteata*
Baptisia 'Lunar Eclipse' PP#25,875	Complex hybrid derived from *B. alba, B. australis, B. bracteata*, and *B. tinctoria*
Baptisia 'Mojito' PP#25,987	Complex hybrid developed from *B. australis, B. bracteata*, and *B. sphaerocarpa*
Baptisia 'Royal Purple' PP#25,508	Complex hybrid developed from *B. australis, B. bracteata*, and *B. sphaerocarpa*
Baptisia 'Sandstorm' PP#25,926	Advanced generation hybrid from *B. australis* and *B. bracteata*
Baptisia 'Spilled Buttermilk' PP#26,319	*Baptisia australis* × *bracteata* selection backcrossed to *B. bracteata*
Baptisia 'Sunny Morning' PP#25,479	*Baptisia sphaerocarpa* × *B. alba*

Here are some of the traits we observed for the species we utilized and their influence on our hybrids:

- *Baptisia alba*: plants were chlorotic and not terribly vigorous on our alkaline (pH = 7.6) clay soil. Our hybrid from it, *B.* 'Sunny Morning', also exhibits a preference for a neutral to acidic soil. But this species does impart purple tinted stems in the spring in its hybrids.
- *Baptisia alba* var. *macrophylla*: the northern genotype of this species tends to produce fewer stems than most of the other species, has no foliage on the lower stems, is one of the last to bloom, and also produces the longest inflorescences, all of which can be seen in its hybrids. These hybrids can take longer to mature in the garden, but the lack of lower foliage on strongly upright stems also results in the ability to interplant closer to other plants.
- *Baptisia australis* var. *australis*: the large vigorous form of the species is also the best known false indigo species. It imparts heat and cold tolerance, vigor, broad soil type and pH adaptability to its progeny, but habits can be irregular and hybrid progeny may grow huge; the original plant of *Baptisia* ×*variicolor* 'Twilite' grew as large as 5.5 ft tall and 9.5 ft wide, with no stem lodging!
- *Baptisia australis* var. *minor*: this is the southern genotype of the species. Germplasm from Texas proved vegetatively hardy in northern Illinois (USDA Zone 5), but flowering was limited. It imparts large flowers, a beautiful low dichotomously branching habit, and finer foliage, along with great heat adaptability and high soil pH tolerance. The true form should be utilized more in breeding, especially for USDA Zones 6 to 9.
- *Baptisia bracteata*: the southern genotypes did not prove to be hardy in northern Illinois. Even the regional genotypes proved to be temperamental in the garden. It is the earliest blooming species we used, the only one with horizontal inflorescences, and along with *B. australis* var. *minor* produces the largest flowers. Both the species and its hybrids are a bit more difficult to root from cuttings. It imparts early bloom, a compact habit, and heavy bloom with large flowers to its progeny. The horizontal inflorescences of the species don't seem to be passed along to its hybrids unless backcrossed to the species.

- *Baptisia sphaerocarpa*: another southern species (we trialed germplasm from Arkansas and Texas) that is perfectly vegetatively hardy and in most years floral bud hardy in northern Illinois. It imparts vigor, soil adaptability, and bushy habits to its progeny. Its yellow flower color is dominant when crossed to white-flowered *Baptisia* and blends when crossed with the blue-violet flowers of *B. australis* producing a mélange of violet, purple, copper, brown, and other odd-colored progeny.
- *Baptisia tinctoria*: the most vexing and intriguing species we utilized. It is the only repeat blooming species of the ones we used, producing short stems of yellow flowers from spring and then well into August. The airy stems and delicate small leaves give it a refined habit. However, southern genotypes did not prove hardy for us. Regional genotypes had to be grown on sharply drained soil, as it is native to almost pure sandy soils in Illinois and Indiana. The small flowers and short inflorescences proved fairly dominant in the crosses we made from it. Intriguingly, it did impart a slightly longer bloom period to our one released hybrid from it, *Baptisia* 'Midnight'. I would encourage breeders located where the species is more amenable to cultivation to work with it to capture its repeat bloom, but to avoid some of its less desirable traits.

***Echinacea*, coneflower, breeding**

The program has introduced four hybrid coneflowers, all marketed under the Meadowbrite™ coneflower series. These are all interspecific hybrids.

Echinacea 'Art's Pride' PP#15090 (Orange Meadowbrite™ coneflower) a 2004 introduction. The first orange-rayed coneflower in the marketplace was a second generation cross of *Echinacea purpurea* and *E. paradoxa*. We crossed the two species in 1998. The first generation hybrids between the two species bloomed in 2000 with light magenta ray flowers, and unlike either parent, were sweetly fragrant. These hybrid plants were crossed in 2000. The second generation hybrids bloomed in 2002 in an amazing melange of white, magenta, yellow, and orange ray flowers (Figure 1). It was from these plants that our introduction was selected in 2002. The hybrids were again all fragrant. Key to the development of orange hybrid coneflowers was the use of a white-flowered selection of *E. purpurea* in the original cross. The cross of *E. purpurea* using the typical magenta ray flowered forms × *E. paradoxa* produced second generation hybrids in muddy magenta, rusts, and violets.

Figure 1. *Echinacea purpurea* × *Echinacea paradoxa* hybrids at Chicago Botanic Garden.

Echinacea 'CBG Cone3' PP#16636 (Mango Meadowbrite™ coneflower) a 2004 introduction. The rays are mango yellow in color. This selection arose as a mutation in tissue culture from *Echinacea* 'Art's Pride' (above), which highlighted the challenge of producing large numbers of coneflowers in tissue culture in earlier years. It proved to be stable when intentionally propagated in tissue culture. I have not encountered any recent reports of *Echinacea* being unstable in tissue culture, presumably a result of the tissue culture protocols being refined over time.

The two selections above and other early orange and yellow-rayed hybrid coneflowers developed from the same parent species by fellow breeders proved to be culturally challenging, often being short-lived in garden cultivation. This challenge no doubt arose from the *E. paradoxa* ancestor. This species is found in limestone glades in the Ozarks, where it is adapted to high pH, limited nutrient availability, and exceptional soil drainage. It forms a taproot adapted to coursing deep into a rock crevasse — not our typical garden cultivation conditions! *Echinacea purpurea* × *E. paradoxa* hybrids appear to be perfectly fertile though, and so advanced generation crosses being made by other breeders are proving more amenable to cultivation, either through careful selection and/or by backcrossing to other, easier to cultivate and maintain garden forms of *E. purpurea*.

Echinacea 'CBG Cone 2' PP#18546 (Pixie Meadowbrite™ coneflower). A more complex interspecific hybrid that combined two hybrid lines. We crossed *E. purpurea* 'Magnus' × *E. tennesseensis* in 1996 and then *E. angustifolia* × *E. tennesseensis* in 1997. Both hybrid lines proved to be reasonably fertile. The former cross was quite vigorous and garden-adaptable, but most of the progeny disconcertingly produced tall inflorescences (like the *E. purpurea* parent) that then branched high up the stems producing multiple flower heads per stem (like the *E. tennesseensis* parent). The stems on most of these hybrids tended to lodge, but a few more compact forms were eventually selected. The *E. angustifolia* × *E. tennesseensis* hybrids were compact, bushy, and floriferous, but a challenge to grow culturally on our wet clay soils (both parent species originate from well drained soils in drier habitats). The two hybrid lines were crossed in 2000 (*E. purpurea* 'Magnus' × *E. tennesseensis,* the seed parent), and the selection made in 2002. It wasn't introduced until 2007, as it proved more difficult to root in tissue culture. But once introduced, it became popular for its bushy, compact habit, and its ability to bloom from July to September (in USDA Zone 5), producing a plethora of flower heads with perky, upturned magenta rays. It appears to have a higher aster yellows resistance than other selections. Time has also proven it to be one of the longer-lived coneflowers currently in cultivation (we have observed high survival of 5-year-old plants). Gardens could use more *E. tennesseensis*-influenced hybrids. However, there are challenges in breeding with this species. *Echinacea tennesseensis* × *E. paradoxa* hybrids were all mules (sterile). Our *E. tennesseensis* × *E. purpurea* hybrids were very difficult to cross to our *E. purpurea* × *E. paradoxa* hybrids, and when we were successful in combining these hybrid lines, the progeny invariably had inferior plant habits and produced ray flowers in muddy colors that faded with age. Our most promising lines that we unfortunately abandoned for other projects were advanced generation crosses of white-flowered *E. purpurea* × *E. tennesseensis*. We produced a few hybrids similar to the Pixie Meadowbrite™ coneflower only with white rays and green disks. These also proved difficult to propagate in tissue culture and so were never introduced. I highly encourage other breeders to purse this line of breeding for longer lived and garden adaptable coneflowers.

Echinacea 'Burgundy Fireworks' PP#23,691. Selected in 2006 and introduced in 2012 for its compact habit, upturned dark burgundy rays that are fused into tubes, and its dark violet tinged foliage and stems, this selection has a complicated pedigree. The fused ray trait appeared as a mutation in a line of *Echinacea* [*laevigata* × *purpurea*] × [*tennesseensis* × *laevigata*]. The dark stems and foliage was derived from a line of *Echinacea* [*purpurea* × *tennesseensis*] × *purpurea*. These two lines were crossed and the progeny then sibbed to segregate 'Burgundy Fireworks'. Most of the selection's sibs were very inferior in habit, but were still quite fertile, indicating there are various lines of interspecific coneflowers that could be pursued. The fused ray trait appeared several times in a number of coneflower lines. During the breeding of 'Burgundy Fireworks' its fused ray trait was shown to be a

simple recessive trait, and could therefore be in theory programmed into other lines in two generations of crossing.

For a more detailed though earlier accounting of *Echinacea* breeding, see Ault (2006).

***Phlox,* phlox, breeding**

Despite the ongoing popularity of phlox, notably the spring-blooming moss phlox, *Phlox subulata* and its relatives, and the summer blooming garden phlox, *Phlox paniculata*, relatively few of the 65 or so species of phlox are represented in the trade, and fewer still interspecific hybrid phlox are available. Probably the most commonly grown interspecific phlox currently in cultivation are *P.* ×*arendsii* (*P. paniculata* × *P. divaricata*) hybrids, *P.* ×*procumbens* (*P. subulata* × *P. stolonifera*) hybrids, and hybrids of *P. subulata* and *P. bifida* that are masquerading as *P. subulata* selections. There are also a handful of hybrid phlox out of Europe (many more selections are sold there), sold as *P.* ×*douglasii* hybrids (an invalid designation as there is a species *P. douglasii*, and so the hybrid name of the same is incorrect). These are reputedly hybrids of *P. subulata* with various western phlox species, or hybrids between various western species. Their actual parentages are not known.

There are challenges to developing interspecific phlox hybrids. Many of the species won't hybridize readily with one another, and some of the combinations that have been successful can only be made in one direction (Zale and Jourdan, 2012). I suspect many of the interspecific hybrids are infertile or only with very low fertility, making advanced generation breeding and selections difficult, if not impossible. *Phlox* ×*procumbens* seems to fall into the infertile category, for example. Some interspecific hybrids are fertile, such as *P. subulata* × *P. bifida*, which allows for more interesting trait selection in advanced generations. Also restricting the development of more interspecific hybrid phlox is the lack of availability of most of the species in the horticultural trade, which can be confounded by the exacting cultural requirements of most of the western desert or montane species.

With these caveats in mind, we launched an interspecific phlox breeding program at Chicago Botanic Garden in 2002. Given the relative ease of cultivating the eastern phlox species, most of our early efforts were directed towards crossing various eastern species. We had many more failures than successes. Most of the crosses attempted between *P. carolina, P. divaricata*, *glaberrima* subsp. *triflora* (=*P. triflora*), *P. ovata* (=*P. latifolia*), *P. maculata, P. paniculata, P. pilosa*, *P. stolonifera*, *P. subulata*, and others failed to produce seed, or the limited seed produced often failed to germinate. One odd hybrid was produced from *P. paniculata* × *P. stolonifera* that produced a stout stem or two upright in spring and then lodging in summer, with a small terminal cluster of pinched flowers. It died after a few years in the garden. Due to the limited germplasm we had for some of these species, the reciprocal crosses were not always attempted, which may have enhanced seed set for some of the crosses. One outstanding plant did come from this earlier work, that being *P.* 'Forever Pink' PP# 24,918, a 2013 introduction from a cross made in 2007. Originally thought to be a cross of *P. buckleyi* × *P. carolina*, careful examination of the two parent plants in later years proved them to both be *P. glaberrima* subsp. *triflora* selections; this highlights the difficulty in proper identification of some of the more arcane phlox selections in cultivation.

In more recent years, we have concentrated our efforts on spring-blooming, interspecific hybrid phlox. We have been evaluating and attempting to cross eastern taxa (*P. subulata, P. nivalis, P. stolonifera*), midwestern (*P. bifida*), and western taxa (*P. albomarginata*, *P. alyssifolia, P. condensata, P. grayi, P. kelseyi, P. diffusa*). As reported in the literature, *P. bifida, P. nivalis* and *P. subulata* are all proving to be interfertile, and in fact many of the supposed *P. subulata* cultivars in cultivation are likely a muddle of hybrids of these three species. All three species and their hybrids prefer well drained soils, full sun, and reasonable moisture availability. *Phlox bifida* naturally occurs either on sandy soils or on limestone outcrops in the Midwest. I have not rigorously evaluated selections of both ecotypes to test if their breeding behaviors vary. Crosses of *P. bifida* with *P. subulata* and *P. stolonifera* have produced vigorous, mounding plants with cleft-petals (hence the bifida species epithet for *P. bifida*). These are proving garden amenable and quite hardy. The *P. bifida* × *P. subulata* hybrids we have developed have been fertile, but the few *P. bifida* × *P. stolonifera* hybrids we have

developed have been sterile.

Phlox ×*procumbens* (*P. subulata* × *P. stolonifera*) is proving to be an odd case. There are a few cultivars in the trade, and we introduced in 2015 our own selection, *P.* ×*procumbens* 'Pink Profusion' PP# 25,883, which produces large one-plus inch wide flowers in deep purple pink. It has been performing better further south (Zones 6-8) than up north (Zones 4-5), not surprising given both of the parents are from the mid-Atlantic region and further south. 'Pink Profusion' appears to prefer a well-drained soil, full sun, and good moisture availability. Given one parent is found on rocky ledges and slopes in good sun (*P. subulata*) and the other is a moist, woodland or shaded streamside plant (*P. stolonifera*) predicting its preferred habitat is a challenge. Crosses like this between species from very divergent habitats need broad testing to determine how they are best cultivated. If all of the *P.* ×*procumbens* cultivars are sterile (the ones we have tested appear so) this limits being able to line breed them and select for different garden conditions.

The aforementioned western species have proven to be a real challenge to keep alive, let alone grow well, in northern Illinois. Our best successes have been in a deep sand bed filled with coarse quartz-sand plus some organic matter. The western taxa hold promise for a variety of factors, including high pH tolerance (*P. albomarginata*, *P. alyssifolia*), high salinity tolerance (*P. kelseyi*), fragrance (*P. multiflora*), shade (*P. diffusa*), drought, heat, cold and other tolerances, as well as novel flower colors, foliage traits, etc. Most of these western taxa are barely represented in cultivation, and there are selected forms reasonably available only of *P. kelseyi* ('Lemhi Midnight' and 'Lemhi Purple'). Two of our first successful interspecific hybrids were *P. albomarginata* × *P. kelseyi* and *P. alyssifolia* × *P. kelseyi*, both producing very compact plants with light violet flowers. The former has produced a few seed in further crossing; the latter appears to be sterile. Our best success to date is a 2015 introduction, *Phlox* 'Violet Pinwheels' PP# 25,884, which resulted from a cross made in 2008 between *Phlox bifida* and *Phlox kelseyi* 'Lemhi Purple'. Its spring flowers with uniquely upturned petals are a deep violet purple, different from most other spring blooming moss phlox in the trade. 'Violet Pinwheels' requires a well-drained soil, good light, and a uniform moisture supply during its growing season. Our effort to cross these and hopefully other phlox species is ongoing.

Veronica, speedwell, breeding

The program has introduced two speedwells. *Veronica* 'Whitewater' PP#22783, introduced in 2011, is a white-flowered branch sport of the popular 'Waterperry Blue', discovered by John Wachter of Elite Growers, Inc. It is an excellent groundcover for sun to partial shade with adequate moisture supply. It carpets itself with glistening white flowers in spring, and can repeat bloom later in the season. Like its clonal parent, it also produces attractive red fall foliage. *Veronica* 'Tidal Pool' PP#23,341, selected in 2009 from a cross made at Chicago Botanic Garden in 2007 between *Veronica armena* and *V. pectinata* 'Rosea' and then introduced in 2012, is a very adaptable groundcover for drier sites and full sun, producing its dark blue-violet flowers in spring followed by attractive and disease resistant foliage all summer. A number of interspecific hybrids were developed at the Chicago Botanic Garden, mostly between the drier habitat, groundcover types, but most of them proved to be poorly adapted to our seasonally wet, poorly drained clay soils and high summer humidity. For the sake of disclosure to assist other breeders, here is a list of the crosses that definitely produced hybrids. Note: *Veronica* taxonomy is a muddle, and some of the selections in cultivation may not be correctly identified to species. Caveat emptor! The most promising crosses we made are marked with an *: **V. alpina* 'Alba' × *V. spicata* 'Silbersee', *V. allionii* × *V. spicata* subsp. *incana*, *V. stelleri* 'Mann's Variety' × *V. allionii*, *V.* 'Giles van Hees' × *V. spicata* subsp. *incana*, *V. armena* × *V.* 'Blue Reflection', **V. turrilliana* × *V. cuneifolia* ssp. *Issaurica*, **V. cuneifolia* × *V. turrilliana*, **V. liwanensis* × *V. oltensis*, *V. oltensis* × *V. armena, *V. cuneifolia* × *V. armena.* A few more-moisture tolerant groundcover speedwell hybrids are still being evaluated at Chicago Botanic Garden.

Other, currently active breeding projects at Chicago Botanic Garden will be presented at a future date.

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Weigela species and cultivar genome size and ploidy estimations: shrub breeding©

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INTRODUCTION

Weigela are among the most popular flowering shrubs for temperate landscapes as they tolerate a wide range of cultural conditions, propagate easily from cuttings, and flower heavily in late spring. The genus is composed of 10 species native to China, Japan, Manchuria, and the Korean peninsula. Since the genus was brought to western horticulture near 1860, over two hundred cultivars have been introduced (Dirr, 2009; Sheffield Botanical Gardens, 2015). Introductions continue today with breeding work emphasizing the development of compact plants, novel foliage colors, and recurrent blooming characteristics. One cultivar, 'Courtalor', Carnaval® weigela is widely promoted as a reblooming polyploid (Pantin, 2015; Wood). Because polyploidy may be associated with ornamental characteristics that breeders may be selecting for, such as reblooming, we set out to investigate the presence of polyploidy in natural populations and extent of polyploidy in available cultivars. This manuscript reports genome size and ploidy estimations for 10 species and 46 cultivars, from a total of 74 accessions.

METHODS AND MATERIALS

Plant material was sampled from plants growing at The Morton Arboretum, the Chicago Botanic Garden, and the Arnold Arboretum of Harvard University. Genome sizes were determined by using a flow cytometer (CyFlow® PloidyAnalyser; Partec. Münster, Germany) with materials and protocols from Cystain PI absolute P test kits (Partec. Münster, Germany). Tissue samples were collected from expanding leaves and co-chopped with an internal standard, a leaf sample of *Pisum sativum* 'Ctirad', with a known genome size of 8.76 pg (Greilhuber et al., 2007). After chopping the sample was filtered through a 30-micron mesh filter (Celltrics®; Partec. Münster, Germany) and then stained with propidium iodide from the test kit. After staining the samples were immediately loaded and analyzed by the flow cytometer. Data was collected until at least 5000 nuclei of the unknown sample and at least 3000 nuclei of the internal standard were counted, CVs were maintained at less than 5% for the sample and the internal standard. Three replications were performed per genotype tested. Data was interpreted by one-way ANOVA ($P<0.05$) and Fisher's LSD for means separation ($P<0.05$). Our genome sizes were compared to reported chromosome counts to infer chromosome number and ploidy level.

RESULTS AND DISCUSSION

Genome sizes of our samples grouped from 1.91 to 2.32 pg of DNA; with one outlier, *W.* 'Courtalor', Carnaval® weigela at 3.03 pg of DNA (Table 1). Looking at literature, Duron and Decourtye report chromosome counts on the cultivar *W.* 'Newport Red' (syn. 'Vanicek') to be $2n=2x=36$, a diploid (1990); Sokolovskaya and Probatova (1985) report chromosome counts of *W. praecox* to be $2n=36$. Comparing these reports to our results we infer that the group with genome sizes of 1.91-2.32 pg of DNA are all diploid ($2n=2x=36$), and because *W.* 'Courtalor', Carnaval® weigela has approximately 1.5 times greater DNA content than the diploid group that it is a triploid (be $2n=3x=54$).

From our sampling across all ten species and from across some of the species ranges it appears that polyploidy does not occur or does not commonly occur in wild *Weigela* populations. Additionally our screening of 46 cultivars uncovered only one polyploid, suggesting that polyploidy among existing *Weigela* cultivars is also not common. On deeper

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investigation to the origins of *W.* 'Courtalor', Carnaval® weigela it was found that it had been derived from mutation breeding program in France. The breeders had artificially induced polyploidy (tetraploids, 2*n*=4*x*=74) by in vitro colchicine applications and backcrossed tetraploids with diploids to recover triploids; leading to three selections *W.* 'Courtared', Lucifer® weigela, *W.* 'Courtamon', and *W.* 'Courtalor', Carnaval® weigela (Duron and Decourtye, 1990). In our work only *W.* 'Courtalor' CARNAVAL was tested and we did not confirm the ploidy level of these other two selections.

Table 1. Relative genome size and ploidy levels determined via flow cytometry for species and cultivars of *Weigela.*

Taxa	Source[1]	Accession #	Relative 2C genome size [mean ± SE (pg)]	2C ploidy level (*x*)
W. 'Courtalor', Carnaval® weigela	MOR	359-2015 ct	3.03 ± 0.02	3
W. middendorffiana	MOR	354-2015 ct	2.32 ± 0.00	2
W. decora	MOR	53-200*1	2.28 ± 0.02	2
W. japonica	ARN	1317-84-A	2.23 ± 0.00	2
W. hortensis	ARN	414-2007-B	2.20 ± 0.01	2
W. 'Sunset', My Monet® sunset wiegela	MOR	221-2014*2	2.16 ± 0.00	2
W. ×*incarnata*	MOR	333-85*1	2.14 ± 0.02	2
W. floribunda	ARN	1019-90-rA	2.13 ± 0.05	2
W. 'Carlton', Ghost® weigela	MOR	348-2015 ct	2.13 ± 0.01	2
W. 'Verweig', My Monet® weigela	MOR	214-2007*2	2.12 ± 0.05	2
W. 'Bokratwo', Merlot Pink weigela PP#21763	MOR	357-2015 ct	2.09 ± 0.02	2
W. 'P. Duchartre'	MOR	1007-80*1	2.08 ± 0.03	2
W. 'Alexandra', Wine and Roses® weigela	MOR	426-2001*5	2.08 ± 0.02	2
W. 'Bristol Snowflake'	MOR	353-2015 ct	2.08 ± 0.04	2
W. 'Argento-marginata Variegata'	MOR	559-71*1	2.07 ± 0.01	2
W. 'White Knight'	MOR	1078-2004	2.07 ± 0.02	2
W. 'Bramwell', Fine Wine® weigela	MOR	164-2008	2.07 ± 0.02	2
W. florida 'Variegata'	MOR	905-62*1	2.06 ± 0.00	2
W. 'Bokraspiwi', Spilled Wine® weigela	MOR	358-2015	2.06 ± 0.01	2
W. 'Pink Delight'	CBG	236-1992	2.05 ± 0.02	2
W. 'Groenewegenii'	MOR	564-71*1	2.05 ± 0.02	2
W. 'Bokrashine', Shining Sensation™ weigela	CBG	639-2012	2.05 ± 0.01	2
W. 'Victoria'	CBG	709-2003*6	2.05 ± 0.02	2
W. 'Bokrafive' Merlot Rose	MOR	355-2015	2.05 ± 0.01	2
W. 'Pink Princess'	MOR	89-75*1	2.04 ± 0.02	2
W. 'Bokrafour', Flamingo Pink® weigela	MOR	356-2015	2.04 ± 0.01	2
W. 'Samba'	CBG	65-2012*3	2.04 ± 0.02	2
W. 'Centennial'	MOR	330-85*2	2.03 ± 0.00	2
W. decora	ARN	81-90-A	2.03 ± 0.01	2
W. 'Candida'	CBG	171-2003*1	2.03 ± 0.02	2
W. 'Elvera', Midnight Wine® weigela	CBG	501-2010	2.03 ± 0.02	2
W. subsessilis	ARN	906-77-E	2.02 ± 0.01	2
W. coraeensis	MOR	423-58*1	2.02 ± 0.02	2
W. 'Bristol Ruby'	MOR	1004-80*1	2.02 ± 0.01	2
W. 'Newport Red' (syn. 'Vanicek')	MOR	1009-80*3	2.02 ± 0.02	2
W. 'Tango'	CBG	66-2012*2	2.02 ± 0.04	2
W. 'Bokrasopea', Sonic Bloom® Pearl	CBG	1178-2014*4	2.01 ± 0.01	2
W. 'Olympiade', Briant Rubidor	CBG	898-1998	2.01 ± 0.02	2
W. 'Java Red' sport	CBG	61-2012	2.01 ± 0.02	2
W. 'Dark horse'	CBG	04R5293*03	2.01 ± 0.01	2
W. 'Red Prince'	MOR	1317-2004*1	2.00 ± 0.01	2
W. 'Walweigeye', Eyecatcher® weigela	CBG	Q4R5295*7	1.99 ± 0.01	2

Table 1. Continued.

Taxa	Sourcez	Accession #	Relative 2C genome size [mean ± SE (pg)]	2C ploidy level (x)
W. subsessilis	ARN	317-2001-C	1.99 ± 0.01	2
W. 'Dart's pink lady'	CBG	79-1999*5	1.99 ± 0.01	2
W. 'Brigela' French Lace™ weigela	MOR	785-2005*1	1.99 ± 0.02	2
W. florida	ARN	82-2010-A	1.98 ± 0.03	2
W. 'Kolmagira', Rainbow Sensation™ weigela	MOR	360-2015	1.98 ± 0.01	2
W. 'Rumba'	CBG	64-2012*10	1.97 ± 0.03	2
W. 'Kosteriana Variegata'	CBG	382-2001*8	1.97 ± 0.03	2
W. subsessilis	ARN	587-53-A	1.97 ± 0.01	2
W. maximowiczii	ARN	167-97-B	1.97 ± 0.01	2
W. praecox	MOR	554-79*11	1.97 ± 0.03	2
W. subsessilis	CBG	249-2008-A	1.96 ± 0.01	2
W. hortensis	MOR	178-85*2	1.96 ± 0.01	2
W. 'Verweil-4', Sonic Bloom® Red	CBG	1202-2013*1	1.96 ± 0.02	2
W. 'Java Red'	CBG	612-2012*5	1.96 ± 0.02	2
W. 'Bokrasopin', Sonic Bloom™ Pink	CBG	961-2013*3	1.95 ± 0.03	2
W. 'Suzanne'	CBG	481-2003	1.95 ± 0.03	2
W. florida	ARN	132-96-B	1.94 ± 0.01	2
W. florida	ARN	422-93-A	1.94 ± 0.02	2
W. florida var. *venusta*	ARN	817-84-B	1.94 ± 0.05	2
W. 'Foliis Purpurius'	CBG	957-1991*1	1.94 ± 0.00	2
W. florida	MOR	319-94*1	1.94 ± 0.00	2
W. hortensis	ARN	279-84-B	1.94 ± 0.01	2
W. praecox	ARN	966-85-D	1.93 ± 0.01	2
W. 'Sunny Princess'	CBG	191-2013*1	1.93 ± 0.03	2
W. 'Styriaca'	CBG	638-2003*3	1.92 ± 0.01	2
W. looymansii 'Aurea'	CBG	1423-2002*2	1.90 ± 0.00	2
W. preacox	ARN	843-84-B	1.90 ± 0.01	2
W. florida	ARN	404-86-B	1.90 ± 0.02	2
W. 'Abel Carriere'	CBG	76-1999	1.90 ± 0.01	2
W. florida	ARN	125-2003-B	1.89 ± 0.00	2
W. hortensis	ARN	30-2001-C	1.88 ± 0.01	2
W. praecox 'Korean Sunrise'	CBG	482-2003*6	1.87 ± 0.03	2

[1]Source Codes: MOR, The Morton Arboretum, Lisle Illinois; ARN: Arnold Arboretum of Harvard University, Boston, Massachusetts; CBG: Chicago Botanic Garden, Glencoe, Illinois.

At the beginning of our investigation we had thought that recurrent blooming may be linked to polyploidy in weigela, but this does not necessarily appear to be the case. Although 'Courtalor' CARNAVAL is a recurrent blooming polyploid, other repeat or re-blooming cultivars such as the SONIC BLOOM series ('Verweil-4' SONIC BLOOM Red, 'Borksopin' SONIC BLOOM Pink, and 'Bokrasopea' SONIC BLOOM Pearl), 'Red Prince', and 'White Knight' all are diploid. Mutation breeding and ploidy manipulation may be viable methods for further improvement in *Weigela*, including further improvement in flower size, heavier recurrent bloom, and improvement in plant stature. The new plant development program at The Morton Arboretum has a weigela improvement program underway.

ACKNOWLEGEMENTS

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A perspective on the importance of managing juvenility in plants: focus on plant improvement and propagation©

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INTRODUCTION

Although I took the opportunity to officially retire from my professorship in horticulture at a major Midwest university, on-going research projects along with increased participation in the projects at Knight Hollow Nursery, Inc. continued to involve me in research activities encompassing both propagation and plant genetic improvement. These activities have also involved discussions with growers and researchers about how to accomplish various goals. In explaining my ideas, I soon became aware that the concept of managing the juvenile/adult phase of development in crops was often not well understood nor its importance well appreciated. Occasionally I ended up taking time to explain my perspective on developmental change in plants and how this would be a major part of the particular project we were discussing. One result of all this "retirement" activity was to include plant juvenility in progress talks I was asked to present. My discussion here today at IPPS is a continuance of this theme.

I am very aware that most of the IPPS audience is thoroughly aware of the importance of managing plant juvenility, so I have skipped over most of the basics. But I do hope that this discussion and the practical example that I highlight will re-emphasize the importance of keeping aware of how plant development influences our everyday progress.

SOME GENERAL THOUGHTS ABOUT JUVENILITY

The concept of juvenility in plants can be quite difficult to discuss since we really do not have a thorough understanding of this part of plant development. For example, if I were given two sections of a plant stem, could I tell which one was more juvenile than the other? Basically the answer is no as we have yet no clear "markers" that I can analyze that will clearly define the juvenile state of these stem pieces. There is ongoing research involving gene expression that hopefully may be able to give us such tools. But for our discussion, what this deficit means is that we are left with circumstantial observations based on plant responses. For example, one of the most reliable markers for juvenile tissue is that it possesses the highest capacity to regenerate missing parts (such as adventitious roots and buds). For the adult phase of development, slower vegetative growth accompanied by the capacity to flower is usually a readily apparent visual marker.

One question I often ask audiences while showing them a flowering potted plant is 'Where is the most juvenile part of this plant?" Intriguingly, this may seem like a simple question, but actually it can be quite complex, again because we do not have clear biological markers. After some thought, three answers are appropriate: the reproductive cells, the roots/rhizomes, and the plant collar. The embryo in seed development can be considered the most juvenile part of a plant; interestingly the seeds form in the most "adult" tissues of a plant (as defined by capacity to flower) and intriguingly undergo complete "rejuvenation." The roots generally do not have a capacity to flower (thus roots are never adult??) and do often retain regenerative capacity (such as root cuttings generating adventitious shoots and adventitious axillary roots). I have always thought that one of the most fascinating morphological parts of a plant is the collar region at the juncture of the root and shoot system. For a plant that developed from a seedling, the collar originated in the highly juvenile embryo and this juvenile trait seems to remain in the collar region throughout the

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life of the plant. The most juvenile shoots of a tree often come from the collar as basal suckers. In the rooting of a stem cutting, maybe all we are doing is in part regenerating a collar region?

EXAMPLE OF THE NEED TO CONTROL JUVENILITY TO ESTABLISH A PROPAGATION PROTOCOL

One of the major projects with which I am now associated involves developing an upper Midwest industry based on growing, processing and marketing American hazelnut, *Corylus americana*. American hazelnut is a native shrub with a center of genetic diversity in the Midwest. Demand for hazelnuts as a component in numerous edible products is high. The combination of these two facts along with a diverse interest of a group of researchers and growers has resulted in the formation of a consortium to develop a new industry (Upper Midwest Hazelnut Development Initiative, http://midwesthazelnuts.org/description.html). All aspects of creating a new industry are being investigated, including sampling and screening native germplasm, perfecting farm management and harvesting protocols, nut processing, and market development. However, one of the major hurdles is not having a commercially reliable clonal propagation protocol. Considerable trials investigating the use of stem cuttings and layerage to clone selections from the wild has been done by cooperators at the University of Minnesota but these efforts so far have not shown a clear route useful at the commercial level. Thus the two universities and KHN were asked to investigate if micropropagation might meet this need. Please note that this work in not complete yet so the observations I present here are just preliminary.

To have a practical micropropagation protocol, at least four stages must be met: isolation of tissue in a sterile environment, stabilization of tissue for growth in microculture, production of high quality microshoots that will provide microcuttings, and microcutting rooting and acclimation to greenhouse environments.

For isolation, the first source of plants was field plantings of native swarms that had been selected over several years of observation for nut productivity. Note in Figure 1 that these plants were showing flower buds and thus most of the shoots were adult. Even non-flowering shoots from these plants were difficult to sterilize and did not perform in microculture (Figure 2). Thus several approaches at rejuvenation were attempted. Divisions of field plants were taken and grown in pots in a greenhouse. Tissue samples taken from growth of flowering shoots again were not successful in microculture. However, when suckers from rhizomes or the collar region (Figure 3) were sampled, more successful sterilization and establishment in microculture was achieved (Figures 2 and 4). More than 50% of the 18 clones selected for propagation were successfully isolated in microcultrure.

Figure 1. An American hazelnut plant in a field of individual selected plants that were obtained from wild swarms for their general productivity. Note the shrub growth form and the flowering of the two year old stems.

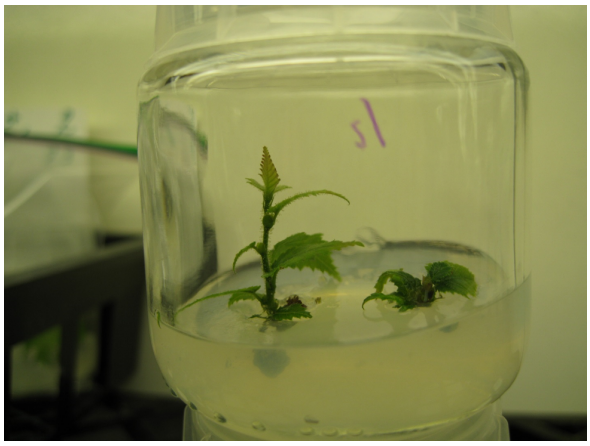

Figure 2. New stem pieces isolated from a highly adult hazelnut stock plant (right) and a non-flowering (juvenile) stock plant sucker.

Figure 3. A division of a field American hazelnut plant potted and growing in a greenhouse. Shoot on the right is from a one year old stem and will set flowers in the fall; shoot on the left is a non-flowering (juvenile) shoot originating from a rhizome or collar bud.

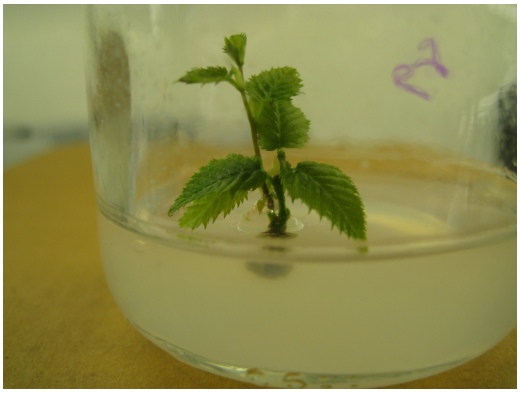

Figure 4. A subcultured microshoot where the original shoot (right) has stopped growing and a new, more vigorous and continuously growing (juvenile?) shoot has emerged.

Continued and more vigorous microshoot growth of tissue was successful in establishing (stabilizing) growing shoot cultures (Figure 5). During 3 to 6 months of subculturing growing microshoots, the emergence of basal shoots (Figure 4) was often evident; such shoots continued more active growth on subsequent subcultures than did the subcultures of the original shoot from which they emerged. This vigorous microshoot growth from the base of established shoots visually resembled the emergence and growth of shoot suckers with greenhouse grown stock plants. With 8 clones, microshoots suitable for use as microcuttings (Figure 5) were obtained.

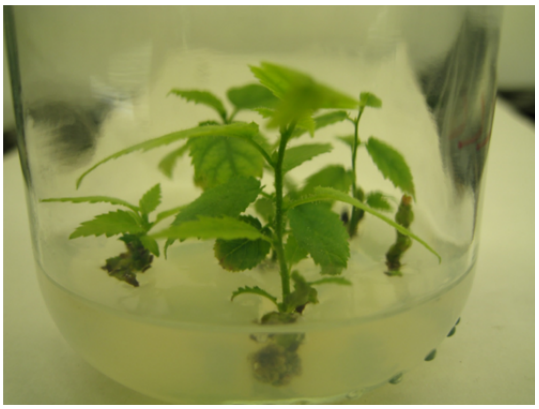

Figure 5. A stabilized shoot culture of an American hazelnut selection. The larger shoots are appropriate for use as a microcutting.

For rooting/acclimation, 1-month-old microshoots were harvested from the shoot cultures, and treated with water-soluble IBA (1000 ppm) dips before sticking in soilless mix. Microcuttings were exposed to 18 h of fluorescent lighting in 1020 flats covered with clear plastic domes. Rooting and or callusing was evident as new leaf regrowth became apparent. Such cuttings were acclimated by slowly removing the plastic dome under the rooting environment. Surviving microplants were potted and moved to the greenhouse.

Unfortunately, the losses incurred during rooting, acclimation, and greenhouse culture were high, with less than 20% of the original microcuttings surviving as rapidly-growing liners. Although roots often formed on 30-50% of the microcuttings, this was usually associated with prominent callusing (Figure 6). When such microplants were moved to more stressful environments (greenhouse), over 90% of the microplants stopped growing and gradually succumbed.

Figure 6. A newly rooted American hazelnut cutting showing significant callus ball at base.

With the general lack of success in both microcutting and the earlier stem cutting trials, at this point we became curious about the innate capability of American hazelnut cuttings to regenerate adventitious roots. To explore this question, we grew seedlings of selected swarms of wild hazelnuts and harvested softwood stem cuttings. After treating with 1000 ppm of soluble IBA, 50 to 100% of the cuttings rooted and the resulting potted plants continued vigorous growth (Figure 7). Similar cuttings taken from more established stock plants in the greenhouse were largely unsuccessful and usually only produced massive callus balls at the base of the stem. Interestingly, similar softwood cuttings taken from rapidly growing plants originating from micropropagation also showed a high capacity to root and successfully acclimate (Figure 8).

Figure 7. Cuttings of American hazelnut during acclimation and early growth in a greenhouse. Cuttings on right are from young seedling stock plants and on left from microcuttings. Note the non-uniformity and deterioration of many of the microcutting-generated plants.

Figure 8. Young plants of American hazelnut from two different sources. Right is a microcutting and left is a cutting from a microcutting-generated stock plant similar to the one on the left. Size of plants is just an indication of differing ages.

DISCUSSION AND CONCLUSION

These early attempts to propagate American hazelnut native germplasm selections using cuttings were frustrating but enlightening:

- Working only with juvenile tissues was critical. Seedling cuttings demonstrated a high capability to root that was not evident in softwood cuttings from more adult

stock plants.

- Establishment of successful microshoots in culture only reliably occurred using source tissues from juvenile growth (such as suckers).
- As has been observed with other micropropagation protocols, continued subculturing of actively growing shoots in microculture seems to lead to further rejuvenation which has been hypothesized as a major part of the "stabilization" phase of establishing a micropropagation protocol.

So where do we go from here with the cloning of American hazelnut via micropropagation? One approach that is being explored is the combination of micropropagation and stem cutting propagation. Although micropropagation has so far not proven commercially successful, it does generate useful and apparently highly juvenile stock plants from which stem cuttings with a high capacity to root can be obtained. Our approach may be to annually generate juvenile stock plants via micropropagation and use these to produce multiple generations of softwood cuttings (Figure 7).

SUMMARY

With our initial trials of generating a cloning technology for American hazelnut germplasm, the general recalcitrance of this species to regenerate roots was evident. The importance of maintaining a juvenile state of the stock used for either cuttings or micropropagation seems critical. Fortunately maintenance of the juvenile state by use of micropropagated stock plants offers an approach to overcome this limitation to the development of this industry.

From hands and feet to robots and spreadsheets©

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INTRODUCTION

It is said "necessity is the mother of all invention," and for our company this is very true. Although we didn't invent anything, certain economic situations made it necessary for us to reinvent our production methods. The economic slowdown that started in late 2008 and dragged out for many years set a new course for Decker's Nursery that we are still traveling. This is the journey we took to mechanize our company.

To better understand the journey, it will help to understand our company. Decker's Nursery was founded in 1921 by Paul Offenberg. He was a professionally trained Horticulturalist from Holland who immigrated here and used his skills to start Paul Offenburg Nursery. Through hard work and a lot of effort, the nursery grew, relocated a couple of times and reorganized into the company that is Decker's Nursery today. The Nursery focuses on propagation and wholesale nursery production.

Decker's Nursery can be broken down into three main departments: container, liner, and field production. The field department produces B&B trees and evergreens on around 90 acres for a very local customer base. The liner department is a national supplier of 4- and 2.5-in. pots and ships to 36 states and Canada. The container department is a regional supplier growing on 26 acres. We grow mainly #1, #3, and #7 for local independent garden centers and landscapers. We carry around 200,000 #1, 225,000 #3, and 15,000 #7 in production and sales for a complete year.

The economic slowdown affected every department in very different ways. The field department went from growing on over 100 acres to growing on less than 60 acres. The liner department actually maintained sales through the slow down by partnering with different introduction companies. The container department lost about one third of its sales. As a whole we dropped around 29% in overall sales.

As a cost saving measure we cut labor … and cut labor … and cut labor. We went from 58,000 work hours at the peak of our pre-recession sales to 29,000 work hours at the bottom of our sales during the recession. We lost one third of our sales and half our labor from 2007-2011. In 2011, we decided we were not going to be able to save ourselves into prosperity. We needed to increase sales and increase production. The general lack of available labor made it impossible to go back to pre-recession practices so we needed to rethink everything.

REDUCING AND MAINTAINING LOW LABOR COSTS

Reducing and maintaining low labor costs while increasing plant quantity and quality was the foundation for all our decisions. In the spring of 2011 we invested a lot in updating equipment and new production techniques in the nursery. We bought a flat filler, an EZ trimmer, and conveyors for the liner department. The container department got a shape trimming machine and a set of pot forks … and so the journey begins.

When we got the pot forks we didn't quite understand what we were setting ourselves up for. The first major problem we ran into was our pots. The pots we were using originally didn't work with the forks. There was a misunderstanding when we bought the forks, and we quickly realized our blow-molded pots would not work. We needed to change all our pots over to a different style, one that had a hard rim around the top for the fork to catch. The problem was exaggerated because of the recession. We had thrown away tens of thousands of plants over 2 years saving all of the pots. We saved so many pots; we didn't buy any 3-gal containers for almost 2 years.

After we got the correct pots and could use the forks to pick them up, things really

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started to move forward quickly. The next problem we had was keeping the pots in rows on the trailers. We use a 4×12 ft tracking wagon made by Mitchell Ellis Equipment to move pots around the nursery. The problem was the pots would shift around after the wagons were moved. This made it impossible to slide the forks back in between the pots and pick them up.

We did try a couple of different theories but ended up building a grid system to set into the trailer. Using some old metal tree stakes (we weren't using because our field production down sized) we created a grid pattern 4×12 pots for #3 that we would set the pots into and they would hold in place even after the wagons have moved around. Little did we realize that we created the pattern to which most of the nursery has been or will be altered to match.

We had the pot forks, the correct pots, and the wagons to move them around. Now we needed the correct equipment to use the pot forks. We owned a New Holland and Bobcat skid steers so we tried both of those with no luck. We rented fork lift that didn't work. The problem was the four wheel design. It would leave ruts in the gravel areas when the operator would turn in tight places. So we made another investment in a Trike forklift.

The Trike forklift has many features that make it successful with the forks. Its open design allows for unobstructed view for the operator. The quad front wheels act as a roller and smoothes out gravel. The three wheel design gives it a zero turn radius without leaving ruts behind and the hydrostatic transmission allows for smooth starts and stops. The Trike forklift was made to work with the pot forks and it really does.

The next few steps we took were more about increasing the efficacy and versatility of the Trike forklift in our nursery. During the beginning of the economic downturn we quickly realized we had more over wintering storage capacity than we had summer time growing space. Until we decided to mechanize it never really mattered. But once we saw the future, we knew we needed to have as much open space that we could get. This would allow for Trike forklifts, trimming machines, and robots to move freely around. So, early in the process we made efforts to cut down as many houses as we could. We also have started a practice of overwintering many items outside to further decrease the need for houses even more.

Along with having unobstructed spaces we wanted the ability to move freely across those spaces; which can cause some real problems with most irrigation systems. Working with Netafim we implemented a completely new irrigation system in the nursery using Oval Tube and Meganet™ irrigation nozzles. Oval tube acts much like a larger fire hose that inflates when in use and flattens when it is empty. The tube is light weight, easy to install, and is highly versatile. With oval tube, we can drive our wagons, trimming machines and Trike forklifts around the open with no physical obstructions.

So we have the pots, we have the forks, we have the wagons with grates, we have the Trike forklift, and we have increased open spaces and now we were ready for the largest step of all … robots. For a couple of years, we realized this was the ultimate goal. After Brian Decker saw a demonstration at Willoway Nursery we envisioned this being implemented and working with all the other systems to really maximize our nursery production. All of the steps we made toward mechanization, we made with this final step in mind.

This spring we leased four robots from Harvest Automation for 3 months as a trial. They actually sent five to make sure we had at least four that would work for us. We received them in late February and started to play around with them inside before spring started. The robots work on a two-wheel system with a large roller acting as a very high tech three wheeler. Two paddles in the front act as gripper that open and close to pick up the pots. Five different electronic eyes help guide and read distances to pick up or set down the pots. There is a reflective tape that is used to act as home base for the robots to read distance and directional orientation.

There is wide range of programing options: pot size, spacing distance, spacing patters, etc. Harvest Automation also gave us a spread sheet to help understand our spaced block size, our un-spaced block size and the frontier. The frontier is the distance the robot travels to pick a pot up and set the pot down. Managing the frontier is very important to maximizing the efficiency of the robots. If the frontier is to short, the robot spends too much time pick up and setting down plants. If the frontier is too long, the robot spends too much time driving.

Early in the spring we laid out long runs of un-spaced pots in our three bay wide

system for the robots to come back in later to space. Ideally, a company would have four or more robots working with one operator. The operator is there to watch the robots and fix any issues that might happen. Certain actions can cause a fault in the robot which needs to be reset by pulling the magnetic flag on its back. If your frontier is too close or too far, if it is too close to another robot, if it bounces up and the electric eye loses the reflective tape it will cause faults that would need to be reset.

Once a week you download the data from each robot and send it to Harvest Automation and they send it back to you as spreadsheets. The one thing I can't stress enough about Harvest Automation's is their commitment to customer service. The amount of information you get from the spread sheets is amazing. Things as simple as number of pots moved in one hour, day, or week to average frontier and faults. They also work with you to help you understand the data and how the use the data to improve efficiency.

The robots are a great display of where our industry is heading, but it's not the place Decker's Nursery is at currently. We decided at the end of our lease to return the robots and invest in other structural and mechanical improvements. There were many reasons we made the decision we did but I'm sure this is a decision we will have to revisit in the future.

TO REVIEW

- We changed our pots and limited new pots to only "wide rim" style.
- We modified our wagons to hold the pots.
- We invested in two Trikes.
- We have bought or made multiple sets of pot forks for #1, #3, and #7 containers.
- We improved our irrigation and fertilization methods to increase efficiencies with our new systems.
- We trialed robots but decided against them.
- We look forward to improve all structures to be Trike compatible.

In conclusion, our ultimate goal was met. We were able to cut labor cost and increase our sales. In 2014 our sales were up by 15% over the pre-recession high of 2007 but our labor hours were down 43% from the same time (Figure 1). Our journey through this process had many twists and turns, but was successful and is far from over.

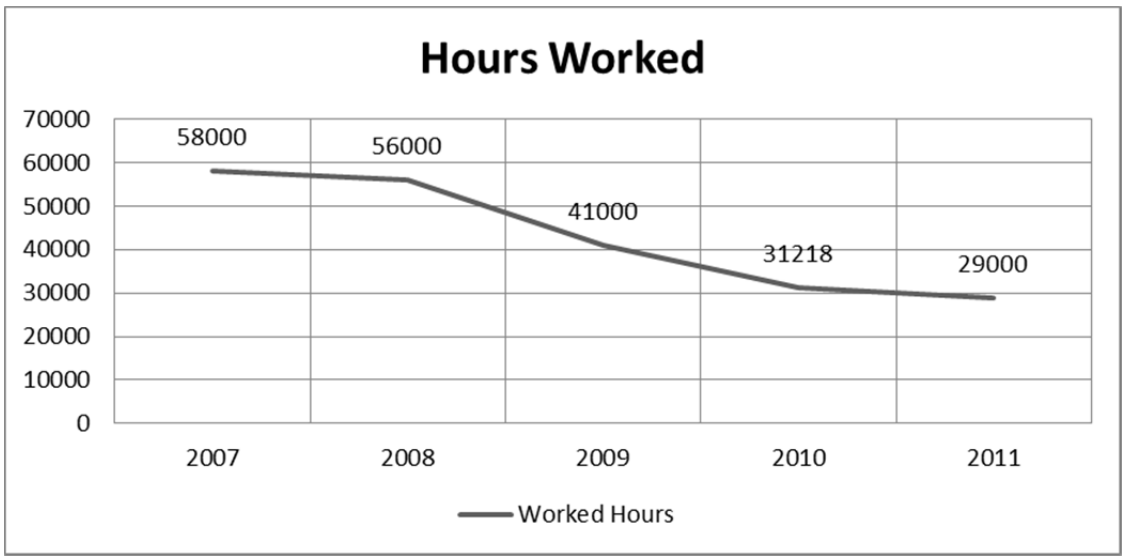

Figure 1. Labor hours from 2007 to 2011.

Winter is coming: protecting container nursery stock from adverse weather events©

M. Emmons[a]

Prides Corner Farms, Inc., 122 Waterman Road, Lebanon, Connecticut 06249, USA.

INTRODUCTION

The fall can be a great time to take a step back from the pressures of the growing season and to reflect on the year. The plants are dormant, pressure to water constantly has been reduced substantially and the extreme cold weather that will be coming has yet to arrive; time to take a deep breath. But any sense of tranquility soon disappears when the realities of winter and what it can bring comes forward. Prides Corner Farms (PCF) goes to great lengths to ensure that our nursery stock is adequately protected. And it does not stop with winter protection. Our plants are under assault throughout the year from events created by Mother Nature. Talking about the weather is not enough. Preparing for what she dishes out is important. Let's start.

OVERVIEW

Since 2011 in Connecticut, we have experienced two hurricanes, a substantial snowfall in October, record winter snowfalls, and a drought of considerable duration. In 2011 alone there was record snowfall in the winter, Hurricane Irene in August, a snowstorm in October (8 weeks after Irene) that left 800,000 electric customers without power in Connecticut, and a Cylindrocladium outbreak (Boxwood Blight) that forced us to destroy thousands of plants. What a year. Many of these events can be managed and any problems mitigated just as long as you adequately prepare.

Drought

Water is a finite resource that very few nurseries have the luxury of consuming without the risk of restrictions. During time of drought it is important to have a plan that allows the nursery manager to stretch out his or her water supply without compromising the quality of the plant material. The most important thing to remember about drought is realizing you are in one before it is too late. Prides Corner Farms has developed what we call "Water Conservation Levels" to guide us through extended dry periods during the growing season. These levels are explained in detail here:

1. Level 1 water alert.

No current restrictions are needed. Ensure that the system is running efficiently and that leaks and clogged irrigation heads are dealt with in a timely manner.

2. Level 2 water alert.

Voluntary 25 to 33% reduction in water consumption is requested. All leaks and plugged heads are to be cleaned immediately. Watering should be looked at very carefully to make sure we are not wasting any water.

3. Level 3 water alert.

A 25 to 33% reduction in water consumption is mandatory. Managers and supervisors must monitor water consumption carefully to ensure that the reductions are being implemented. Twenty-five to 33% of the nursery must have their watering needs completed by 9 a.m. No zone or growing area can run unless leaks and plugged heads have been addressed and fixed.

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4. Level 4 water alert.

A 33 to 50% reduction in water consumption is mandatory. Fifty percent of the nurseries watering needs must be completed by 8 to 9 am. Washing vehicles on the nursery is prohibited. Blocks of plants that are not full should be consolidated to water more efficiently. No zone or growing area can be run unless all leaks and plugged heads have been dealt with. Managers and supervisors must be directly involved with any and all watering decisions.

5. Level 5 water alert.

We are in imminent danger of running out of water in 10 days to 2 weeks or less. Water reductions of 50 to 75% are mandatory. As many plants as possible must be consolidated to reduce the area of the nursery that needs water. Seventy-five to 100% of the nursery's watering needs must be completed by 8 to 9 a.m. Managers and supervisors must take complete control and responsibility for watering needs. Washing of any equipment on the farm is prohibited.

Over-wintering strategies

1. Temperature protection.

Great effort goes into ensuring that the plants grown by Prides Corner Farms are protected from any winter weather event. Whether that event is extreme cold or substantial snow, we need to be able to react in a way that mitigates any damage. More than half the plants grown by PCF would be difficult, if not impossible to over-winter without some form of additional protection. The nursery is located in USDA Zone 6a where the average annual extreme minimum temperature is -10 to -5°F. Prides is in USDA Zone 6a: the average annual extreme minimum temperature is -10 to -5°F. Fortunately there are many plants that do just fine with minimal attention; that is placing them in an over-wintering structure and covering them with a white sheet of over-wintering film is adequate for their survival. Here are the plants that survive with minimal protection (Table 1).

Table 1. Plants that survive with minimal protection.

Amelanchier	*Sorbaria*
Aronia	*Spirea*
Betula	*Symphoricarpus*
Callicarpa	*Taxus*
Chionanthus	*Thuja*
Forsythia	*Viburnum dentatum*
Hamamelis	*Viburnum opulus*
Hydrangea paniculata cultivars	*Viburnum rhytidophyllum*
Hydrangea arborescens and cultivars	*Viburnum trilobum*
Ilex verticillata and cultivars	*Wisteria*
Juniperus (most)	*Sambucus*
Malus	*Salix*
Philadelphus	*Rhus*
Physocarpus	*Rhododendron catawbiense* taxa
Picea	*Rhododendron* 'PJM'
Potentilla	

Then there are the plants that we feel require additional protection as shown in Table 2. There are various reasons for protecting these plants. As shown some plants have fairly high root kill temperatures and need additional protection to protect the roots specifically. These plants are indicated by the letter "R". Still others require protection from leaf desiccation, indicated by the letter "D". Some plants do fine through the winter but can be a

challenge during the transition period between winter and spring waking up too early, indicated by the letter "T". The letter "H" refers to heated house.

Table 2. Plants requiring additional protection

Abelia (H)	*Hypericum patulum* 'Hidcote' (T, H)
Acer palmatum cultivars (R)	*Ficus* (H)
Evergreen azaleas (D)	*Ilex* × *meserveae*, *I. crenata* cultivars (R, D)
Buddleia (R, T, D)	*Kalmia latifolia* cultivars (D)
Buxus (T)	*Leucothoe* (D)
Calluna and *Erica* (D)	*Magnolia* (R)
Caryopteris (R)	*Myrica* (D)
Chaenomeles (T)	*Osmanthus* (H)
Chamaecyparis pisifera 'Filifera Aurea' (D)	*Pieris* cultivars (D)
Clethra alnifolia (D)	*Rhododendron* 'Scintillation' (R, D)
Cornus florida (R)	*Rhododendron* 'Capistrano' (R, D)
Cotoneaster (R)	*Rhododendron* 'Purple Passion' (D)
Cytisus (D)	*Rhododendron* certain lepidotes (D)
Deutzia gracilis 'Nikko' (T)	*Rhododendron* yakushimanum cultivars (D)
Hibiscus (R)	*Rosa* (R, T)
Hydrangea macrophylla (R, T, H)	*Syringa* certain genera and cultivars (R, T)
Hydrangea quercifolia cultivars (R, T, H)	*Viburnum dilitatum*, *V. plicatum* f. *tomentosum* (R)

All three of these winter challenges (root kill, leaf desiccation, and spring transitional problems) can be overcome successfully by using a poly blanket within the over-wintering houses (Figure 1).

Figure 1. Creating a poly blanket within the over-wintering houses.

The amount of protection given using this blanket is illustrated clearly by the digital thermometer that reflects the temperature in the house and the temperature in the house under the poly blanket (Figure 2).

Figure 2. Comparison of outside and inside temperatures under a poly blanket.

To protect the plants from root kill and winter desiccation it is important to be a good weatherman and know what kind of weather is coming in advance. Covering the plants before a sharp cold snap keeps the root balls from freezing solid therefore protecting sensitive roots and allowing plants to replace moisture during the respiration process. Continue to use the poly blanket in the spring to cover and protect plants during the transition period in the spring.

2. Snow load.

There is no worse feeling in the world than seeing over-wintering structures that have succumbed to the weight of a heavy snow load. Prides Corner Farms mitigates these heavy snow events by proactively bracing houses before any snowfall occurs. With the advice of Dr. John Bartok, Agricultural Engineer, University of Connecticut, we place a 2 in. × 4 in. × 8 ft board every 20 ft in a house. In large houses that are 26+ ft wide it is recommended that the 2 in. × 4 in. × 8 ft boards be placed under the side purlins. For the smaller 14-ft wide houses the boards are staggered at an angle (Figure 3).

Figure 3. Placement of supports to prevent snow load damage.

CONCLUSION

Major weather events are going to happen and although they can't be stopped there are ways to protect valuable nursery stock. Being a good weather man is essential. The winter of 2013-2014 was one of the coldest we've had in a long time. Protecting your stock from the most intense weather will bring huge dividends come spring. The effort is worth it. Also, with global warming there is greater fluctuation in temperatures and the transition period during early spring can be a dangerous time for many plants if adequate protection is not given.

Embracing technology and innovation at Spring Meadow Nursery©

D. Joeright[a]

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INTRODUCTION

Over the last 10 years, the team at Spring Meadow Nursery has put forth great effort to create a culture amongst our staff that embraces the use of technology, automation and new ideas to innovate and improve many aspects of our production process. The willingness to invest in new technology and automation is the first step towards achieving the goal of increased efficiencies, improved quality, and long-term profitability. Certain considerations and calculations must be made before one decides to invest time or money into a new piece of equipment in any production process.

1) Desire or necessity. Everyone desires the next new technology, but is it necessary? Would it result in a measurable improvement to particular production process? Or is it just a fancy show-piece for your friends? Any investment in a new technology must be targeted to a specific process that needs improvement.
2) Efficiency. Increasing efficiency in any process directly reduces labor costs, and increases profitability. Automation and new technologies are not always needed to increase efficiency. Significant gains in efficiency can often be achieved through simple changes in production practices, such as using "lean" principles to reduce waste, lower supply inventories, and diminish non-value added work.
3) Investment cost and payback. Investment cost must be considered along with calculated payback. Payback is the amount of time required for an investment expense to pay for itself through increased efficiency, or increased quality. Better efficiency results in reduced labor costs. Better quality results in the ability to reduce shrink and increase prices, thus sustaining or increasing profitability. A payback of 1 year or less should be an easy decision for any business. A payback of 2-5 years is still acceptable for most businesses. Gaining consensus on a payback period of 5-10 years requires more careful calculations, planning, and at least a 10-year plan for your business.
4) Maintenance. All equipment requires maintenance. It requires not only reactive (breakdown) maintenance, but preventative maintenance. Whenever maintenance hours interfere with operational hours, time and money are lost. When considering a new piece of automation, one should always try to witness a demonstration if possible, and calculate the expected maintenance involved, including preventative maintenance.

CONSIDERATIONS AND CALCULATIONS

Technology

Installing wireless internet (Wi-Fi) throughout the greenhouses has encouraged propagators, growers, shippers, and sales managers to communicate live from the field. Tablets and smart phones can be used to access desktop computers, or environmental computers. IPads allow growers to capture and send images, send/receive emails, and check voicemails. Growers can also monitor crop inventory, availability, order status, irrigation schedules, and environmental conditions (Figure 1).

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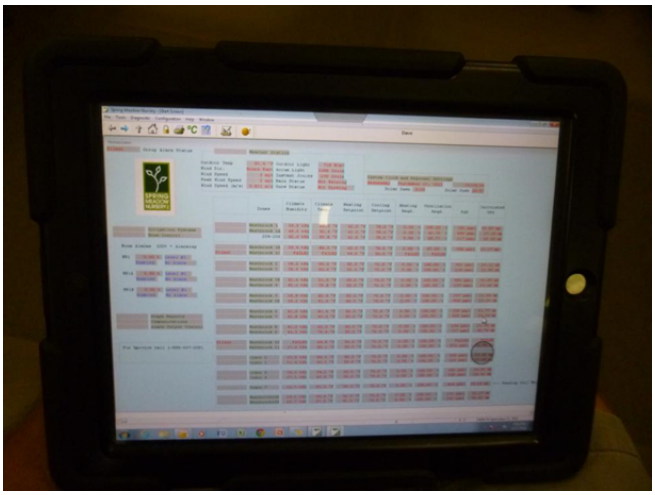

Figure 1. Wireless tablets for environmental and irrigation control.

Spring Meadow recently upgraded to Argus Controls® for environmental control of greenhouse climate and irrigation. Using VPD accumulation for propagation has proved better than time-based misting. Using tablets in the field in conjunction with Argus resulted in more accurate decision making in misting and irrigating.

Automation

In 2015 we installed a new medium mixer (Figure 2). This allowed production of a more stable, consistent medium mix with triple the output capacity of our old mixer. A stable growing medium is yielding better quality, faster finishing time, and more consistent crops.

Figure 2. AgriNomix automated soil mixer.

Many different recipes can be programmed into the machine and stored in its memory. A single push of a button (Figure 3) by the operator changes the mix for specialty crops requiring different proportions of the medium components.

Figure 3. Touch-screen control of AgriNomix soil mixer.

Grading machine

In 2007 we invested in a robotic grading machine (Figure 4) that sorts our liner trays into four possible grades based on size. Robotic gantries disassemble the trays after which each individual plant is photographed and then placed back into a tray based on its grade (Figure 5). This machine allows us to grade our plant material as it moves from propagation areas into finishing houses, as well as when plants are prepared for shipping.

Figure 4. Tuinbouw Technisch Atelier (TTA) Plugsorter grading machine.

Figure 5. Tuinbouw Technisch Atelier (TTA) Plugsorter showing final grades.

Innovation

Trimming, or pruning, is a common practice for any production nursery. In some cases it can be very labor intensive, while in others it can be automated. Trimming liners with hedge-trimmers is one method that has been used in the past at Spring Meadow Nursery (Figure 6). This was a labor intensive process where we could achieve 600 trays per man-hour.

Figure 6. Manual trimming with hedge-trimmers.

In 2008, an idea was hatched for a 24-fot wide trimming machine that could trim an entire bay of plants at once. A giant cutting blade was purchased and a prototype on wheels was constructed (Figure 7).

Figure 7. Prototype of 24-foot automatic trimming machine.

Successful tests of our prototype led to an overhead rail system designed to support heavy machinery. This rail system would ultimately be the transport method for our 24-ft wide trimming machine which was fully realized in 2010 (Figure 8). This new machine quickly proved its value by cutting over 10,000 trays per hour.

Figure 8. Completed 24-foot automatic trimming machine on rail system.

A series of rotating brushes sweep clippings across the cutting blade and onto a conveyor belt that dumps them to a sidewalk where they can be easily collected and disposed of (Figure 9).

Figure 9. Clean collection of clippings by automatic trimming machine.

A rail system designed specifically for our trimming machine soon opened the door to other opportunities for automation. Working with an outside company, Spring Meadow purchased a boom sprayer in 2014. This machine would also ride on the same rails (Figure 10). It is also 24-ft wide and travels down a bay while spraying at a 30° angle to lean plants over and contact the undersides of leaves. Once the sprayer reaches the end of a run, it automatically reverses direction and sprays again at a 30° angle from the opposite side, resulting in complete coverage of the crops and minimized exposure to applicators.

Figure 10. Automatic spraying machine using rail system.

Currently in 2015 we are trialing a 24-ft wide tray moving cart utilizing the same transport rails (Figure 11).

Figure 11. Powered moving cart using rail system.

Lean practices

Using lean manufacturing principles have helped to increase many of our efficiencies, often with little or no reliance on automation. Progressive sticking helped us to increase our sticking output by 20%. Progressive transplanting also resulted in a 20% increase in output. This occurred without any additional input, just a simple change in the process. In 2015 we looked at our rail system again and began using simple platforms suspended from the rails that would act as rolling tables. Using these platforms and with help from installation of concrete floors, we were able to reduce order packing and shipping labor by 50% (Figure 12).

Figure 12. Packing orders on rolling platforms.

The idea behind these simple platforms is to work smarter, not harder. Bringing the work up off of the floor makes it more comfortable for the worker. A comfortable worker is likely going to produce better quality work and do so more efficiently than an uncomfortable worker. We also now use these devices for hand sorting (Figure 13).

Figure 13. Manual grading on rolling platforms.

We are trialing them for cutting harvest (Figure 14).

Figure 14. Harvesting vegetative cuttings on rolling platforms.

We use them for maintenance of overhead irrigation systems, and we have even tried them for weeding (Figure 15).

Figure 15. Pulling weeds from rolling platforms.

In nearly all cases, we were able to improve our efficiencies, and increase productivity. These are just a few of the areas where we have been successful at innovating and implementing technology in the last 8 years at Spring Meadow Nursery.

New Plant Forum©

Compiled and Moderated by C. Tubesing
Presenters:

J.R. Ault[1,a]

[1]Director of Ornamental Plant Research, Chicago Botanic Garden, Glencoe, Illinois, USA.

Phlox × *procumbens* 'Pink Profusion' PP#25,883
Phlox 'Violet Pinwheels' PP#25,884
Tradescantia 'Tough Love' PP#25,988

B. Hendricks[2,b]

[2]Klyn Nurseries, Perry, Ohio, USA.

Cephalanthus occidentalis Magical® 'Moonlight' buttonbush
Penstemon calycosus
Syringa reticulata subsp. *pekinensis*, 'WFH2', Great Wall™ Peking tree lilac

B. Horvath[3,c]

[3]Intrinsic Perennial Gardens, Inc., Hebron, Illinois, USA.

Allium 'Windy City' PPAF
Festuca 'Cool as Ice' PPAF
Geum 'Citronge' PPAF
Sedum ellacombianum 'Cutting Edge' PPAF

M.D. Yanny[4,d]

[4]JN Plant Selections, LLC, Milwaukee, Wisconsin, USA.

Aesculus glabra 'J.N. Select', Early Glow™ Ohio buckeye
Juniperus chinensis 'J.N. Select Blue', Star Power™ Chinese juniper
Juniperus virginiana 'J.N. Select Green', Emerald Feather™ eastern redcedar
Spiraea fritschiana 'J.N. Select A', Pink-a-licious™ Fritsch spirea

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***Aesculus glabra* 'J.N. Select', Early Glow™ Ohio buckeye**

Figure 1. *Aesculus glabra* 'J.N. Select', Early Glow™ Ohio buckeye.

Early Glow™ buckeye is a seedling selection found by Michael Yanny in 1981 (Figure 1). The original tree, at about 30 years old, is approximately 30 ft tall × 20 ft wide. Early Glow™ buckeye gets its name from its bright red fall color in mid to late September in Southern Wisconsin. It is typically the first tree to get fall color each year at Johnson's Nursery. Its form and growth rate seem to be similar to *A. glabra* seedlings, though it does show better late season foliage quality than straight species plants. Early Glow™ buckeye produces very few seeds giving it great potential as a street tree. It seems the reason for the near seedless nature of the tree is that fewer pistils elongate enough to be easily pollinated.

Grower's licenses are available from Chicagoland Grows® Inc.

***Allium* 'Windy City' PPAF**

Figure 2. *Allium* 'Windy City' PPAF.

An Intrinsic Introduction. Dark green glossy foliage reaches 15 in. tall and wide (Figure 2). Dark rose purple flower clusters of sterile flowers begin to color in June and open in July on 18-20 in. stems. Full sun is best. Average well drained to dry soil. Very drought tolerant.

***Cephalanthus occidentalis* Magical® 'Moonlight' buttonbush**

Figure 3. *Cephalanthus occidentalis* Magical® 'Moonlight' buttonbush.

Cephalanthus occidentalis or buttonbush is a native shrub growing naturally in bogs or in moist areas to a height and width of 6-10 ft. Magical® 'Moonlight' buttonbush is a distinctive, compact form with slightly smaller glossy leaves and a mounding habit to about 5 ft height and wide (Figure 3). The distinctive globular white flowers appear in mid summer and are hummingbird and butterfly magnets. A perfect choice for the rain garden or bioswale yet readily adapts as an ornamental shrub in average garden soils.

***Festuca* 'Cool as Ice' PPAF**

Figure 4. *Festuca* 'Cool as Ice' PPAF.

This one was selected for its lighter green spring emergence giving the plant a slight bicolor look and better heat tolerance in summer. Vigorous plants turn blue in summer. Flower stems reach 18 in. with foliage reaching 24 in. wide. Zone 4-8. Full sun to light shade, well drained soil is best (Figure 4).

***Geum* 'Citronge' PPAF**

Figure 5. *Geum* 'Citronge' PPAF.

Creamy orange flowers emerge in May from red stem and buds on 18 in. stems (Figure 5). Wide folded petals overlap giving a nice full effect. Heavy blooming plants have some reb-loom too. Full sun, moist rich soil. Zone 4-8.

***Juniperus chinensis* 'J.N. Select Blue', Star Power™ Chinese juniper**

Figure 6. *Juniperus chinensis* 'J.N. Select Blue', Star Power™ Chinese juniper.

Selected at Johnson's Nursery in1998 by Michael Yanny from a crop of open-pollinated *J. chinensis* seedlings. A 12-year-old plant is 13 ft tall × 5 ft wide (Figure 6). The plant gets its name from the beautiful blue-green, star-like juvenile foliage which gives it a delicate, almost sparkling texture. It maintains the juvenile foliage for about 15 years then slowly begins to develop scaly, non-star-like, soft, bright green, adult foliage. In addition, it begins bearing silvery-blue berries (cones) at this time. They are attractive to birds. It is an extremely fast growing cultivar even surpassing *J. chinensis* 'Mountbatten' by 1 ft of growth on 6-year-old field-grown plants. This is an excellent plant for use as a screen where deer are a problem.

Grower's licenses are available from JN Plant Selections, LLC.

Juniperus virginiana 'J.N. Select Green', Emerald Feather™ eastern redcedar

Figure 7. *Juniperus virginiana* 'J.N. Select Green', Emerald Feather™ eastern redcedar.

Selected at Johnson's Nursery in 1998 by Michael Yanny from a block of seedlings of open pollinated *J. virginiana* 'Canaertii'. A 12-year-old plant is 12 ft tall and 5 ft wide (Figure 7). The plant has an upright, ascending branching habit. Emerald Feather™ grows very fast compared to other selections in commerce and doesn't require staking. The plant has a fresh, bright green color that makes an excellent back drop for plantings of flowering shrubs and perennials. It is an excellent plant for screening purposes. The plant has tiny, silver to blue berries that are relished by birds. Emerald Feather™ performs best in well-drained soil in full sun. Its resistance to deer browsing is presently unknown.

Grower's licenses are available from JN Plant Selections, LLC.

Penstemon calycosus

Figure 8. *Penstemon calycosus.*

Penstemon calycosus is an herbaceous 2-3 ft perennial native to the eastern USA. The plant is very adaptable as a garden plant performing far better in eastern gardens than the western natives. It grows well in light shade to full sun in moist but well drained to dry soils. flowering in late spring to early summer (Figure 8). Flowers can range in color from light violet to purple and are produced on terminal panicles against a background of glossy green lance-shaped leaves. The clone we have chosen has dark bluish-purple flowers. The plant is easily propagated by division or softwood cuttings.

***Phlox ×procumbens* 'Pink Profusion' PP# 25,883**

Figure 9. *Phlox* × *procumbens* 'Pink Profusion' PP# 25,883.

'Pink Profusion' definitely lives up to its name as our trial plants have bloomed for as long as 8 weeks, commencing in mid to late March and extending well into June in northern Illinois (USDA Zone 5) (Figure 9). The flowers are quite large for a *P*. ×*procumbens*, 1 and ¼ inch wide with broad overlapping petals, and the oversized blossoms can completely cover the plant at peak bloom. The petals are an attractive deep purplish pink, and the flowers have a conspicuous deep reddish purple center eye surrounded by a white halo. Two year old plants grew to 13 in. wide × 8 in. tall in bloom and 3 in. tall out of bloom. Like other *Phlox* ×*procumbens* selections, 'Pink Profusion' produces a dense mass of central stems in the spring, which after blooming become lax on the ground and may or may not layer in to form irregularly spreading mats. Plants may die back during the summer, but will produce a mass of blooming stems anew the following spring. Best grown in full sun to partial shade and on a moist, but well-drained soil amended with organic matter. It is readily propagated by shoot tip cuttings taken from new growth in spring to early summer. Developed by Jim Ault at Chicago Botanic Garden from a cross made in 2007 between *Phlox* 'McDaniel's Cushion' and a pink-flowered seedling of *P. stolonifera*. USDA Zones 6-8 (5b with protection). A Chicagoland Grows® Inc., plant introduction.

***Phlox* 'Violet Pinwheels' PP# 25,884**

Figure 10. *Phlox* 'Violet Pinwheels' PP# 25,884.

This delightful hybrid phlox breaks new ground in the spring blooming moss phlox genre. The notched, upturned petals truly look like they could take flight with a good breeze, hence the "violet pinwheels" cultivar name (Figure 10). The ¾ inch-wide flowers open a vivid purple color and age to a still vivid violet color, rare colors among the moss Phlox. We have observed 5 weeks of bloom commencing as early as late March and extending as late as mid-May, spring temperatures depending, in northern Illinois (USDA Zone 5). The plants consist of low, slowly spreading mounds of dark green needle-like foliage that is soft to the touch. Three-year-old plants have grown to 18 in. wide × 4 in. tall and have been evergreen year around. This plant is a perfect marriage of eastern and western phlox, combining unique beauty with adaptability to heat, cold, alkaline and saline soils. It is best grown in full sun on well drained soils and with an adequate water supply. 'Violet Pinwheels' is readily propagated by shoot tip cuttings taken from new growth in spring or autumn. This plant was developed by Jim Ault at Chicago Botanic Garden from a cross made in 2008 between *P. bifida* and *P. kelseyi* 'Lemhi Purple'. USDA Zones 4-8. A Chicagoland Grows® Inc., plant introduction.

***Sedum ellacombianum* 'Cutting Edge' PPAF**

Figure 11. *Sedum ellacombianum* 'Cutting Edge' PPAF.

Yellow edged, bright green foilage on mounding plants look good from spring to fall providing a bright contrasting plant for full sun to part shade (Figure 11). Well drained soil.

***Spiraea fritschiana* 'J.N. Select A', Pink-a-licious™ Fritsch spirea**

Figure 12. *Spiraea fritschiana* 'J.N. Select A', Pink-a-licious™ Fritsch spirea.

Pink-a-licious™ Fritsch spirea originated from a selection made by Michael Yanny from a crop of open pollinated seedlings of *Spiraea fritschiana* started in 2000 at Johnson's Nursery. The likely pollen parent is *S.* ×*bumalda* 'Norman'. Unlike its white flowered mother, Pink-a-licious™ has abundant purplish, pink flat-topped clusters of flowers in June (Figure 12). This cultivar has a wonderful, compact habit. It grows to 2-3 ft tall × 2-3 ft wide in Southern Wisconsin. The fall color on this plant can be outstanding, being a combination of the colors of a fruit salad. Often times the plant will have fall colors of pineapple yellow, watermelon pink, honey dew chartreuse, and cantaloupe orange all on the same plant at the same time. Pink-a-licious is easy to grow, requiring full sun to partial shade, and is tolerant of a wide range of soil types.

Grower's licenses are available from Chicagoland Grows® Inc.

***Syringa reticulata* subsp. *pekinensis*, 'WFH2', Great Wall™ Peking tree lilac**

Figure 13. *Syringa reticulata* subsp. *pekinensis*, 'WFH2', Great Wall™ Peking tree lilac.

This is a distinctive Peking lilac with an upright form, ascending branches and cherry-like exfoliating bark and crisp, dark green foliage maturing into a 15-20 ft small tree (Figure 13). Clusters of pure white flowers emerge about 10 days earlier than *S. reticulata*. The distinctive form makes it an ideal choice for use as a street tree. It easily develops a central leader and is a vigorous grower making up quickly to a saleable plant. It is recommended to plant field liners from potted liners rather than bare root. Licensed growers can easily propagate the tree by budding or grafting.

***Tradescantia* 'Tough Love' PP# 25,988**

Figure 14. *Tradescantia* 'Tough Love' PP# 25,988.

A new direction in spiderwort breeding! Most of the hybrid spiderworts in the

marketplace were developed using eastern species that prefer some shade and a moist soil. (Figure 14). 'Tough Love' was developed from two Great Plains species that are naturally found in full sun to light shade on dry rocky, clay, to sandy soils. Try 'Tough Love' in a challenging dry site and see how it performs for you. While best on dry sites, we have found it tolerates a wet clay soil as well. It has performed very well on the trial roof garden at Chicago Botanic Garden. The one inch wide, vivid reddish-purple flowers are borne in great profusion in May, literally covering the centers of the plants. Like all spiderworts, each flower lasts but a day. Plants will repeat bloom through late August. Unlike many spiderworts, 'Tough Love' tends to remain evergreen through the summer months, or if severely stressed, the foliage may disappear but is replaced in autumn. Clump-forming with broad, daylily-like, leathery foliage. Two-year-old plants from division were 11 in. tall × 16 in. wide at peak bloom, making this one of the more compact selections in the marketplace. Easy to propagate by division in early spring or autumn. Developed by Jim Ault at Chicago Botanic Garden from a cross made in 2006 between an open-pollinated hybrid seedling of *T. tharpii* and the species *T. occidentalis*. USDA Zones 4-8. A Chicagoland Grows® Inc., plant introduction.

Light-emitting diode lights can make rooting cuttings easier and safer©

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INTRODUCTION

Bailey Nurseries in St. Paul roots over 9 million cuttings every year. This includes a portion grown from tissue culture. These micro-cuttings started in January are the *Syringa vulgaris* hybrids (frequently called the French hybrids lilacs). These micro-cuttings lilacs are shipped to Minnesota from a tissue culture laboratory in Oregon.

Minnesota in winter is not the ideal place to root micro-cuttings in a greenhouse. Cold temperatures, low humidity, and low light conditions make rooting cuttings a real challenge. The current method, which uses small tents to better control the environment, yields variable results, and the cuttings require a lot of labor to maintain.

When we began reading about how European growers were using light-emitting diode (LED) lights to root cuttings, it piqued our interest and several questions came to mind. Here are some of the key questions:

- Where can we install some LED lights?
- Will they produce a good crop?
- Which crops will benefit from this system?
- Will they simplify grower care and improve rooting?
- Will they last in the environment in which we'll eventually put them?

METHODS AND MATERIALS

In 2011, we worked with Philips and Hort Americas [our supplier and technical support – the contact info is Chris Higgins (chiggins@hortamericas.com)] to design a propagation chamber using LED lights in one of the buildings.

We started the initial trial in February 2011. We partitioned a section of the germination chamber. This chamber provides a constant 75°F temperature in the winter, has light provided by 8 ft-long fluorescent tubes, has misting capacity (fog nozzles suspended from the ceiling), and readily available electricity. The tent we created was partitioned from the rest of the chamber with black and white plastic to avoid light contamination from the fluorescent lights (used for germination). We used three Cannon carts tied together side-by-side to form one large shelf that could hold up to 15 trays. The light source under this tent was provided by five Philips GreenPower LED production modules made of 70% deep red and 30% blue lights. The distance from the shelf to the module was 15 in. These modules are 5 ft long, which matches the size of the Cannon carts. We had room for 15 flats.

A range of cuttings were taken from plants in the greenhouses, including *Spiraea*, *Celastrus*, *Physocarpus*, and *Hydrangea*, to name a few. The cuttings originating from the greenhouses were stuck in 38-cell plastic trays (standard size of 11×21 in.) and treated with IBA. We also added 900 micro-cutting lilacs to the LED area. These micro-cutting lilacs were grown in three 288-trays. The medium used was Preforma and without the use of IBA.

In 2012 we purchased more GreenPower LED production modules. We created and partitioned six stalls from the main germination chamber. We are now able to move the Cannon carts in and out of the chamber with total ease. Each cart has five shelves, or layers, with 15-in. spacing between shelves. The modules are mounted on the frame of the stalls and are tilted 90° towards the center of each shelf. Each shelf is lit by two modules. The light

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cycle is 16 h on and 8 h off. During the off time, the mist cycle is also turned off. During the day time, the mist cycle is controlled by a timer. The cuttings receive more mist during the first 2 days. As they acclimate and develop roots, the mist is reduced each day. A grower is responsible to check the lilacs 5 or 6 times a day. This check only takes a few minutes each time, as every element (moisture, temperature, and light) stays constant. The mist water is treated with a reverse osmosis system. This system prevents the build-up of calcium carbonate on surfaces, especially the diodes. The first roots are seen after a few days. At 10 days, most cuttings will show some roots. At 2 weeks, some roots will be at the bottom of the cell and coming out of the drain holes. This is when they will get fertilized. A liquid solution of 50 ppm nitrogen is applied over the top of the cuttings. As early as 3 weeks, but preferably at 4, the lilacs can be moved to a greenhouse. Once in the greenhouse, they are fertilized again. It is important to acclimate them to ambient light, under some shade. Full sun will burn the foliage. After a few days of acclimation, they can be planted in the sand.

This setup can accommodate a total of 150 flats. Because of the multi-layer production design, we now think in number of plants per cubic foot instead of square foot. During the winter of 2012, we rooted 16,000 micro-cutting lilacs, or 25% of the schedule. And during the winter of 2013, we rooted 66,000 micro-cutting lilacs, or 100% of the schedule. This practice was repeated in 2014 and 2015.

Our normal greenhouse growing practices require the presence of a grower every 30 min during work hours, or more often when the light level (sun/cloud) keeps changing. In this system, these tissue culture lilacs are rooted inside several small tents (24 flats each) in one of our greenhouses. This greenhouse is heated by steam pipes buried in the sand and by forced air. The tents are used to create a micro climate that is easier to control than trying to control the entire greenhouse. Depending on the level of sun intensity, on how much moisture is in the air and on how often the heaters are running, the grower in charge has to adjust: the mist cycle, which is done by hand; the amount of shade, which is a combination of different layers of plastic covering the tent; and the level of venting, which is done by opening or closing these same layers of plastic to match the outside growing conditions. This takes place all day long.

In the LED chamber, there is none of this constant monitoring. We have experienced a significant reduction of crop monitoring. The fogging system fills the entire room with fog, and for this reason, no hand misting is necessary. The fog keeps the cuttings turgid, the fluorescent tubes in the germination chamber maintain a constant air temperature of 75°F and the LED lights provide the proper light quality, intensity, and duration to promote plant growth.

RESULTS

Right away in 2011 we were able to answer several of our questions. The first year indicated that growing under LED lights is possible. We were able to root cuttings with minimum maintenance. These cuttings (*Hydrangea paniculata* and *Spiraea*) were transplanted into larger containers (quart size) and again grown under LED lights in a different tent, outside the germination chamber. We were able to root and grow and take cuttings from these plants. This second generation of cuttings was also rooted under LED light. This meant that this last generation of cuttings were plants that had never seen sunlight.

In regards to crop quality, not all crops responded equally to these new growing conditions. It was ideal for the micro-cuttings, but plants like *Rhus typhina* 'Bailtiger', Tiger Eyes® staghorn sumac for example had issues because of the high level of humidity. We experience cuttings growing roots above the soil line (because of the light, temperature, and humidity). Spireas and hydrangeas were more prone to this reaction than other crops like roses. The aesthetic value of the plugs was reduced by the presence of these roots.

The next observation on plant quality was that plants needed to be moved out as soon as they were rooted. Keeping them under these growing conditions (high humidity) was not helping. The growing conditions were promoting the growth of botrytis rapidly. Weekly sprays were required to keep this fungus under control.

Seed germination was also successful in these conditions (under LED lights) when compared to the normal conditions (fluorescent light) in the germination chamber. What was observed using impatiens seeds was that these seedlings were shorter under the LED light source than under the fluorescent light source. This can be an advantage for a bedding plant grower. Shorter plugs make for a better finished product. Germinating under LED light can also provide some safety if the seedlings are not moved fast enough to a greenhouse, as they are elongating at a slower pace under LED than under fluorescent light.

This system is not without a significant capital investment, even for a small scale (150 flats), as described in this paper. It is important to determine the benefits from this system. Here the principal goal was to increase the yield and reduce or simplify labor associated with rooting micro-cutting lilacs. The second goal was to determine if other crops would benefit from this system. The focus was put on crops that are difficult to root. The problem crops are *R. typhina* 'Bailtiger', Tiger Eyes® staghorn sumac; *Amelanchier*; *Cotinus;* or *Daphne*, for example. Several trials have been performed with mixed results. Regarding these trials, the main source of problems (causing failure) is the management of the level of moisture. The mist of this chamber is not adequate for all crops. As a result, we are seeing good and poor rooting success on the same crop. Proper management of the moisture level in the chamber is critical for success. It works for the micro-cuttings (Table 1).

Linked to high humidity level, one more observation is necessary. The Philips GreenPower LED production modules are rated to 95% humidity. Our chamber exceeds this level. Everything is wet, and droplets form on the diodes and other surfaces. From our experience, we believe water finds its way into the modules in two ways. The first is via the splicing that connects the power cords. Even though our process has improved, we still see water getting into the power cord. This is not a defect of the production modules. It is directly linked to our setup and the level of moisture in the air, which is more like 100%. The second way for the water to enter the module is around the diodes. Because water enters the module, over time it will damage some of the electronic components inside the module. These two observations result from the "extreme" environmental growing conditions, conditions that exceed the manufacturer recommendations.

Table 1. Rooting percentages — germination chamber versus tents in the greenhouse.

Cultivars	2012	2013[1]	2014	2015[2]	Tents (7-year average)
Albert F. Holden		72	92	42	61
Krasavitsa Moskvy (syn. Beauty of Moscow)		87	93		74
Charles Joly			85	93	
Common white lilac		93	94	95	87
Ludwig Spaeth		90	95		67
Madame Lemoine		96	86		71
Président Grévy			96	87	
President Lincoln		100	94	100	72
Sensation		96	85	94	80
Wedgwood Blue			88	97	
Yankee Doodle	95	51	81	45	63
Wonder Blue		95	96		83
Declaration			85	44	
Miss Ellen Willmott		89	89		75
Pocahontas				98	

[1]In 2013 the 'Yankee Doodle' arrived with cold damage (frozen). This explains the low rooting percentage.

[2]In 2015 'Albert F. Holden', 'Yankee Doodle', and 'Declaration' experienced spray damages from a contaminated backpack with herbicide residue.

When a yield percentage is missing, that cultivar was not grown that year.

SUMMARY

Growers can benefit greatly from using the right source of LED lights, for the right crop, and in the right circumstances. Here is a summary of these benefits:

Advantages:

- Increased yield
- Increased plant quality
- Reduced crop timing
- Reduced and simplified grower care
- Reduced greenhouse cost, heat and maintenance
- Freed up greenhouse space
- Accelerated propagation, and
- Propagation made possible at any time of the year, as long as a cutting source is available

Disadvantages:

- Investment cost
- Durability and reliability
- Directional light

CONCLUSION

Under the controlled environmental conditions of the germination chamber (uniform temperature, light and humidity), the Preforma plugs remain moist, the micro-cutting lilacs keep turgid, the temperature and light source is constant and the cuttings never get stressed. There is little to no grower care required in these growing conditions. Because the cuttings are not stressed as much as in the greenhouse under the tents, the yield (percent of rooted plants) is increased, and grower time is reduced.

Where ecology meets economy©

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This paper is designed to be an introduction to a discussion that will take place at this meeting between people involved with natural areas management and conservation and those doing plant propagation, plant breeding-introduction, and nursery stock production. Various papers will be presented after this one, all providing information and research results as food for the discussion.

The issue of invasive plant species has become a prominent one in the last 15 years. Invasive plants are causing destruction of natural ecosystems in many unmanaged land tracts. The amount of money spent by natural areas managers to control invasives has become a major part of their budgets. Many but not all of the plant species which have been identified as invasive originated from the ornamental plant industry. Because the industry has been a major incubator of new invasive plant species, most efforts to control the expansion of more invasive plant species have been centered on preventing new ones from entering from the industry. Many states have enacted regulations to control the sale and possession of various invasive species, while some others have instituted voluntary invasive plant control measures in cooperation with their green industries. Some states have little to nothing formally in place to deal with the problem. Federal regulations of invasive plants presently in the country are minimal at this time.

The issue has strained relations between some in both the natural areas community and the green industry. It is not uncommon to find land managers that resent the nursery industry because they see it as the source of their biggest problem. I know of numerous nursery people who have voiced their concerns about the government regulating their plant inventories. Another common complaint from both land managers and nursery people is that they want sound science to determine what should be regulated and what shouldn't.

My stance on this as a plant propagator and plant breeder-introducer as well as a producer of local ecotype native plants is smack-dab in the middle! It is a call for cooperation. The industry needs to respect and work with the land manager-conservation community to help them preserve their natural areas so that huge portions of their budgets don't continually get eaten up by invasive species control. And the land manager-conservation community needs to respect and work with the industry so as not to severely impact the businesses that are a major conduit for connecting the general public to plants and the natural world.

To me the single most important aspect of this discussion is the concept of connection. By this I mean the connection of the human population to the natural world. I believe this to be the most important thing that all of us do, conservationists, land managers, plant propagators, plant breeders, and nursery people alike. We affect the future of our world by influencing people with our plants and the environments we create and, or maintain. We provide or protect much of the beauty and magnificence that the natural world has to offer. We introduce many people to the art of growing plants and the intricacies of ecology. We are all in this together and need to appreciate that fact. We are all connected both ecologically and economically.

I think the respect aspect between the various parties is critical to making progress on the invasive plants issue. I am amazed at the knowledge I have come to appreciate from being involved with land manager-conservation people over the past 15 years. I find the use of fire as a management tool fascinating! It can save a lot of time and be very effective in controlling troublesome species both native and non-native. Many land managers know chemical control methods for some extremely difficult to control plant species such as Japanese knot weed and reed canary grass. If green industry were to work more with land

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managers, surely much could be learned to benefit their businesses.

The reverse is true as well. Nursery people can produce and grow plants like no one else. Land managers and people involved with ecological restoration can learn much by collaborating with nursery growers and propagators. Growing plants can be invaluable to understanding plant behavior in various environments. There is much to be learned by the conservation-land manager community from the nursery industry.

One of the most interesting aspects of my work is the knowledge I gain from observing plants in the wild in areas typically taken care of by hard working land managers. I learn how plants in the wild behave in particular soils and with other organisms in their environment. It is very useful to be able to see various ecotypes of plant material in their native habitat and observe their differences. It has been important to me in developing regionally adapted seed strains and cultivars for use in traditional landscaping. I value the natural areas with the same fervor that I do IPPS, libraries, and my old college professors. All are incredible sources of useful knowledge that I utilize to make a living.

As the presentations proceed today on the various subjects of invasive species regulation, sterile cultivars and their testing, conservation of endangered species, and production of native plants, I hope everyone will consider how they can help the cause of connecting people to the natural world so we can all continue to make a living at it and future generations will also. Ecology and economy can never truly be separated. We have to all work together.

Ecology—Economy
By Michael Yanny--2012

Ecology—Economy

It's a two letter difference
in language and life

Economy—Ecology
Ecology—Economy

Ecology is economical,
Survival of the fittest
Economy is ecological
Work together or go broke.

Ecology—Economy
Economy—Ecology

The economy of the world requires humans to work together through trade of goods and services. Without the goods and services the economy crumbles.

The ecology of the world requires goods and services be provided by the various biological components of the system. Without the goods and services the ecology crumbles.

Economy—Ecology
Ecology—Economy

Ecologists must understand economics.
It's a part of their science,
just like physiology and taxonomy.
Economists need to feel the natural world or they will break it.

Ecology—Economy
Economy—Ecology

Ecologists are human.

So are Economists.
They have a common life form
    and an interest in a better life.
That's good!!!

Economy—Ecology
Ecology—Economy

Life is rich
    With quarters and pine cones,
        nickels and acorns,
            flowers and dollars.
There is no reason not
    to work together
        for the common good
            of living well.

Ecology—Economy
Economy—Ecology

It's only a two letter difference
    in language and life

Ecology—Economy

Identifying invasive plant species: what plant propagators need to know about the science behind invasive plant assessment protocols©

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Abstract

Although only a very small number of introduced plant species ultimately become invasive in the United States, those that do can cause a number of harmful effects within our natural communities. Some of these invasive species are woody in nature (trees and shrubs), and these typically have a past or current horticultural connection. Thus, plant propagators of woody plant species need to remain informed of how plants are identified as invasive and which species are beginning to spread in their state. In this paper, I present additional reasons for why plant propagators should care about this issue, what they need to know about how states assess plant species as invasive, and newer issues involving cultivars that also provide unique opportunities for plant propagators. Ultimately, plant propagators are encouraged to become better engaged with efforts to assess invasive plants in their own state and to contribute to the dialog about invasive plant issues in the United States.

Keywords: assessment, cultivars, invasive species, woody

INTRODUCTION

Our world today is filled with an amazing diversity of cultivated plants, many of which are highly desired by the gardening public for traits such as flower or fruit color. Even from the earliest of times in the United States, plant explorers have been sent out throughout the world to gather seeds and cuttings of the most unusual, hardy, or sensational plants to cultivate and promote back home. More recently, global trade of commodities such as plants has become more the norm than the exception. As a result, over 25,000 plant species have been introduced to the United States since European settlement (Pimentel et al., 2005). In many cases, these plant introductions were accidental, such as when seeds hitchhike in ship ballast water or are carried along in imported soil. In other cases and especially with woody species, non-native plants have been purposely imported into the United States with the very best of intentions — whether it be promoting fireblight resistance in fruit trees in the Pacific Northwest, preventing soil erosion on road cuts in the South, or introducing fruiting shrubs for wildlife in the Northeast. Unfortunately, a small number of these non-native plants escape cultivation and spread to negatively impact natural areas across the United States, causing unforeseen and widespread effects (Sakai et al., 2001) that were never anticipated during the original introduction. These plants are known today as invasive species. According to the federal definition provided by President Clinton's Executive Order 13112, an invasive species is "an alien [non-native] species whose introduction does harm or is likely to cause economic or environmental harm, or harm to human health." In short, invasive plants can be thought of those plants that jump boundaries into natural areas, where they spread and eventually outcompete native plant species, negatively affect animals that live there, and/or alter natural ecosystem processes.

Scientists do recognize, however, that not every imported species will become invasive (Richardson and Rejmánek, 2011). Ecologists use the "Law of Tens" to talk about the potential for an imported species to spread. For example, if 1,000 plant species are imported

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into a new area, it is estimated that only 10% of those (100) may escape cultivation. Of those, only 10% will begin to establish (10) and of those, only 10% will ultimately become invasive (1 species) — and then usually only after a number of decades called a "lag period." Therefore, the term "non-native" is *not* equivalent to "invasive" because there can also be some non-native species that do not pose a threat to natural ecosystems. In addition, there may be some introduced species that are still in their lag period and have not yet shown any invasive tendencies. Furthermore, not all invasive species are non-native (despite the federal definition) as scientists recognize some native species, such as white-tailed deer, as invasive.

The fact that only a small proportion of all introduced species become problematic however, does not lessen the importance of the issue. Although it is difficult to put a dollar cost on invasive species, Pimentel et al. (2001, 2005) have estimated that invasive plant species cost at least $35 billion per year in the USA in reduced revenue from agriculture, forestry, recreation, control and removal costs, etc. Consequently, invasive species not only affect the integrity and ecology of our natural areas, but they are also quite financially costly for land managers and owners of federal, state, and private natural lands. In other words, invasive species are a concern that ultimately will affect everyone.

WHY SHOULD YOU CARE?

Why should plant propagators, especially those working with woody plants, care about invasive species? There are several reasons. First, the majority of woody invasive species have a horticultural connection in their current or past history. For example, 82% of 235 invasive woody species surveyed were used in horticulture at some point in time, even if they are no longer used today in that way (Reichard and White, 2001). These include shrub species such as Amur honeysuckle (*Lonicera maackii*) or common buckthorn (*Rhamnus cathartica*). Second, plant propagators often focus on specific traits during development because these are the traits desired most by gardeners. It turns out that these are the same traits that are found most often in invasive species (Sakai et al., 2001). For example, species that are invasive are most likely to have prolific flowering, high fruit production (often desired by gardeners for attracting wildlife), rapid growth, survival in diverse habitats, tolerance to stress, and have a history of multiple introductions. This last characteristic is important because many introductions may be necessary before a species can successfully establish within an area. For example, many European birds such as the European starling only established in the United States after they were imported and introduced multiple times to Central Park in New York City by a Shakespeare enthusiast who wanted to introduce all the birds found in Shakespeare's plays into the park (Ehrlich, 1989; Mirsky, 2008). For cultivated plants, multiple and repeated introductions typically occur as part of the large-scale production and distribution of ornamental plants to multiple points across the country. This aspect of commercial plants has been largely ignored by scientific researchers but undoubtedly plays a role in certain species invasions.

Finally, plant propagators should care about invasive plant species because it just makes good business sense. Although not as common as with herbaceous species, some woody plants can begin to seed within a landscape and become perceived as a pest species, appearing in places where it was not planted nor wanted (for example, Callery pear seedlings appearing in residential yards in southwestern Ohio). Over time, invasive plants can rapidly overwhelm a landscape aesthetically and ruin the ornamental novelty of the species so often desired by gardeners. Customers will not see the value in paying money for a plant that they can just dig up from their neighbors' yard or a park down the street. On a more positive note, plant propagators who pay attention to emerging species invasions can potentially increase their profits by anticipating future changes in product availability and offer alternatives (especially as invasives are becoming increasingly regulated in certain states). For example, breeders and propagators can begin developing sterile cultivars of species that show signs of invasiveness, thereby anticipate future demand for this type of product well before other competitors are even aware of the problem.

Will the invasive plant issue ever affect you as a plant propagator? The answer is most likely yes — if you work on woody species, you will probably encounter this issue during

your career. Highly popular ornamental plant species that are now considered invasive in one or more USA states include Japanese barberry (*Berberis thunbergii*), Norway maple (*Acer platanoides*), burning bush (*Euonymus alatus*), purple loosestrife (*Lythrum salicaria*) and Callery pear (*Pyrus calleryana*). Based on past history, it is highly likely that plant propagators today are currently developing species improvements and cultivars that will unintentionally become invasive in the future. What can be done now to prevent this from happening and ultimately help plant propagators continue to be successful in their businesses? But to even begin to answer this question, we must first ask: How do we even identify plants as invasive?

INVASIVE SPECIES ASSESSMENT PROTOCOLS

Whether a plant is labeled as invasive ultimately depends on where the plant falls along a gradient of invasion severity — in other words, "How abundant is the plant within the landscape?" and "What are the impacts of that plant within the natural ecosystem?" For example, the occasional solitary plant growing in a forest would usually escape the notice of most people and would not elicit any discussion of potential invasiveness. On the other end of the spectrum, an extensive carpet of a non-native species spread across an entire hillside with multiple, detrimental effects on surrounding plants and animals can easily be perceived as being invasive (especially if there are multiple reports of the same behavior in other locations). Where then, along this continuum, is a species first recognized by some authority as being "invasive"? This is where invasive species assessment protocols become important.

Many USA states have now adopted their own protocols and procedures for how to identify a plant species (or cultivar — more on this below) as invasive. The creation of a single, state-wide list of invasive plants is critical to prevent confusion among the general public in terms of which particular plants should be excluded from sale or at least closely regulated, and which plants should be promoted for gardening and other uses. Even more importantly, a single list is instrumental for green building councils who determine which plants are necessary for projects to get LEED certification, as well as for determining which plantings are permitted in developments that have adopted their own restrictions. Having a single recognized list also arguably levels the playing field for the nursery industry so that competition is fair and just among all of its members. It is important to remember, however, that the creation of a single statewide list does not preclude the creation of other regional lists by local parks and arboreta, but it does at least create some level of basic consistency across the state.

Invasive plant assessment on the state level is an ever-evolving process. Historically, invasive plant assessments in many states were quite casual, and often involved surveying land managers for the names of their most problematic species targeted for removal on their properties. The names of these plants were then combined together to form the invasive list for that particular state. However, over the last few years, many states have moved towards adopting more scientifically rigorous protocols. For example, the first list of invasive plants in Ohio was created in 2000 by surveying land managers across the state. Unfortunately, the nursery industry was inadvertently excluded from this conversation even though some of the listed species were of nursery importance. So when the Ohio Invasive Plants Council (OIPC) realized that the invasive plant list needed to be updated to recognize new invaders (such as lesser celandine, *Ficaria verna* [syn. *Ranunculus ficaria*]), the organization created an entire new assessment process that would be objective, transparent, and based on scientific data. The nursery industry was specifically invited to be a part of this process (in both the creation of the protocol as well as its implementation), as were representatives from research, land managers from local parks, state lands, and federal lands, non-profit organizations, and the general public. The final assessment protocol and its policy of implementation ultimately were approved by the leadership of the OIPC and the Ohio Nursery and Landscape Association (ONLA). Today, periodic assessments in Ohio are conducted by a five-person team, which includes two representatives suggested by the ONLA and approved by the OIPC. Other states as well have been purposely reaching out to engage nursery professionals, plant breeders, propagators, and horticulturalists in their invasive

plant assessment processes.

Many states have or are forming their own assessment protocols, and there are several generalizations that can be made. First, many protocols can be classified into two types, depending on the purpose of the resulting list for a particular state (Buerger et al., 2016). On one hand, species are identified as invasive purely for educational or informative reasons (for example, currently in Indiana, Michigan, and Ohio), while on the other hand, a plant species may be listed as invasive for purposes of regulation (Illinois, Minnesota, Wisconsin, Connecticut). Educational protocols usually rank assessed species as Invasive, Not Very Invasive (or similar wording), or Need More Information, and these protocols often involve a point system. In contrast, regulatory protocols classify assessed species as Needing Some Regulation (Prohibited, Restricted, etc.), No Regulation, or Need More Information and these are often based on committee discussions using a non-point system or decision tree (or in some states, a point system is only used to guide the initial committee discussions). Second, the size and composition of the committee typically conducting these plant assessments varies by state (ranging from 5 members in Ohio to many more members in Wisconsin, depending on the species that is discussed). In contrast to the past, most states now increasingly recognize the horticultural industry and plant propagators as critical members of the assessment process and include them in discussions. For example, the Midwest Invasive Plant Network (MIPN) has been engaging various members of the green industry over the past 3 years with their Invasives in the Trade Working Group.

Assessments for various states typically consist of a mix of questions, some of which aim to predict whether a plant will invade. This is especially true for protocols developed for regulation purposes, as their intent is often to prevent future plant invasions (in contrast to just identifying plants that are already established invaders). The questions in the protocols for different states can generally be grouped together into at least five major categories (Buerger et al., 2016):

1) Current Distribution. These questions are designed to assess how widespread is the plant within natural areas locally, regionally, and sometimes even nationally. Plants growing in dense numbers within natural areas across regions where they were not planted will achieve a score or generate the most points that indicate the strong possibility that the plant may be invasive. In contrast, plants that are limited in number or not yet present within a given state may generate a low score for this particular set of questions. Some states also include questions about the distribution of the plant in surrounding areas or nearby states (if the plant is not yet present outside of cultivation in the state in question). This is critical because research has indicated that the best predictor of invasiveness in plants is whether the plant is invasive in a nearby location or similar habitat (Reichard and Hamilton, 1997; Kolar and Lodge, 2001; NAS, 2002).
2) Establishment and Expansion Capability. Plants that are most likely to be identified as invasive typically are those that are experiencing rapid expansion across multiple environments (or have the potential to do so). In some cases, these plants may have already established in a location and are just now showing early signs of spread or are otherwise already beginning to expand geographically. Questions within this category often refer to the biological characteristics of plants, such as seed production, vegetative spread, and seed dispersal ability (Sakai et al., 2001).
3) Ecological Impacts. This series of questions are motivated by the fact that some invasive plants have larger negative impacts on natural ecosystems than other plant species. The highest scores for these questions are often given to plants that outcompete native plants, reduce survival and reproduction of animal species, and negatively impact ecosystem processes such as nutrient cycling, fire regimes, and forest succession. This type of information is documented most often in the scientific literature for invaders widely distributed across their introduced range.
4) Socio-Economic and Cultural Impacts. A subset of states (such as Michigan and Florida) also consider the economic contribution of the assessed species as part of their normal assessment process. Most often this refers to the role of the species in

horticulture, or its potential or current use as feed for cattle, biofuels, or other means of generating financial income within the state. Essentially, the "invasiveness" of a species is downgraded slightly if its removal from industry would cause undue financial hardship on state residents or industries. Consequently, this category of questions is most prevalent in states whose goal is to regulate invasive species. In other states (such as Ohio), the economic importance of a species is not included in the assessment as it is considered separate from explaining why a species may be biologically invasive.

5) Prevention, Control, and Management. Several states acknowledge the importance of control and management costs of assessed plants. In this case, plants that are most difficult to remove from natural landscapes generate the highest assessment scores. As with the socio-economic questions, this category is not used in some state protocols because the cost of invasive removal is not considered by itself to be a biological reason why a plant may become invasive on its own. In many cases, however, land managers find this category of questions to be extremely helpful in prioritizing their management plans.

Regardless of the categories of questions above, most state protocols require evidence to support each answer. Ideally, this would consist of a scientific study published in the peer-reviewed literature. In some cases, this involves documentation of the occurrence of a plant in natural areas, using mapping sources such as BONAP (http://www.bonap.org), USDA PLANTS database (http://plants.usda.gov/java/; note that the "I" species notation here indicates "Introduced", and not "Invasive"), or EDDMapS (https://www.eddmaps.org). Ultimately, effective protocols must yield answers and final assessments that are easily understandable, transparent, and clearly based on scientific evidence in order to be convincing to a broad range of constituents.

THE CULTIVAR QUESTION

As more states develop assessment protocols, there is increasing focus on the role of cultivars in species invasions (see for example, Knight et al., 2011) and how they should be dealt with in the assessment process. Although there is not yet general consensus, many states currently group cultivars with the parental species. In this case, if a given plant species is assessed as invasive, all known cultivars are also listed as invasive, unless shown otherwise. In other words, cultivars are "presumed guilty unless proven innocent." In other states, cultivars are assessed separately from the original species, either using the same protocol (as in Ohio) or a separate protocol developed specifically for cultivars (Florida, Indiana, and New York). In Ohio, this process is particularly difficult because of the frequent lack of biological information regarding specific cultivars in the scientific literature.

An important challenge to the assessment of cultivars often involves the identification of escaped individuals. Are escapees the cultivar itself (usually rare), offspring of cultivars planted nearby, offspring from seeds dispersed from naturalized populations that were initiated by seeds of cultivars, or are they members of the parental species? Although escapees are typically identified through morphological traits such as growth form or leaf color and shape, this can be deceptive in some cases and genetic methods remain the best way to conclusively verify the identity of escaped individuals. For example, individuals of Japanese barberry (*Berberis thunbergii*) are sometimes found in natural areas, but their ornamental origin has been questioned because wild plants produce green leaves, lacking the red/purple coloration of most popular cultivars. However, it has since been shown using greenhouse crosses that a proportion of offspring of purple cultivars can indeed produce green leaves (Lehrer et al., 2006a; Lehrer and Brand, 2010). Even more importantly, an individual plant can shift from producing purple to green leaves during a single growing season, depending on the amount of light available (Lehrer and Brand, 2010). Furthermore, genetic tests of wild individuals have confirmed their cultivar parentage (Lubell et al., 2009), and Japanese barberry cultivars are known to produce seed (Lehrer et al., 2006b) that germinate and grow in natural conditions (Lubell and Brand, 2011) with their offspring capable of also producing seed in woodlands (Brand et al., 2012). Consequently, the identity

of escaped individuals in natural areas must be examined carefully because wild individuals may not morphologically resemble their cultivar parent. A straightforward way to overcome the difficulty of determining which cultivars have or will contribute to invasive populations is to determine if a cultivar is capable of producing viable seeds or other propagules that can disperse away from the maternal plant.

An additional concern for cultivar assessment is the potential for different cultivars of certain plant species to cross-fertilize one another, creating hybrids and potentially triggering invasive populations. This has been seen, for example, in Callery pear (*Pyrus calleryana*) trees in which any given cultivar is self-incompatible (such as 'Bradford', 'Chanticleer', or 'Autumn Blaze') but the combination of cultivars (or a cultivar and its rootstock) together results in cross-fertilization and seed production (Culley and Hardiman, 2007; Culley et al., 2011). Thus an individual cultivar is technically not invasive, but the species is invasive because of the different cultivars that are produced and distributed together across the county (Culley and Hardiman, 2009). Similarly, popular cultivars of *Lythrum virgatum* such as 'Morden Pink' and 'Morden's Gleam" (often sold as alternatives to the highly invasive *L. salicaria*) are now known to produce seeds following cross-pollination with each other or with introduced *L. salicaria* growing nearby (Lindgren and Clay, 1993; Amon et al., 2007). This highlights the fact that cultivars cannot be examined in isolation of one another but they must be grown together in an array of genotypes to best determine which may have any potential to spread.

More recently, researchers and plant breeders have begun to focus on the development of low fecundity ("sterile") cultivars that may serve as practical alternatives to highly invasive, but ornamentally popular plant species (e.g., Callery pear, Japanese barberry, etc.). This is most important in states with invasive plant regulation but it also provides a way in which ornamental plant breeders can be perceived as being environmentally friendly. However, the concept of sterility is still debated by researchers – such as whether sterility is permanent or transient, and whether seed sterility is sufficient or whether pollen sterility is also important. In addition, vegetative growth is rarely addressed in cultivars and could be important, especially for plants that disperse by vegetative fragments growing near waterways where water dispersal is common. Scientific studies have shown that even cultivars with very low seed production can still potentially trigger an invasion (Knight et al., 2011). However, most researchers agree that a permanent, completely sterile plant may not be realistic in the long term. For example, some states, such as Oregon where butterfly bush (*Buddleja davidii*) is regulated, define sterility as less than 2% seed production in order for cultivars be approved for statewide sale. In other words, this level of seed production is viewed as an acceptable level of risk in the state. Many states in the Midwestern USA are now working together to best define the concept of sterility for cultivars and what would be an acceptable standard.

CONCLUSIONS

In order to remain profitable, plant propagators need to remain cognizant of invasive plant assessment in their state, especially for ornamental plant species or cultivars that are just beginning to spread but have already been determined to be invasive in other states. There are many opportunities for plant propagators to become actively involved in the discussion of and specifically, the assessment of invasive plants. A good starting point is to contact the invasive plant council (sometimes known as the exotic pest council) in their state, if such a council exists and is active. If a plant propagator lives in the midwestern United States, a good resource is also the Midwest Invasive Plant Network MIPN; see http://www.mipn.org). Many of these organizations would like to engage plant propagators and breeders in their discussions, in recognition of that fact that woody invaders in particular often have a past or current horticultural use. There is also increasing recognition that we all have a common interest in protecting our natural resources and working together to create practical ways to reduce the harmful impacts of invasive species in our communities. Horticulturists need to also engage in these discussions so that they can be part of solutions that allow them to remain commercially viable while effectively reducing

current and future species invasions. One proactive approach is for plant breeders and propagators to begin to develop sterile cultivars so that they can be well positioned to offer alternatives if the associated species is identified as invasive in the future. Other solutions may also be found if plant propagators actively engage in discussions with land managers, academic researchers, and other interested parties who recognize that they all have a common interest – to reduce species invasions in our natural communities.

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Sterile cultivars (or close to it) — is this a viable option for the nursery industry?©

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INTRODUCTION

Some popular landscape plants have proven over time to exhibit invasive tendencies. The realization that these plants are invasive has led to legal bans of known invasive ornamental species in some states. For example, *Berberis thunbergii* and *Euonymus alatus* have been illegal to grow, sell, and transport since 2004 in New Hampshire and since 2009 in Massachusetts. In 2013, New York began a legal phasing out of *Berberis* and *Euonymus*, and Minnesota and Wisconsin have initiated partial bans on the most fecund Japanese barberry cultivars.

Many of the characteristics that make plants invasive, also make them good landscape plants. Invasive plants are typically tough adaptable plants that perform at a high level in managed landscapes. In addition, they often are highly ornamental and some are unpalatable to deer, making them even more useful in regions where deer populations have exploded. Use of native species or non-invasive exotic species as alternatives to invasive species has had some success. However, there are some invasive species for which it is hard to find replacement plants that provide the same set of ornamental characteristics and landscape performance traits that are delivered by the invasive plant. For these hard to replace invasive species, there is considerable interest in the development of sterile forms of these plants. Gagliardi and Brand (2007) found that the green industry strongly supported the development of sterile forms of ornamental plants as a solution to the invasive issue.

DEVELOPING STERILE FORMS OF INVASIVE PLANTS

Species undergoing breeding work

Several university and arboretum plant breeders are focusing considerable effort on development of sterile forms of important landscape plants that are invasive. The list of taxa that breeders are working on includes *Acer platanoides, B. thunbergii, Buddleja davidii, Campsis, Cotoneaster, E. alatus, Hibiscus syriacus, Hypericum, Ligustrum, Malus, Miscanthus, Prunus,* and *Spiraea*. We are just beginning to see some of the bred sterile plants enter the market. An example of sterile plants that have been big sellers recently are some of the newer *Buddleja* hybrids that are either completely sterile, or produce much reduced numbers of seed.

Breeding and evaluating plants for sterility is a long-term process, which can be technically challenging. In addition to the challenges inherent in developing sterile plants, there are many other impediments to the use and acceptance of sterile landscape plants. Some states already have legislative bans of invasive species in place. These bans include all forms of a species, including horticultural cultivars. In states with existing plant bans, new legislation will be required that will allow for exemptions for sterile cultivars before they can be used. Reversing existing legislation is often even more difficult to make happen than establishment of the original legislation. Undoubtedly there is also some loss of market for particular species where plant bans have been in effect. Customers who have gotten the message about the invasiveness of a particular species will need to be re-educated about new sterile forms in order to overcome concerns they now have about invasiveness.

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Public trust issues

In a broader sense, there are probably some significant trust issues the public will have with sterile cultivars of invasive ornamentals. To a great extent this lack of trust stems from the public's poor understanding of plant genetics, plant growth, and plant reproductive biology. Further exacerbating this situation are "pseudo scientists" who use the internet and other venues to spread their conviction about sterile plants, which is often not founded in scientific fact or evidence.

Misinformation about two ornamental plants, *Lythrum virgatum* 'Morden Pink' and *Pyrus calleryana* 'Bradford', has placed a great mistrust of plant breeders and the nursery industry in the public's psyche. Almost without exception, when presenting the topic of sterile invasive plants to the gardening public, or general public, I am questioned about whether I can "guarantee that the plants I breed won't revert or change to fertile plants like Morden Pink loosestrife or Bradford pear did." Information about *L. virgatum* 'Morden Pink' from the Agriculture Canada Morden Research Center was misinterpreted initially and some catalogs listed the genotype as sterile, even though it was known to be female fertile by the scientists working with it. 'Morden Pink' was used in crosses to create 'Morden Gleam' and 'Morden Rose' clearly demonstrating a lack of sterility. Incorrect plant catalog information became "fact" over time and soon everyone believed 'Morden Pink' was sterile. To make the situation worse, isolated garden plants of 'Morden Pink' appeared sterile to growers and gardeners because of the complex tristylous reproductive mechanism used to force outcrossing in this genus (Anderson and Ascher, 1993). In tristylous plants, the combination of flowers styles and stamens on each genotype will be short, medium, and long in length. Only stamens and styles of the same length produce seed set. Therefore, 'Morden Pink' has to outcross with another genotype in order to match its style length with another genotype or species with appropriate stamen length. When other *Lythrum* genotypes or species are present, 'Morden Pink' produces lots of seed.

Pyrus calleryana 'Bradford' is another ornamental plant that has added to the public's negative perception about sterile plants. *P. calleryana* was brought to the USA as a fire blight resistant rootstock and for potential use in breeding to incorporate fire blight resistance into *P. communis* (Whitehouse et al., 1963a). Many sources incorrectly state that 'Bradford' was bred to be sterile. However, it was a seedling selected from seed obtained from China (Whitehouse et al., 1963b). Initially, 'Bradford' was observed to produce little fruit, but this lack of fruit production was due to self-incompatibility, not sterility (Zielinski, 1965). Exposure to new cultivars or genotypes of *P. calleryana* resulted in significant fruit and seed production by 'Bradford' (Culley and Hardiman, 2007). This scenario has resulted in the public believing that "sterile" Bradford reverted to a fertile condition and that sterile plants will all eventually become fertile again. The bottom line is the public no longer trusts plant breeders or the nursery industry when it comes to the topic of plant sterility. This is a significant impediment to the use and acceptance of sterile invasive plants.

Methods to produce sterile or near sterile plants

On the positive side, methods do exist that can be used to create sterile or near-sterile forms of plants. These plants will not spontaneously "revert" to a fertile condition. Significant advances have been made in transgenic technologies that can be used to create sterile plants. However, the use of transgenic methods to produce sterile forms of ornamental invasives is not currently a viable option. The negative public opinion about transgenic plants and the regulatory hurdles that must be cleared are currently too large for pursuit of this strategy to develop sterile invasive landscape plants.

Ploidy manipulation is the most often used method for the creation of invasive plants that produce no seeds or few seeds. In most cases, the goal is to develop triploid plants, which will typically have low fertility due to unpaired chromosomes during meiosis. One way to make triploid plants is to take advantage of triploid endosperm tissue that is produced as a result of double fertilization in the ovule. Three 1N nuclei are produced by a pollen grain that lands on the stigma. One becomes the tube nucleus, which forms the pollen tube through the stylar tissue. The other two nuclei are generative nuclei, which enter the

ovule. One generative nucleus fertilizes the 1N egg to form the 2N embryo and the second generative nucleus combines with a pair of 1N polar nuclei to create the 3N endosperm, which develops into an important food source for the growing embryo.

In non-endospermic seeds the endosperm food reserves are transferred relatively quickly to the cotyledons of the developing embryo making it challenging to take advantage of this tissue. In endospermic seeds, the cotyledons are small and the endosperm remains large even in a fully developed seed. For endospermic seeds, the 3N endosperm tissue is accessible and can be used as a source of natural triploid cells. Triploid endosperm cells can be induced to form callus in vitro and eventually to form shoots. Shoots can then be rooted to form triploid plantlets. Endosperm derived triploid plants will have two sets of maternal chromosomes and one set of paternal chromosomes. Triploid *Euonymus alatus* has been produced using this procedure (Thammina et al., 2011).

Triploid endosperm is only useful for a limited number of plants due to inaccessible endosperm or recalcitrance in vitro. So a more common approach to creating triploids is to first create tetraploids from diploid plants. Mitotic inhibitors such as colchicine or oryzalin are used to double the chromosome number in plant cells. Meristematic tissues, such as the plumules of germinating seeds or shoot apical and lateral buds are the targets of mitotic inhibitor treatments. Plants are produced from the tetraploid shoots and grown to flowering size. Crosses are then made between tetraploid and diploid plants to create triploids.

Triploid plants must be thoroughly evaluated to determine their level of fertility. Some will be fully sterile, but others will express low and variable levels of fertility. In genera or families where apomixis is known, triploid plants can utilize asexual embryo formation to produce large numbers of viable seeds. While development of triploids can be relatively straight forward in some species, other species possess a triploid block where it is very difficult to obtain triploid seeds, most often due to failure in endosperm development that ultimately results in embryo failure (Köhler et al., 2010).

Berberis thunbergii has a strong triploid block. Despite extensive efforts to generate triploid barberry in my breeding program by crossing 4N and 2N plants, I have only been able to generate four individuals. All four plants have not produced any seeds despite flowering and producing fruit. Unfortunately, all of these triploids have green foliage and are not as desirable as ornamentals as they would have been with purple foliage. Although triploid barberry has been a difficult achievement, it has been relatively easy to produce large numbers of autotetraploids. While many autotetraploid barberry have been fertile, others have been highly seed infertile and we have dwarf or compact tetraploid genotypes with purple, yellow, or green foliage that will soon be available in the trade.

Another method that has been used to create infertile landscape plants has been wide interspecific or intergeneric hybridization. In *Buddleja*, interspecific hybridization, especially when three or more species are involved in the cross, has produced very reduced seed production or even complete sterility (Werner and Snelling, 2011). Similarly, in *Berberis*, a tri-specific cross involving *B. verruculosa, B. gagnepainii*, and *B. vulgaris* has exhibited low seed set (Brand, unpublished). Two of the species involved have blue/black fruit and one has red fruit, so having genomes from different ends of the *Berberis* spectrum helps reduce fertility.

Regardless of how putative sterile plants have been produced, it is of the utmost importance that they are thoroughly studied and documented to perform as claimed. Without thorough confirmation of the level of sterility, there is the risk of a plant becoming another *L.* 'Morden Pink' or *P. calleryana* and further eroding the public's confidence in sterile plants. First, ploidy should be confirmed using flow cytometry and chromosome counts. If a plant is an intergeneric hybrid, its hybridity should be confirmed through both morphological and genetic analysis.

To accurately document seed production, putative sterile plants must be planted with appropriate fertile controls in a replicated planting. One must provide for genetic outcrossing and outcrossing with various ploidy levels by including multiple genotypes. Plants must be allowed to mature enough to insure that reproductive capacity isn't overlooked simply because plants are too young. Brand et al. (2012) found that barberry

cultivars on average increase fruit production over 1000% when comparing 5-year old to 10-year old plants. Several plants that appeared sterile at 5 years of age were producing seed at 10 years of age.

Reduced fertility an acceptable option?

When completely sterile plants cannot be achieved are plants with reduced fertility an acceptable option? How reduced does seed production need to be in order to be acceptable? Knight et al. (2011) make the case that long-lived woody plants will need to have extremely low levels of seed production in order to insure no population growth. Brand et al. (2012) developed predictive information about Japanese barberry seedling establishment in the wild using a combination of seed production, seed germination, and seedling survival data. Barberry genotypes producing about more than 50 seeds per year would likely result in one or more seedlings becoming established in an unmanaged woodland. To be most useful, similar data will need to be established for each invasive species that is being considered for sterile cultivar development if absolute sterility cannot achieved.

The nursery and landscape industry has been supportive of cultivar exemptions for sterile or near sterile genotypes of important invasive landscape plants. The best example of a working cultivar exemption for sterility is one established in Oregon for *Buddleja* (Oregon Department of Agriculture, 2011). Regulation of *Buddleja* is through the Oregon Department of Agriculture (ODA). To be approved for sale, a *Buddleja* genotype must produce 2% or less viable seed or be documented to be an interpecific hybrid. In 2015 there were 18 approved cultivars of *Buddleja* that were legal to use in Oregon. To gain approval for exemption, one can either submit independent research documenting the level of fecundity to the ODA for review, or they can pay to have Oregon State University evaluate the fecundity of a plant. For interspecific hybrids, proof of parentage information must be submitted to ODA for review. The *Buddleja* cultivar exemption program in Oregon seems to be successful so far and can serve as a model for other states to follow with additional plant species (Contreras and McAninch, 2013).

In New York, where Japanese barberry has recently been legally banned, a decision-making tree was developed to support a cultivar exemption program. Barberry cultivars do not necessarily have to be completely sterile to be approved for sale, but must produce low numbers of seed and meet several other criteria that collectively would result in low risk of establishment in unmanaged areas. Minnesota and Wisconsin have taken a slightly different approach with their recent bans of *B. thunbergii* and cultivar acceptability. They used data developed at the University of Connecticut (Brand et al., 2012), which documented seed production levels for 45 cultivars. Minnesota and Wisconsin legislation bans the species (*B. thunbergii*) plus 25 cultivars, which produce high numbers of fruit. Lower fruiting cultivars are still legal, but the language included in the legislation states that when horticulturally acceptable seedless cultivars become available revisions should be made to reduce the seediness considered acceptable for use.

Massachusetts, which has a long-standing ban on all *B. thunbergii* and cultivars, has formed a committee to explore the possibility of sterile cultivar exemptions. New Hampshire is not considering cultivar exemptions to its barberry ban at this time. A concern that is often voiced by those considering support for cultivar exemptions for sterile plants is how can one be sure of the identity of a plant. Often, sterile cultivars may be hard to distinguish from fertile forms of the same plant. Mechanisms need to exist to help prevent the sale of fertile plants either intentionally or accidentally. As genetic testing of plants becomes increasingly routine and affordable, it will become reasonable to require random genetic checks to confirm the identity of sterile plants on the market. In addition, sterile plants will all be patented and licensed to specific growers, making tracking of plant material fairly straightforward. Sterile plants will probably all be sold with individual plant tags that get carried forward with the plant from propagation to final sale, again making plant tracking easier.

Given the number of plant breeders currently focusing effort on the development of sterile forms of invasive landscape plants, there will undoubtedly be numerous new sterile

plants arriving in the market in the next decades. It is likely that exemptions in plant bans to allow for the use of sterile cultivars will become widespread and commonplace.

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Protocols for testing the invasiveness of plants in Florida©

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INTRODUCTION

Globalization has facilitated the movement of non-native species worldwide through increasing connectedness between isolated ecosystems (Meyerson and Mooney, 2007). Only a small proportion of non-native species introduced to a new range become established, and those species that do become invasive have significant economic and ecological impacts, often resulting in reduced biodiversity and changes in biogeochemical cycling, hydrology, and disturbance regimes (Gordon, 1998; Mack and D'Antonio, 1998; Vitousek et al., 1996). Invasive species can be defined as an organism (plant, animal, fungus, or bacterium) that is not native and has negative effects on our economy, our environment or our health.

Florida and California lead the continental United States in the number of invasive species (Vitousek et al., 1996). In particular, Florida is notorious for its conspicuous invasions by plants and animals including the Burmese python (*Python bivittatus*), lionfish (*Pterois volitans*), giant African land snail (*Lissachatina fulica*), and old-world climbing fern (*Lygodium microphyllum*). The combination of the peninsular shape and a northern frost boundary creates a subtropical island with biogeographical implications including reduced native fauna and flora, and increased susceptibility to biological invasions (Ewel, 1986; Gordon, 1998). Additionally, approximately 85% of all non-native plants enter the US through Florida (Simberloff, 1996). It is estimated that over 25,000 species have been introduced to the state with over 1400 establishing, many of those in sensitive natural areas (Gordon, 1998; Adams et al., 2011). To date, over 15% of natural areas have been invaded by one or more non-native plant species (Jubinsky et al., 2007). Once these species take hold, there are significant impacts to recreation and species are expensive to manage with management costs in the tens of millions of dollars (Langeland, 2013).

There are many common biological traits associated with invasive species including high relative growth rates, longer flowering and fruiting periods, high fecundity, efficient propagule dispersal, short minimum generation times, tolerance to a wide range of habitats, and efficient resource utilization (Gordon, 1998). Unfortunately, many of these biological traits are also common in most horticultural and landscaping plants. In fact, 60% of all the invasive, non-native species are linked to the ornamental plant trade, forestry, or agriculture (Grotkopp et al., 2010) and 82% of the invasive woody plants in the USA were introduced through horticulture or landscaping (Reichard and Hamilton, 1997). But not all non-native plants intentionally introduced become invasive and many are economically beneficial with total sales of the nursery and landscaping industry in Florida topping $15.3 billion in 2010 (Florida Nursery Growers and Landscape Association, http://www.fngla.org). Effective screening tools can utilize information regarding the traits associated with invasive species to assess the invasive potential of non-native species to prevent future invasions and not hinder economic growth.

WHAT IS THE ASSESSMENT?

A subcommittee of the UF/IFAS Invasive Plant Working Group created the UF/IFAS Assessment in 1999 to provide status and risk assessments for nonnative species in Florida's natural areas. These recommendations reduce invasion into natural areas by ensuring that plant species with invasive characteristics are not recommended for use by UF/IFAS faculty. The UF/IFAS Assessment has three assessment protocols: the Status Assessment for non-native species already present in the state, the Predictive Tool for species proposed for

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release (or a new use), and the Infraspecific Taxon Protocol to assess cultivars, subspecies, or hybrids of known invasive species.

Status assessment

The Status Assessment provides a well-defined system to determine if a nonnative plant species is (or is at risk to be) invasive in Florida's natural areas. Recommendations reached through the Status Assessment are intended to prevent invasions and reduce the spread of current invasions. The Status Assessment is intended *only* for plants that currently occur in Florida and is not intended to provide evaluations of species that have not yet been introduced to the state. To account for differences in how a species will perform in different regions of the state, Florida has been divided into three zones — North, Central, and South. These zones are based roughly on the USDA hardiness zones (http://planthardiness.ars.usda.gov/PHZMWeb/), and conclusions are developed for each zone independently. For example, some species may be invasive in all parts of the state, while others are limited to particular zones (e.g., subtropical South Florida). Additionally, species are systematically re-evaluated to document changes in their status, and conclusions are amended when necessary.

The Status Assessment consists of questions about ecological, management, and economic aspects of the species and also the species' potential to expand into non-invaded zones. At least three experts (i.e., land managers or scientists) in each region familiar with the status of the species complete questionnaires for the status assessment. These experts provide the following information:

- Distribution of the species (i.e., how many acres are occupied and the habitat types invaded).
- Long-term alterations to ecosystem processes (i.e., changes in fire regimes, allelopathic interactions, and changes in community structure).
- Life history traits related to fecundity (i.e., number of viable propagules, time to reproductive maturity).
- Management practices (i.e., which management methods are used, difficulty in implementation, and cost).

Their responses are incorporated with information gathered from an extensive literature search (herbaria records, peer-reviewed primary literature, floras) to reach UF/IFAS Assessment final recommendations.

There are four possible results of the Status Assessment:

1) Not considered a problem species at this time, may be recommended.
2) Caution, may be recommended but manage to prevent escape.
3) Invasive and not recommended except for "specified and limited" use approved by the UF/IFAS Invasive Plant Working Group.
4) Invasive and not recommended.

The conclusions include plans for reassessment, after either 2 years for "caution" and 10 years for "not a problem" and "invasive." Additionally, any species may be reassessed whenever additional relevant information becomes available that might change the conclusions of the Status Assessment.

Predictive tool

The purpose of the Predictive Tool is to decrease invasions in Florida's natural areas by ensuring UF/IFAS faculty do not recommend the use of plant species not yet introduced or only limitedly introduced to Florida that have a high risk of becoming invasive. The Predictive Tool is a weed risk assessment (WRA) protocol consisting of 49 questions used to evaluate species either new to the state or proposed for a new use. Weed risk assessments have proven to be a cost-effective tool where adopted. Economic analysis conservatively estimated that implementation of WRA will save Australia $1.67 billion (USA) dollars over a period of 50 years (Keller et al., 2007). Gordon et al. (2008) tested the accuracy of the predictive tool and determined that 90% of major invaders and 70% of non-invaders were accurately categorized by the protocol across a range of geographies (including Florida). The

accuracy of the predictive tool minimizes the occurrence of false positives and effectively predicts low-risk plant species that may be economically beneficial and nonnative plant species that have a high risk of invasion.

Questions presented in the Predictive Tool are answered by conducting thorough literature searches, using sources such as herbaria records, agency reports, and peer-reviewed primary literature. The questions in the predictive tool address the following areas:

- History of the species (i.e., domestication/cultivation)
- Biogeography (i.e., native range vs. proposed release sites, invasive status in other regions)
- Life history traits (i.e., plant type, growth habit, modes of reproduction)
- Ecology (i.e., persistence attributes, allelopathy, dispersal mechanisms)

Each question receives a numerical score between -3 and 5 points (most -1, 0, or 1), and conclusions are made based on the cumulative score. There are three potential outcomes of the predictive tool:

- Low risk of invasion (<1 point)
- High risk of invasion (>6 points)
- Evaluate further (between 1 and 6 points)

Thresholds for each conclusion were established at scores to prevent the introduction of many serious invasive species, to limit the rejection of species that have not become invasive to 10%, and to limit the number of species requiring further evaluation to 30% (Pheloung et al., 1999).

Like the Status Assessment, conclusions for the Predictive Tool are separately derived for North, Central, and South Florida. If the conclusion is "evaluate further," an additional tool called the Secondary Screen is used. The Secondary Screen is a decision tree consisting of a small subset of risk assessment questions that vary based on life form (Daehler et al., 2004). Trees and shrubs are evaluated on shade tolerance, stand density, dispersal, and generation time. Herbaceous plants (and small stature shrubs) are evaluated on their palatability to herbivores, their status as an agricultural weed, and their stand density (both decision trees are applied to vines) (Daehler et al., 2004). The addition of this supplemental tool has reduced the number of species requiring further evaluation by an average of 60% (Gordon et al., 2008). Additionally, the Status Assessment was revised to direct species to the Predictive Tool in the following two cases:

- Species that have not escaped into Florida's natural areas but are recent arrivals to the state or are known to cause problems in areas with climate and habitats similar to Florida
- Species that are being proposed for new uses (e.g., biofuel or biomass planting) that will result in significantly higher propagule pressure

The Predictive Tool has also been written into the ITP and is used in cases where obvious traits of the infraspecific taxon will alter its risk of invasion relative to the resident species.

Infraspecific taxon protocol

The Infraspecific Taxon Protocol (ITP) is an internal tool for UF faculty, particularly the UF/IFAS Assessment staff and the UF/IFAS Invasive Plant Working Group, to independently evaluate cultivars, varieties, hybrids, or subspecies of resident (nonnative species found in Florida) invasive species to determine if all taxa associated with particular species should receive the same recommendations.

UF/IFAS Assessment staff may initiate an ITP evaluation if new sub-specific taxa or hybrids are being recommended by UF/IFAS faculty or others. UF/IFAS faculty can also initiate an ITP evaluation when they want secondary testing of a taxon whose resident species has received a “do not recommend” conclusion (e.g., to obtain UF/IFAS approval to release a cultivar for commercial use). The petition for assessment must be accompanied by evidence demonstrating that the taxon is a distinct entity and has characteristics that will reduce its invasive potential compared to resident species. Examples of taxa that have been evaluated with the ITP include five cultivars of *Eucalyptus grandis*, three cultivars of *Ruellia*

and four *Lantana* taxa. The conclusion "not a problem species" was found for two of the *Ruellia* cultivars and all of the *Lantana* taxa. Even though the ITP is used infrequently, it does allow development of recommendations for taxa selected for uses (i.e., landscaping, biomass plantings) that may result in widespread dispersal and higher propagule pressure. The ITP consists of 12 questions to determine the following information:

- If botanists/field personnel will be able to distinguish the taxon from the resident species (or other infraspecific taxa) in the field
- If the taxon can regress (or hybridize) to characteristics of the resident species
- The fecundity of the taxon
- If the taxon displays invasive traits that cause greater ecological impacts than the resident species

Depending on the answers, conclusions may be drawn from the ITP, or the infraspecific taxon is directed to the Predictive Tool or the Status Assessment. Recommendations made directly from the ITP fall into the same possible categories outlined in the Status Assessment. Final recommendations and supporting data from the ITP must be evaluated by at least three experts (e.g., professional botanists, horticulturalists, plant breeders). If the ITP cannot be completed because of a lack of appropriate evidence, lack of three suitable experts, or if a consensus cannot be reached among the experts, then the conclusions for the resident species are applied to the infraspecific taxon.

Appeals must be addressed to the UF/IFAS Invasive Plant Working Group for case-by-case review. Recommendations for infraspecific taxa that have been assessed or evaluated using the ITP are listed in the online "Conclusions" table independently from the conclusions of the resident species. These follow the same reassessment schedule as the Status Assessment (http://plants.ifas.ufl.edu/assessment/conclusions.html).

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Increasing diversity and availability of native woody plants in the nursery industry©

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NATIVE PLANTS

There is a misconception that native plants, in general, are not that ornamental based on what's observed in the wild. There is some truth to this but through cultivation, many can become excellent landscape plants. In fact the nursery industry embraces many native plants that under standard nursery practices become excellent ornamentals. Good examples include many mainstream landscape plants. Trees such as *Acer saccharum, Amelanchier laevis, Asimina triloba, Cercis canadensis, Cornus florida, Fagus grandifolia, Nyssa sylvatica* as well as many species of *Quercus*. Shrubs including *Aronia*, *Clethra alnifolia*, *Cornus sericea*, *Diervilla lonicera* and *Ilex verticillata* are widely available.

Another problem is how we define native. It can be defined in different ways depending on how they will be used.

To the purest and those that are working with natural areas a native plant is one that occurs in a particular region, ecosystem, or habitat without human intervention. It is commonly accepted that the flora present at the time Europeans arrived as the species native to the eastern United States.

A more liberal approach may be taken when dealing with the public. It is often necessary to look beyond the local natives to create exciting landscapes and it is commonly accepted to look at natives east of the Mississippi or Midwest natives in our area.

A common fallacy is that natives will outperform introduced plants. This may not necessarily be true. The old adage of "right plant, right place" must still be followed and understanding of the conditions in which a plant grows is very important.

Nativars, love them or hate them, are a growing opportunity for nurserymen to meet the native people part way. There is a debate raging whether "nativars" have the same ecosystem services as the true native species. Many nativars are actually selections made from native species that have improved traits and not hybrid plants. Are they as beneficial as native species to wildlife? The answer is unclear without further research. The upside is that they awaken an interest and awareness in native plants to the general public.

Often when people refer to natives they are focused on herbaceous plants whether they are to be used in a prairie planting or in a pollinator garden. Woody plants are often overlooked. With a little research a nurseryman or landscape designer may find that they are actually using more natives and cultivars of natives than they realize.

Understanding the environment in which plants will be grown can open multiple opportunities to choose good native species and cultivars. Promoting these plants for their environmental value is an overlooked sales opportunity for many.

Another opportunity is to look at pollinator friendly plants. In reality pollinators probably visit as many nonnative plants and nativars as pure native species for their nectar. This is only part of the story. These pollinators, especially butterflies need plants on which to rear there young. *Lindera benzoin* is host to the spicebush swallowtail and *A. triloba* provides the food source for the zebra swallowtail. Other woody plants that can attract butterflies include trees such as *Populus* spp., *Ptelea trifoliata*, *Ulmus* spp., *Sassafras*, *Magnolia virginiana*, and *Salix* spp. Vines such as *Passiflora edulis* (syn. *P. incarnate*) and *Aristolochia* are also host plants.

WHEN GOOD PLANTS GO BAD

On the flip side we must also be aware that some of the plants that have been staples

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in the nursery industry have become bad performers. Plants are regional and invasiveness of a species is rarely national. It is imperative that as an industry we take a proactive approach and get involved in the invasive issue. Play a part in working with the natural areas people to understand the ramifications of invasiveness as it affects us all. Be prepared to bring facts not emotion to the table when discussing these plants. It is a fact that *Lonicera maackii* is invasive, especially in southern Ohio. Recently the invasiveness of ornamental pear has been observed in multiple areas. We may not like these facts, but we must face the reality that some plants will have to go out of or be limited in production. Sound science such as the rating system used in Ohio can help better understand which plants have the potential for invasiveness.

SUMMARY

Growing native plants is an opportunity to be environmentally friendly while developing a physically sound marketing program that can be a great sales opportunity.

Two sides of the same coin — finding common ground among plant conservation professionals and commercial propagators©

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INTRODUCTION

If ever asked, "Why should we appreciate plants and nature?" I can quickly and without hesitation reply, "All life depends on plants. Without the world's flora, life as we know it would not exist." But I have known many others, my friends and colleagues in commercial nurseries and other related professions, who know plants primarily as the source of their livelihood. In growing plants for sale, these people contribute significantly to the global economy and provide well for their families and others.

Are these in fact competing value systems or are they two sides of the same coin, where plant diversity — both wild and cultivated — contributes to our quality of life? I believe that in exploring this question, we can come to a greater understanding of why plants matter and better learn how we can work together for a better tomorrow.

For me, I think, I've believed in the value and importance of plants and nature for nearly all of my life, or at least it seems that way. Perhaps this understanding was a serendipitous result of being born in the 1970s, raised in a middle-class family in rural Ohio, steeped in educational television shows like *Wild Kingdom* and *Nature*, and influenced by a number of well-meaning teachers along the way, including my mother, an avid gardener. And if this were not enough, I grew up in a time when environmental concerns were increasingly in the public eye.

Two years after I was born, Peter Raven had this to say in the opening to his 1976 essay on plant conservation:

> "The roughly 300,000 species of green plants and algae provide the means by which the energy of the sun that reaches the earth's surface is locked up in chemical bonds. By carrying out this process, the plants and algae provide all of the food for from ten to thirty times as many heterotrophic organisms, including all the animals and man himself. … the diversity of plants is the underlying factor controlling the diversity of other organisms and thus the stability of the world ecosystem. On these grounds alone, the conservation of the plant world is ultimately a matter of survival for the human race."

In the 1970s, when Raven and other notable visionaries were espousing the virtues of conserving plants and nature, the world was in the midst of an environmental awakening of sorts. Spawned by a post-war realization that our planet was indeed a small place and getting smaller, globally-minded conservation organizations began to spring up including the International Union for the Conservation of Nature (1948), The Nature Conservancy (1951), and World Wildlife Fund (1961).

Many existing gardens, zoos, museums, and other centers of learning, including the Missouri Botanical Garden, The Royal Botanic Gardens Kew, the San Diego Zoo, and the National Museum of Natural History, among many others, were turning at least part of their attention and mission towards global environmental concerns. In the USA alone, the 1960s and 1970s witnessed a boom of new gardens and institutions including the National Tropical Botanical Garden, Chicago Botanic Garden, and the Marie Selby Botanical Gardens, all of which came onto the scene as conservation organizations.

As a result of this period of heightened ecological awareness, the children of the 1960s

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and onward have been steeped in conservation language, science, and culture. It would seem that if these trends continue, we would all soon become aware of the value of plants and agree to conserve them at all costs.

CHALLENGES TO THE CONSERVATION ETHIC

Despite indications that the world is ready to embrace conservation, tangible, large-scale results on the ground are often elusive. Remaining tracts of land continue to be cleared, species declines appear to be accelerating, and global climate change threatens the survival of even the most protected places and habitats on Earth. Putting it bluntly, Peter Karieva, former Chief Scientist and Vice President of The Nature Conservancy, along with his coauthors state, "By its own measure, conservation is failing." (Kareiva et al., 2012).

If conversion to a conservation ethic were simply a result of one's cultural environment and a concerted conservation campaign, it would be an easy matter for those in industry and the general public alike to adopt a conservation mindset. To the contrary, very real challenges in economics, short term gain vs. long term investments, and the simple but confounding issues that we as a species are creatures of habit, all compete with conservation in both philosophy and practice.

Exacerbating these very real challenges for conservation, fewer people today identify themselves as environmentalists than in the previous decade. Between 1989 and 2008, the percentage of the USA public that self-identified as environmentalists decreased from 76% to 41% (Marvier, 2013). As I and others have stated countless times, many are perhaps disenchanted with the onslaught of "doom and gloom." Still others might simply be unaware of the importance of plants and the environment despite intensive education campaigns by the Nature Conservancy, the Center for Plant Conservation, and many other conservation organizations large and small. In a world with so many challenges, as well as so many new and emerging opportunities, protection of nature seems distant and irrelevant to the daily lives of many.

THE OTHER PERSPECTIVES

Some of you might share my upbringing and worldview while many of you see things differently. Considering the wealth of experiences out there, it is no wonder. Take for example the following: a person who has only known the inner city her or his whole life. It must be really hard for someone to know and care for nature if they grew up away from the fields, woods, and wildlife that I took for granted as a child. Or still more challenging, I wonder what it would be like growing up effectively "in nature" but being so poor that I was preoccupied with where my next meal would come from.

On the opposite end of things, I often think about what it is like to be "any kid" in the USA today, a kid who has only known a connected world, on line, always looking at a screen, virtually free — but technically bound. In this case, I particularly worry about my son who by the age of 2 could already navigate an iPhone. I worry about how I will teach him to love the world outside when the world inside is so bright and captivating.

But there is cause for optimism because so many of us do appreciate plants, if only for different reasons. For me, I could have taken a path into the commercial plant industry, working in horticulture and plant propagation which I have done at various stages through my life. But instead, I jumped on the conservation bandwagon, driven by science, and found myself receptive to Raven's call to conservation long before I knew who he was. But I am fully aware this is only one of many ways to look at plants. The real trick, and the truly difficult part of what I believe needs to be done, is finding creative ways to appreciate and save nature regardless of our differences and perspectives. Preserving our quality of life and maintaining our livelihoods at the same time is essential. To do this, we have to go back to what it means to do conservation, and begin listening again to others to learn what it is we should be doing.

FOR THE FUTURE OF OUR PLANET, WE MUST BE OPEN TO NEW IDEAS

The oft repeated quote, "We have two ears and one mouth so that we can listen twice

as much as we speak" comes to mind. Attributed to Epictitus and referenced everywhere from church sermons to Forbes Magazine, this message speaks to the value of learning from others so that one can grow within. For conservations sake, I would take it a step further and say that we also have two eyes — we need to both listen and watch what others are doing so that we might create a focus — a mission — that stands the greatest chance of success.

We cannot do this alone and we certainly can't do it by forcing our conservation message upon others. We have to make conservation speak to them in whatever way works best. A number of prominent conservationists seem to agree with this notion including Peter Kareiva mentioned before, Emma Marris, environmentalist and author of *Rambunctious Garden*, and Greg Aplet, Senior Science Director at The Wilderness Society. These and many other scientists and environmentalist writers are advocating for a more introspective approach to conservation, one where we are open to new ideas and to experimentation.

In a recent editorial in the scientific journal *Nature*, Tallis and Lubchenco (2014) proposed "a unified and diverse conservation ethic; one that recognizes and accepts all values of nature, from intrinsic to instrumental, and welcomes all philosophies justifying nature protection and restoration, from ethical to economic, and from aesthetic to utilitarian." In this way, the authors argue, we will be able to fully embrace the role nature plays in society and, in turn, engender support and concern for the natural world among us all.

To accomplish this, we need not only listen to others, but experiment with new approaches and watch what happens … and see how others react. Marris and Aplet write in a 2014 New York Times editorial, "in the face of great uncertainty, we should hedge our bets and allocate large swaths of land to … restoration, innovation and hands-off observation." These new experiments go well beyond the borders of parks, preserves and remote wilderness. Proponents argue that we should be taking advantage of fallow farm lands, increasing numbers of vacant lots in cities, and abandoned industrial sites throughout the USA and the world. It is here that more people will see the results of conservation work and will provide opportunities for feedback and engagement including citizen science.

Through increased exposure and participation, we also stand a chance of engendering support; those who once did not care for plants and nature might begin to do so. And plant propagators have a role to play in this. It has been shown that when people are engaged in conservation that they increasingly become advocates for the mission and practice of conservation (Johnson, 2014). And what better way to engage people than to bring nature to where they live? New approaches might best include maintaining endangered species not just in preserves but also in cities and private collections at times. And in creating ways that a diversity of plants are available, not just those commonly used in landscaping and horticulture, we will create more opportunity for us both commercially and environmentally.

I recently spoke with Emma Marris and she joked with me that she would love to see golden lion tamarins (an endangered New World monkey) swinging through the cities of the Southeast USA in place of squirrels. While this might be a bit farfetched, the potential to introduce endangered trees and shrubs into city landscapes might be closer to reality. In doing so, we might further engage the public in plant conservation, a practice that was previously relegated to "the experts" for decades. And although this prospect has not been attempted on any meaningful scale to date, Peter Raven (1976) has suggested that lay enthusiasts might serve conservation by maintaining endangered plants in private collections, managed as part of distributed populations and in conjunction with botanic gardens and other enthusiasts.

PLANT DIVERSITY IS THE FUTURE — LET'S PRESERVE IT

I am comforted in knowing that I am not alone in my concern and love for plants; organizations like the Center for Plant Conservation are made up of some of the most devoted and passionate plant lovers there are. And when I see industry professionals actively engaged in discussion on how to be more sustainable and diverse in their businesses, like I heard so many discussing at the IPPS meeting in Cincinnati, I am again encouraged about the future.

In the end, just imagine how wonderful the world could be if the entirety of its people cared deeply about our only flora. The diversity of plants in nature has led to the infinite varieties and cultivars we know and love in our managed landscapes as well as in the food that graces our tables. To maintain and to continue to advance this diversity for everyone's good requires the ability to respect and embrace a diversity of ideas on how the world ought to be. As plant growers and plant lovers, whether garden enthusiasts, plant conservationists, or commercial plant propagators, it is our responsibility to manage and preserve this diversity. Let's embrace this notion and engender support for an environmentally and economically greener tomorrow. So long as we all care for plants and nature in some meaningful way, we all benefit in the end.

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Abscisic acid: a new management tool to improve quality and marketability of vegetable transplants©

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Abstract

Abscisic acid (ABA) is a plant hormone that triggers adaptive responses to water stress, including stomatal closure and shoot growth suppression. Our goal is to explore the potential of ABA in improving quality and marketability of vegetable transplants. First, we examined the stress control effect. In muskmelon (*Cucumis melo* L.) seedlings subjected to water withholding, pre-stress foliar spray of ABA improved the maintenance of leaf relative water content by limiting transpirational water loss. This effect was linear to ABA concentration (0.2 to 7.6 mM). Upon rewatering, the ABA-treated seedlings showed faster photosynthetic recovery and greater dry matter accumulation than the untreated seedlings. Second, we examined the height control effect for producing compact transplants. The effectiveness of height control by ABA varied among crops, cultivars, and growth stages: final transplant height was reduced by up to 20% in bell pepper (*Capsicum annuum* L.), whereas the benefit of height control was limited by overall growth delay in jalapeño and watermelon [*Citrullus lanatus* (Thunb.) Matsum & Nakai]. Overall growth suppression, however, may be of value as a growth holding strategy. When ABA was applied immediately before the maturity stage, all tested cultivars of bell pepper, jalapeño, and watermelon reduced excessive shoot growth (up to 29% 4 days after treatment) and prolonged the transplant marketability. One of the negative side effects observed across these experiments was leaf chlorosis, although it was concentration-dependent and mostly reversible within 7 days. Importantly, field evaluations demonstrated that the growth modulation by ABA was only transient with no negative impact on marketable yield. These results suggest that, with optimal concentration and application timing, ABA can be developed as a new management tool for vegetable transplants.

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Establishment and multiplication of firechalice in plant tissue culture©

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Abstract

Firechalice, *Epilobium canum* (Greene) P.H. Raven subsp. *garrettii* (A. Nelson) P.H. Raven, is a small and thinly branched plant that is difficult to germinate from seed. In order to increase the number of selected individuals rapidly, plant tissue culture would be the propagation method of choice. Single-node stem explants from a selected plant were examined for their ability to establish on Murashige and Skoog (MS) medium or Woody Plant Medium (WPM). Murashige and Skoog medium was found the best salt formulation particularly when supplemented with 4.4 µM benzyladenine (BA). During Stage 2, different plant growth regulators, such as BA, kinetin (Kin), 6-(γ,γ-dimethlyallylamino) purine (2iP), thidiazuron (TDZ) and meta-topolin (mT), were used in the media in different concentrations (1.1, 2.2, 4.4 or 8.8 µM). All the cytokinins tested induced the explants to form the most shoots and shoot dry weight when used at 4.4 or 8.8 µM in the medium. A concentration of 8.8 µM BA or mT were most effective for promoting shoot multiplication, with these concentrations inducing means of 13.7 or 14.1 shoots per explant, respectively. All but one cytokinin failed to affect shoot heights at the highest concentrations used, but 4.4 or 8.8 µM TDZ decreased shoot height by at least 54% compared to the control shoots. These results indicated that firechalice shoots established the best on MS medium for Stage 1 and 4.4 or 8.8 µM meta-topolin in the medium resulted in explants forming the most and largest shoots during Stage 2.

INTRODUCTION

Epilobium canum subsp. *garrettii* (also known as *Zauschneria garrettii*) common name firechalice or hummingbird flower is in *Onagraceae* family. This species is sometimes called "orange carpet" because the plant spreads as a ground cover, and its flowers are bright orange-red and attractive to hummingbirds. This species is relatively small, usually 30 to 46 cm tall and 30 to 61 cm wide (Love et al., 2009). Since plants grow easily in dry areas and have several good characteristics that are useful for urban landscapes, the plants should be propagated asexually to retain the desired characteristics. Axillary shoot proliferation is the best tissue culture technique for true-to-type reproduction. Plants used in axillary shoot culture will undergo the four stages of micropropagation. Stage 1 is establishment and stabilization of shoot cultures. During this stage the best basal medium to use must be determined. For example, Murashige and Skoog (MS) medium, Woody Plant Medium (WPM), or Driver-Kuniyuki walnut (DKW) medium can be used to establish shoots in vitro. Stage 2 involves inducing axillary shoot proliferation by increasing the level of cytokinin in the medium. Explants usually respond to high concentrations of cytokinin and produce many shoots (Einset, 1986). Benzyladenine is the most widely used cytokinin in the micropropagation industry, yet meta-topolin a relatively new synthetic cytokinins, can be used as an alternative to BA and zeatin.

RESEARCH OBJECTIVE

The goal of this research was to develop a micropropagation procedure for rapid production of a selected firechalice plant that was collected near Tony Grove Lake, Cache County, in northern Utah. We demonstrate that firechalice can multiply quickly in the first

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two stages of micropropagation so that hundreds or thousands of a selected clone can be made available to production nurseries.

MATERIAL AND METHODS

Stage 1

Firechalice shoots were established in tissue culture by testing two types of media. Single-node explants were placed on MS medium (Murashige and Skoog 1962) or WPM (Lloyd and McCown 1980). Murashige and Skoog medium contained 4.3 g L^{-1} mineral salts and 5.9 µM thiamine-HCl, 8.1 µM nicotinic acid, 4.9 µM pyridoxine-HCl, 53.3 µM glycine, 100 mg L^{-1} myo-inositol, 30 g L^{-1} sucrose, solidified with 7 g L^{-1} agar, was adjusted to pH 5.7, and included 4.4 µM BA. Woody Plant Medium contained 2.3 g L^{-1} salts, the same concentrations of thiamine, nicotinic acid, pyridoxine, glycine, and myo-inositol as MS medium, and contained 20 g L^{-1} sucrose, was solidified with 7 g L^{-1} agar, was adjusted to pH 5.2 and included 3.5 µM BA.

Stage 2

Shoot explants used in this part of the study were taken from shoot cultures grown on MS medium supplemented with 4.4 µM BA. Different cytokinins in different concentrations were used: benzylaminopurine (BA), Kinetin (Kin), 6-(γ,γ-dimethyl allylamino)-purine (2iP), Thidiazuron (TDZ), or meta-topolin (mT) at 0, 1.1, 2.2, 4.4, or 8.8 µM. Stem explants ~1 cm tall were placed on MS media containing different cytokinins and grown for 30 days before taking data. Statistical analyses for number of shoots, shoot height, and shoot dry weight were analyzed as by two-way analysis of variance (mixed model procedure) (Proc Mixed, SAS 2012) when comparing different plant growth regulators used at various concentrations. For Stage 2 analyses, cytokinin and cytokinin concentrations were used as independent variables. If the interaction between the cytokinins and their concentrations was significant for a growth parameter, then effects of the growth regulator concentrations were tested for each individual growth regulator. Significant differences between treatment means were determined by least-square means at the 5% level when comparing plant growth differences of explants placed on different media.

RESULTS

In Stage 1, shoot explants on MS medium produced at least 2 fold more new shoots, grew almost 3 times taller, and produced 4 fold more shoot dry weight than those on WPM (data not shown). After three subcultures of firechalice shoots on MS medium containing 4.4 µM BA, the shoots had stable growth (consistent foliage size and color), and the shoots were then used in Stage 2 experiments.

The effects of cytokinins in Stage 2 had to be analyzed separately due to an interaction between type of cytokinin and cytokinin concentrations. The two most effective cytokinins for promoting shoot multiplication were BA and mT. A concentration of 8.8 µM BA induced about 13.7 shoots to form per explant, whereas 4.4 µM mT induced 13.5 shoots to form per explant (Table 1 and Figure 1). The highest BA concentration (8.8 µM) increased shoot dry weight ~2.3 fold compared to the control stems. In contrast, 8.8 µM meta-topolin increased shoot dry weight about 2.6 fold compared to control shoots.

The other three cytokinins used in this study either had minimal or detrimental effects on the growth of firechalice shoots. For instance, even though 8.8 µM TDZ increased the number of axillary shoots formed by 2.9 fold and shoot dry weight by 4 fold over the control treatment, shoots height on medium supplemented with 8.8 µM TDZ were 2.5 times shorter than control shoots. Neither Kin nor 2iP concentrations affected shoot heights, yet 8.8 µM kin or 2iP increased shoot dry weights by 2.3 fold each compared to controls.

Table 1. Effects of plant growth regulators (cytokinins) on the mean number of shoots, mean shoot heights, and mean shoot dry weights of firechalice shoots grown on MS medium for 4 weeks. Data are means of six shoots in four vessels per treatment.

Plant growth regulator	Concentration (µM)	Number of shoots	Shoot height (cm)	Shoot dry weight (mg)
BA	0	3.1 a[1]	3	25 a
	1.1	7.4 b	3	32 ab
	2.2	10.6 bc	3.7	34 ab
	4.4	10.2 bc	2.9	36 b
	8.8	13.7 c	2.6	57 c
mT	0	2.3 a	3	27 a
	1.1	7.1 b	3.8	35 ab
	2.2	8.5 b	3.1	60 bc
	4.4	13.5 c	3.7	65 bc
	8.8	14.1 c	3.3	72 c
TDZ	0	2.1 a	3.5 d	24 a
	1.1	5.5 b	2.4 c	36 a
	2.2	5.2 b	2.1 bc	41 a
	4.4	5 b	1.6 ab	71 ab
	8.8	6.2 b	1.4 a	98 b
Kin	0	1.9 a	2.4	16 a
	1.1	3.1 b	3.0	25 ab
	2.2	3.9 b	2.9	32 b
	4.4	3.9 b	2.7	31 b
	8.8	7.9 c	3.3	36 b
2iP	0	2.5 a	3.0	23 a
	1.1	3.3 ab	2.7	19 a
	2.2	3.8 b	2.8	33 ab
	4.4	4.3 b	2.5	34 ab
	8.8	6.2 c	3.1	44 b

[1]Different letters within a column for each individual growth regulator indicate significant differences between means as determined by least-squares means tests at $P \leq 0.05$ level (n=24).

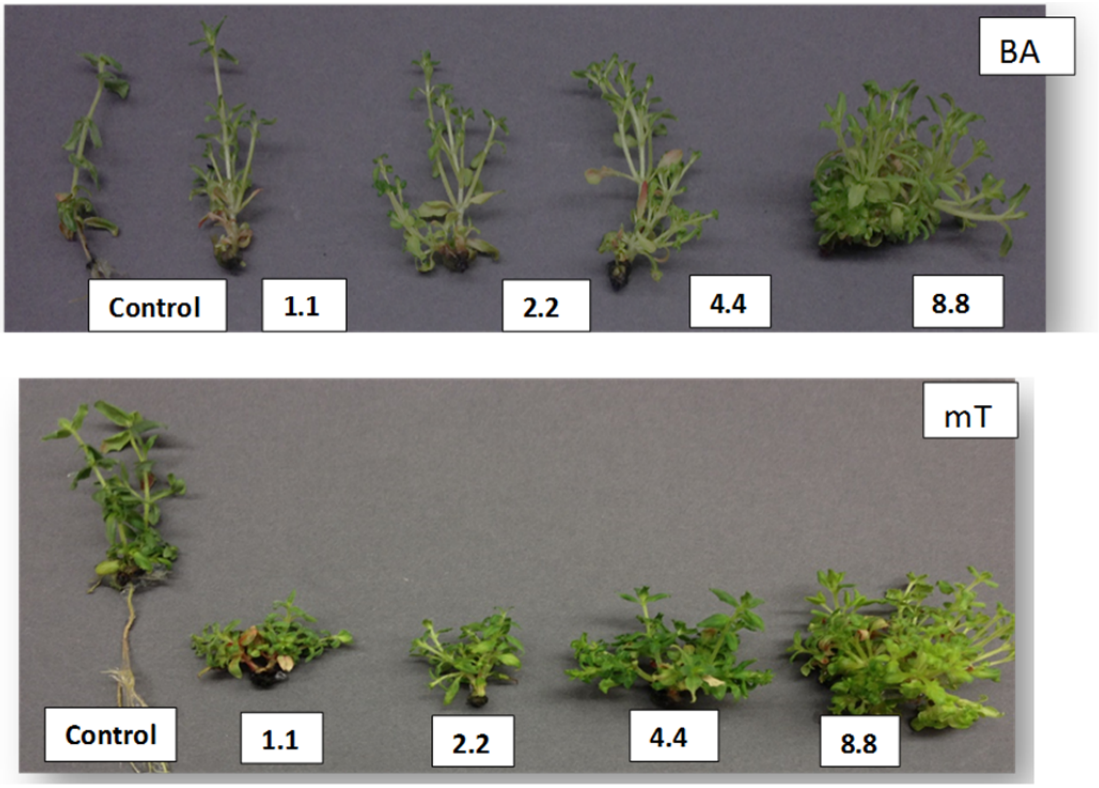

Figure 1. Effects of different concentrations of BA and mT on firechalice shoot multiplication after shoots were grown on MS medium for 4 weeks.

DISCUSSION

In Stage 2 studies with firechalice, mT promoted shoot multiplication the best, even a little better than BA. This information is important for propagators who have to decide which cytokinins to use in their media. Besides looking for the best plant responses in culture, propagators must also consider the costs of the biochemicals used. The cost of mT from *Phyto*Technology Laboratories in 2015 was $257 per gram, whereas the cost of BA from this same company was $5 per gram. The higher cost of mT failed to justify its use in commercial propagation since BA, which was 51 times cheaper, promoted shoot multiplication almost as well as mT. In contrast, addition of TDZ to shoot multiplication medium should be avoided since it inhibited shoot height growth of firechalice.

CONCLUSION

Exact duplicate plants could be rapidly increased for firechalice by using in vitro culture. MS medium was the best medium for establishing firechalice stem explants in Stage 1. During Stage 2, shoot explants were multiplied the best by using BA or mT at 4.4 or 8.8 µM.

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Micropropagation of a selected clone of *Amelanchier alnifolia*©

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Abstract

Shoots of serviceberry, *Amelanchier alnifolia,* propagated in tissue culture often fail to form roots readily. In vitro cultured shoots from a selected dwarf plant were examined for their ability to form roots when the basal salt concentration was adjusted or different plant growth regulators were used in the medium. Different concentrations of Murashige and Skoog (MS) salts were used (full, ½, ¼, and ⅛ strength). In addition, the plant growth regulators indole-3-butyric acid (IBA) or naphthaleneacetic acid (NAA) at concentrations of 0, 0.5, 1, 5, or 10 µM were tested for their ability to induce root formation. The effects of 2 µM benzyladenine (BA) on root formation were tested by combining BA with five NAA concentrations. The ⅛ strength MS treatment induced 38% of the shoots to form roots, whereas roots failed to form on shoots grown on full strength MS medium. The mean number of roots per responding shoot was 1.6. Indole-3-butyric acid and NAA concentrations induced root formation on full strength MS medium. The best rooting was achieved with 10 µM IBA or 10 µM NAA, and the percentage of shoots forming roots was 33% for IBA treated and 67% for NAA treated shoots. The mean number of roots per responding shoot were 6.1 and 2.5 for 10 µM IBA and 10 µM NAA treated shoots, respectively. Shoots treated with BA combined with NAA formed callus at their bases but failed to form roots. This study demonstrated that ⅛ basal salts or 10 µM IBA or NAA were effective for inducing root formation on serviceberry shoots produced in vitro.

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Seed germination studies of *Vitex agnus-castus*©

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INTRODUCTION

Vitex agnus-castus, also known as the chaste tree, is a plant that is grown for its ornamental qualities such as its delicate-textured, aromatic foliage and spikes of lavender flowers that bloom mid- to late-season and attract butterflies. It is also a plant that deer will not eat. *Vitex* is a shrub that grows 5 to15 ft tall with a spread of 15-20 ft and is winter-hardy to USDA Zone 7. The leaf of this deciduous plant is palmately compound, lanceolate shaped with pinnate venation and is bluish-green to green in color (Gilman and Watson, 1994). The *Vitex* plant was recently applauded by the nursery industry as a useful landscape plant, however, there are breeding opportunities to improve the ornamental value of this plant (Dirr, 2015). *Vitex* would benefit from additional breeding in order to develop new characteristics such as a more compact growth habit and additional flower colors.

The long-term goal of this research is to breed and improve *Vitex agnus-castus*. However, the first part is to understand the seed physiology of this plant. The objective of this research was to determine if there are dormancy requirements for the successful germination of seeds from *Vitex agnus-castus* (Bewley and Black, 1982).

MATERIALS AND METHODS

Several experiments were designed to investigate if there are exogenous or endogenous dormancy requirements for the germination of *Vitex agnus-castus* seeds. Five experiments were designed to examine stratification, scarification, scarification + stratification, gibberellic acid treatment, and scarification + gibberellic acid.

1) For stratification, 20 seeds per replication were wrapped in moist paper towels and placed in plastic bags. The bags of seeds were placed in a refrigerator (4°C) for either 4 or 8 weeks. After their treatment, seeds were removed, sown in germination medium in the greenhouse, and evaluated for percent germination.
2) For scarification, 20 seeds per replication were soaked in concentrated sulfuric acid for either 1 or 2 h. After scarification, the seeds were rinsed thoroughly with distilled water to stop the scarification process. The seeds were then sown in germination mix and placed in the greenhouse until germination.
3) For seeds that might have double dormancy, there was an experiment that examined both scarification and stratification. For each replication, 20 seeds were scarified as described in 2 above then they were wrapped in moist paper towels and placed in plastic bags. The plastic bags were placed in the refrigerator (4°C) for either 4 weeks or 8 weeks before the seeds were sown in germination mix and placed in the greenhouse for germination evaluation.
4) For the gibberellic acid test, 25 seeds per replication were soaked in different concentrations of gibberellic acid (GA_3) for 24 h. The concentration tested were 250 ppm, 500 ppm, and 1000 ppm. There were two controls in the experiment: one control was distilled water and another was 19% ethanol. After 24 h, the seeds were sown in germination mix and placed in the greenhouse to germinate.
5) The final experiment tested the effects of both gibberellic acid and scarification on seed germination. For this experiment, 20 seeds per treatment were scarified with concentrated sulfuric acid for different scarification times of 15, 30 and 60 min. After scarification, the seeds were soaked in gibberellic acid (GA_3) with a concentration of 5000 ppm for 24 h before the seeds were sown in germination mix and placed in the greenhouse.

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RESULTS

The best seed germination from the stratification and scarification experiments was 30% for the seeds that were stratified for 4 weeks (Figure 1). The second best germination rate was 20% for seeds that were scarified for 1 hour followed by either 4 weeks or 8 weeks of stratification. The worst treatment, with no germination, was scarification for 2 h followed by 4 weeks of stratification. The second worst treatment for germination was the scarification for 2 hours (Figure 1).

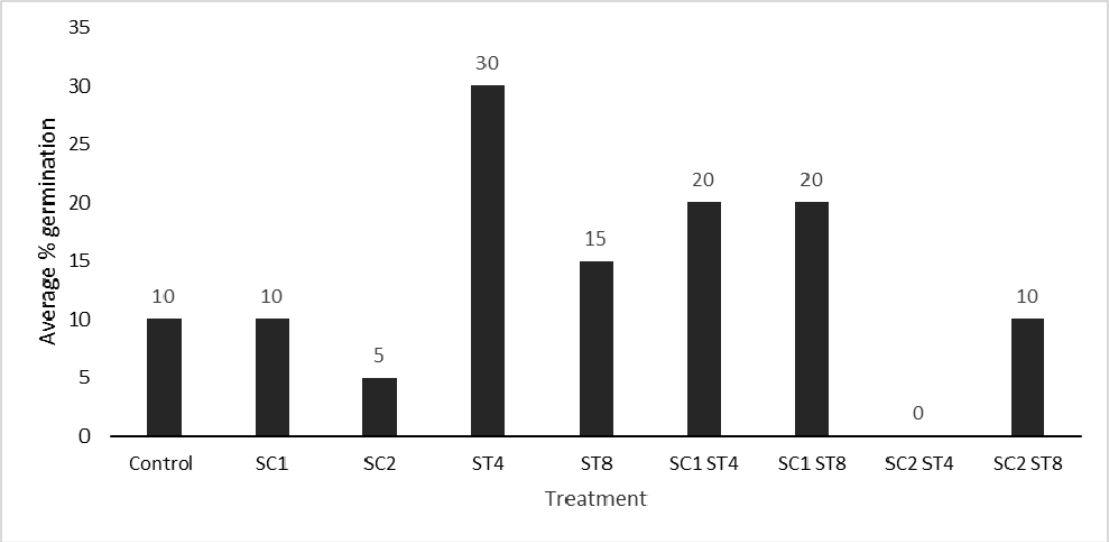

Figure 1. Comparison of different seed treatments to assess dormancy requirements for *Vitex agnus castus*. Seeds with no pretreatment before sowing (control) were compared to those that had been scarified for 1 h (SC1), scarified for 2 h (SC2), stratified for 4 weeks (ST4), stratified for 8 weeks (ST8), scarified for 1 h followed by 4 week stratification (SC1 ST4), scarified for 1 h followed by stratification for 8 weeks (SC1 ST8), scarified for 2 h followed by 4 weeks stratification (SC2 ST4), and scarified for 2 h followed by 8 weeks of stratification (SC2 ST8).

The application of gibberellic acid to *Vitex* seeds did not improve germination when compared to the control seeds (Figure 2). Seeds that were treated with 250 ppm and 500 ppm GA_3 had no better germination percentage than the control seeds. Seeds that were treated with ethanol or 1000 ppm GA_3, did not germinate.

Seed scarification combined with gibberellic acid treatments had some interesting results. The best germination of *Vitex* seeds was obtained during this experiment, however the treatments were not significantly different from the control seeds (Figure 3).

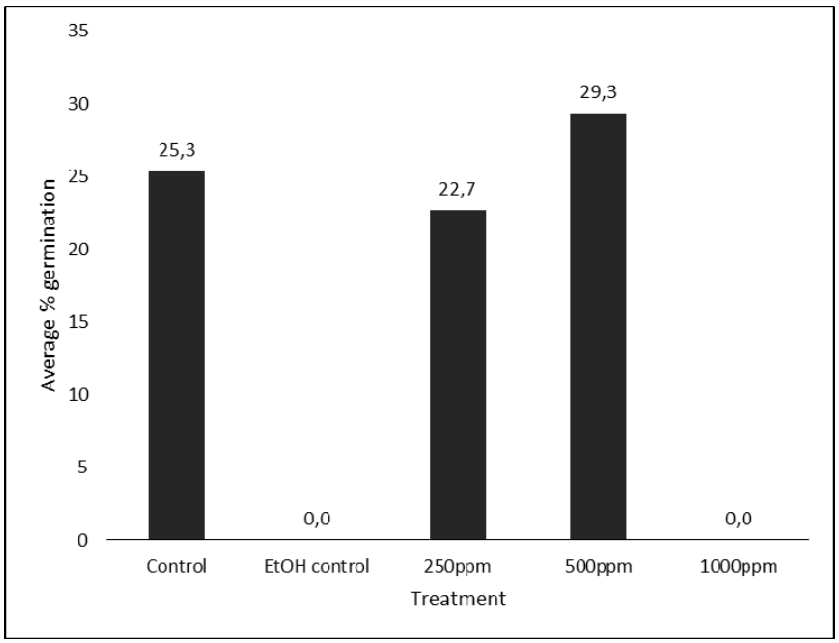

Figure 2. Effect of different rates of gibberellic acid (GA_3) or ethanol on the percent germination of *Vitex agnus-castus* seeds.

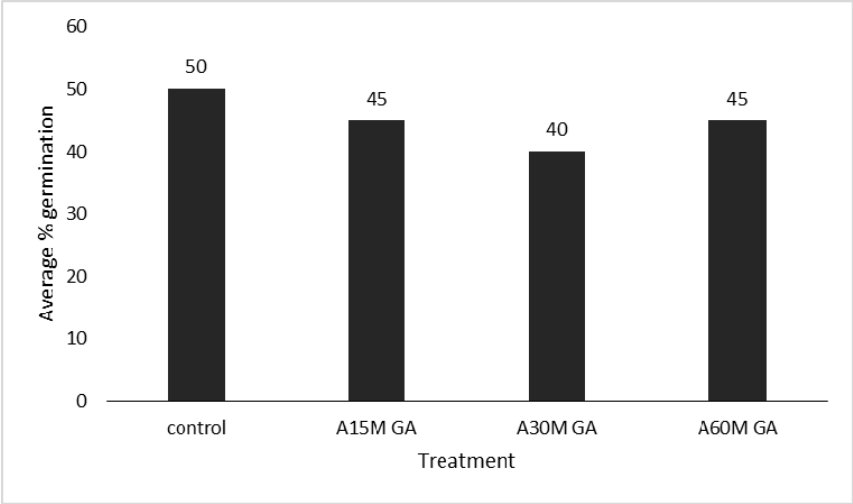

Figure 3. Average percent germination of *Vitex agnus-castus* seeds when soaked in 5000 ppm gibberellic acid (GA_3) followed by scarification for 0 minutes (control), 15 minutes (A15M GA), 30 minutes (A30M GA), and 60 minutes (A60M GA).

SUMMARY

There are many factors that affect seed germination and this research demonstrated that there is more to learn before the factors that are necessary for uniform and reliable seed germination of *Vitex agnus-castus* are fully understood.

These experiments demonstrated that the factors that affect seed germination of *Vitex* are unclear and complex. The seeds responded to scarification, stratification, and gibberellic acid treatments. This suggests that there might be a dormancy factor that plays a role in the germination of their seeds. However, the greatest average percent germination of all treatments was only 50%. When all of the different treatments (Figures 1-3) were compared, it appears that some conclusions can be made: (1) scarification of the seeds for 2 h is too long, (2) treating seeds with ethanol or 1000 ppm GA_3 is not beneficial, (3) there

may be a benefit to treating the seeds with GA_3, and (4) stratification may also be beneficial for enhancing germination.

Because no treatment produced superior and consistent seed germination, no definite and final protocol for treatments of *Vitex agnus-castus* seeds can be outlined. Although there were two sources of fresh seeds that were used for these experiments, it is assumed that the seeds did not have a high level of viability. It is possible that the flowers on *Vitex* plants do not produce large numbers of viable seeds.

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Traditional and in vitro development of new clover (*Trifolium* spp.) plants©

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Abstract

The objectives of the study were to cultivate and breed *Trifolium repens*, grow and micropropagate various species of *Trifolium,* and develop protocols for the genetic manipulation of *T. repens* in vitro. Because white clover is a self-sterile hermaphrodite, cross-pollination is necessary to create viable seed from genetically different parents. As a result of exposing *T. repens* to 6-benzylaminopurine (BAP) in vitro, adventitious shoot formation was initiated and it was observed that a concentration of 1 mg L^{-1} BAP is optimal for adventitious shoot initiation. Other species of *Trifolium* responded similarly to that of *T. repens* while cultivated in vitro. Colchicine and Surflan® (chemical mutagens) were used successfully to produce mutations in *T. repens*. The plants exposed to these mutagens demonstrated physical mutations such as an increase in leaflets per clover and thicker petiole tissue. This research provides evidence that plant tissue culture can be used to micropropagate endangered *Trifolium* species and chemically induced mutations which resulted from this study.

INTRODUCTION

The clover plant, *Trifolium* spp., comes in all shapes, sizes, and colors and provides benefits to the surrounding flora and fauna. These benefits include the fixation of nitrogen, favorable nectar and nutrient density, and potential for growth as an ornamental plant. This study utilized plant tissue culture for the development of new clover cultivars as well as for the establishment and micropropagation of several clover species, both common and endangered, in vitro. Research into the manipulation of white clover plants in vitro is limited. Therefore, the methods that were used to establish clover plants in tissue culture were based on responses of other plants in vitro (Kyte et al., 2014).

As a nitrogen fixer, clover can convert freely available nitrogen into ammonium compounds through nodules in its roots. This unique and valuable trait is a result of a symbiotic relationship between the clover plant and *Rhizobia* bacteria present in the soil (Frame, n.d.). White clover is also rich in a number of essential nutrients such as calcium, phosphorus, magnesium, potassium, and protein (Søegaard, 1993). Furthermore, white clover has been found to improve the daily gains of cattle (Hoveland et al., 1991), to benefit surrounding plants (Parente and Frame, 1993), and to correlate with an increase in carrying capacity of a pasture for deer (Stevens et al., 1992).

In a study by Quesenberry (2002), the importance of several species of clover was evaluated throughout the United States. Quesenberry (2002) estimated that the amount of nitrogen fixed by clover in the United States for 1 year would equate to $525 million based on the price of nitrogen in the form of NH_4NO_3. Quesenberry also estimated the value of good quality red clover hay to be a $6.4 billion dollar market.

White clover is a stoloniferous plant which branches out from growth nodes along its stolon. From these nodes, roots may form on the surface which is in contact with the ground; offshoots, or runners, may branch out as a form of asexual reproduction; and/or petiole and leaves may form (Figure 1). White clover, *T. repens*, is known for having white flowers and moderately sized leaflets with a distinguished white marking on its leaflets (Frame, n.d.). Red clover, *T. pratense*, is similar in structure to white clover in that it is stoloniferous,

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however, red clover is less of a cover crop and it tends to grow significantly higher off the ground than white clover. Red clover is also known for having red colored flowers as opposed to white clover's white to pink flowers (Brickell and Zuk, 1997).

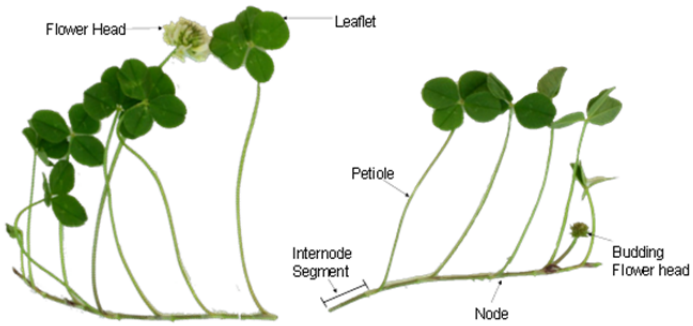

Figure 1. Structure of a white clover plant.

Driven by its benefits and potential as an ornamental plant, Lee (2007) created a new cultivar of white clover which was produced through mutagenic exposures; the new cultivar of clover was patented under the name 'Lucky Together'. By using the mutagen ethyl methanesulfonate (EMS) alongside hormones and growth regulators, Lee was able to develop a new cultivar of clover with aesthetically pleasing qualities, tolerances to Korea's environment, as well as the benefits that clover previously offered. Lee's research provides evidence to support the idea that random mutations in plants as a result of chemical exposure can yield characteristics which have aesthetic appeal as well as practical uses for society (Lee, 2007).

Bae et al. (2009) also successfully mutated and bred white clover for specific traits. One of the traits bred were multifoliate clovers. Multifoliate clovers have greater than three leaflets. The four-leaf clover is the most common naturally occuring multifoliate clover. Bae et al. (2009) used gamma radiation to mutate clover seeds while the seeds were in their developing stages. After exposure, the surviving plants were left to grow and 11.7% of the surviving population exposed to 25 Gy exhibited mutation in leaf number compared to the control which had 0% mutation in leaf number. The plants that expressed mutations in leaf number were then cultivated specifically for the multifoliate trait (Bae et al., 2009).

Polyploidy has also been induced in plants as a result of mutagen exposure. In a 2008 study, polyploidy was achieved by exposing *Rhododendron* seedlings to Surflan®, an herbicide, which contains 40.4% oryzalin, a known chemical mutagen (Jones et al., 2008). Colchicine and oryzalin are both known mutagens and have been observed to cause random mutations such as polyploidy and mixoploidy. Colchicine is a spindle fiber interrupter and is known for its use outside of the agricultural world for treating gout inflammations (Schlesinger et al., 2006). Polyploidy and mixoploidy have been recorded as a result of exposing plants to oryzalin and colchicine by Ascough et al. (2008) and Schlesinger et al. (2006). These studies further support that the chemical mutagens, oryzalin and colchicine, are capable of producing random mutations in plants.

Phenotypic mutations, i.e., physically apparent mutations, in clover plants can be observed in nature as well. One of such mutations is multifoliate clovers. Multifoliate clovers are the foundation of the legend of the "lucky" four-leaf clover.

This study approached the primary steps for development of new clover cultivars differently than previous studies. Chemical mutagens, oryzalin and colchicine were used to mutate clover plants in vitro rather than using radiation or EMS. This study also worked toward the establishment of protocol to micropropagate selections of clover, both common and endangered, in vitro. The methods that were used were derived from the existing research concerning clover plants, as well as the information which has been around on growing plants in vitro since the beginning of the 20th century (Kyte et al., 2014).

MATERIALS AND METHODS

Traditional cultivation of white clover

White clover seeds were scarified using 400 grit sandpaper to remove part of the seed coat and allow water to more readily initiate germination. The scarified clover seeds were set in petri dishes containing a paper filter and water and remained in the dishes until there were visible signs of germination (1-5 days). As clover seeds germinated, they were individually transferred into plastic greenhouse trays containing a 1:1 ratio of potting mix to sand medium. They were grown for 8-10 weeks, the point at which the plants began to flower.

When the white clover plants began to flower, those plants that expressed unique phenotypic characteristics were separated and transplanted into plastic pots. The plants were crossbred by transferring pollen from one floret to the stigma of another floret on a genetically distinct plant with a toothpick. Seeds were harvested 3-5 weeks after transferring the pollen.

Sterilization of clover seeds

Accepted standards of aseptic technique were followed while working under a laminar flow hood equipped with a HEPA filter. To be sterilized, seeds of white clover were placed into glass beakers, submerged in 95% ethanol, agitated with a swirling motion for 30 seconds, and then had the ethanol decanted. The seeds were set aflame to burn off remaining ethanol and scarify the seeds. This flaming process was repeated three times for each set of seeds to ensure sterility. Once completed, the seeds were aseptically transferred onto ¼ strength Murashige and Skoog medium in disposable petri dishes, labeled, and placed in the growth chamber to germinate.

In vitro growth regulator trials

Full strength Murashige and Skoog (MS) medium (Murashige and Skoog, 1962) was prepared in glass culture tubes with the following concentrations of the cytokinin 6-benzylaminopurine (BAP): 0, 0.125, 0.25, and 1.0 mg L^{-1}. The pH of the media were adjusted to a range of 5.7-5.8, agar was added 7 g L^{-1}, and culture tubes were autoclaved.

White clover plantlets were aseptically transferred into tubes of the four different BAP concentrations; there were 16 replications used per treatment. The plants were placed in a 22°C growth chamber in a randomized complete block design. After four weeks the plants had their roots and foliage removed and were subcultured onto fresh media with the same levels of BAP. After a total of 8 weeks the plantlets were removed and their shoot numbers were counted (Figure 2).

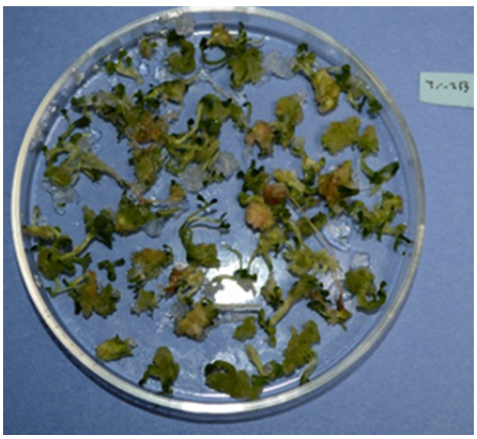

Figure 2. White clover plants produced in vitro on a Murashige and Skoog (1962) medium containing 1 mg L^{-1} 6-benzylaminopurine.

Chemical mutagen studies

1. Surflan application.

Once aseptic white clover seeds germinated, 8-day old seedlings were exposed to varying concentrations of Surflan (40% oryzalin) for three different times under the protection of a laboratory fume hood. The Surflan concentrations that the plants were subjected to were 0.1%, 0.5%, 1.0%, and a control with 0 Surflan. Plantlets in each of these four treatments were exposed to the mutagen for 30, 60, and 90 minutes. There were a total of 144 seedlings that were subjected to these treatments. Following their respective treatments, seedlings were rinsed in sterile water and remained in sterile water until subcultured into Magenta™ GA-7 culture vessels containing ¼ strength MS media; there were five seedlings per magenta. After 4 weeks, the plants were subcultured onto fresh media. After another 4 weeks had elapsed, the plantlets were subcultured again, but this time individually into test tubes containing ¼ strength MS after having their roots and foliage removed. Data on the survival of the plants exposed was recorded 8 weeks after their exposure. Approximately 5 weeks after the subculture into test tubes, the surviving plantlets were transplanted into plastic cell packs containing a perlite-peat based medium and moved to the greenhouse.

2. Colchicine application.

The other mutagen used in this study was colchicine. It was administered to the plants via the growing medium; a different manner than the Surflan. The four solutions of colchicine had concentrations of 0, 0.1, 0.5, and 1.0 g L^{-1} colchicine.

The colchicine solutions were filter-sterilized through a 0.45 µm hydrophilic cellulose acetate membrane filter. One liter of one quarter strength MS medium with 7 g L^{-1} agar was prepared and poured into 250 ml quantities prior to autoclaving. After autoclaving, each of the colchicine solutions were loaded into their respective sterile syringes connected to filter sterilizers and were pumped into the still-liquid, autoclaved media. The colchicine media were then poured into sterile petri dishes and allowed to cool and solidify under the laminar flow hood.

After the colchicine medium were cooled, plants were subcultured onto them. There were four plants placed in each petri dish. The cultured plants were then placed in the growth chamber at 21°C with 24 h lighting. After 48 h, half of the petri dishes were removed from the growth chamber and the plantlets were subcultured onto ¼ strength MS and returned to the growth chamber. Six days after the initial culturing of plantlets into the colchicine media, the plants that remained on colchicine media were subcultured onto ¼ strength media and also returned to the growth chamber. These two exposure periods provided for pulse durations of 2 and 6 days. After 5 weeks, the surviving plantlets were subcultured into culture tubes containing ¼ strength MS after having their roots and foliage removed. After 5 additional weeks, the plantlets were transplanted into cell packs containing a peat and perlite mix and moved to the greenhouse where they were acclimated. Plantlets that initially exhibited unique phenotypic characteristics were identified and rather than moved to the greenhouse, were subcultured onto fresh medium and remained in the growth room.

RESULTS AND DISCUSSION

Traditional cultivation of white clover

Scarification of white clover seeds with 400 grit sandpaper allowed for sufficient removal of the seed coat for improved germination. The scarified seeds began to swell within 24 h after imbibition of water. The cultivation of white clover under the conditions outlined in the methods sustained healthy plants through seed harvest. Hybridization procedures consistently produced 1 to 3 seeds per fertilized ovary.

The successful harvest of clover seeds was delayed due to the self-sterility of clover. The plants that were initially cross-pollinated were clones of each other, so no viable seed

was able to be harvested. It was not until genetically distinct plants were cross-pollinated that viable seed was able to be harvested. These hybrid seeds were harvested 3-5 weeks after pollination, or when the flower head had dried out entirely and the seeds were able to easily fall out of the florets. The seeds ranged in color from yellow to brown but all maintained similar size. The viability of the seeds harvested was verified by scarifying 12 of the harvested seeds and repeating the germination process. Offspring that were grown from hybrid seed and produced from parent plants with multifoliate clovers also produced multifoliate clovers. Some of the offspring raised adopted the distinguished white "v" mark variegation present in the male parent while the remaining offspring expressed the trait with less opaqueness.

In vitro growth regulator trials

The results from these experiments demonstrate that the concentration of 1.0 mg L^{-1} BAP was most effective in the induction of the greatest number of adventitious shoots on white clover plants (Table 1). The BAP concentration of 0.5 mg L^{-1} was also an acceptable level to successfully micropropagate white clover. Statistical T-tests with these data showed the strongest statistical difference (P=0.05) when plants were exposed to 1.0 mg L^{-1} BAP (Table 2).

Table 1. Average number of shoots produced on clover plants that were grown in vitro on media with different levels of 6-Benzylaminopurine (BAP). Standard deviation (STDEV) and standard error (SE) are also shown.

	0 mg L^{-1}	**0.25 mg L^{-1}**	**0.5 mg L^{-1}**	**1.0 mg L^{-1}**
Mean	3.6	6.7	12.5	19.8
STDEV	1.2	5.2	12.7	12.9
SE	0.3	1.3	3.2	3.2

Table 2. T-test results on shoot production of clover plants growing on three different levels of 6-benzylaminopurine (BAP). P=0.05.

	25 mg L^{-1} BAP	***0.25 mg L^{-1} BAP***	**10 mg L^{-1} BAP**
0.0 mg L^{-1} BAP	0.0335	0.0136	0.0012
0.25 mg L^{-1} BAP		0.1.48	0.0012
0.5 mg L^{-1} BAP			0.1164

The information that was collected from this study with white clover allows for its implementation into a plan for the micropropagation of other clover plants. This was especially helpful for the clonal propagation of the new clover plants that were created as a result of this mutation breeding study (Figure 2). The in vitro cultivation of different species of *Trifolium* was performed in the same manner as white clover. The plants that were initiated and maintained in vitro using 1 mg L^{-1} BAP on MS medium were *T. incarnatum, T. wormskioldii, T. dichotomum* (syn. *amoenum*), and *T. pratense*. The endangered plant *T. dichotomum* also responded well in vitro. These results suggest that the methodology that was developed here can also be successful with all other species of *Trifolium*.

Surflan application

During the early stages of growth, all of the plantlets that were subjected to Surflan, except for the control plants, exhibited stunted growth. After 3 weeks, the 0.5% and 1.0% levels of Surflan killed at least 50% of the treated plants (Figure 3). An LD_{50}, or median lethal dose, is used as an indicator when mutations can be induced by a chemical. However, no physical mutations were observed in the plants exposed while in vitro. Later, as the plants grew to maturity, they expressed very few phenotypical mutations. Because not all mutations are phenotypically expressed, these results cannot guarantee that no mutations

occurred to these plants.

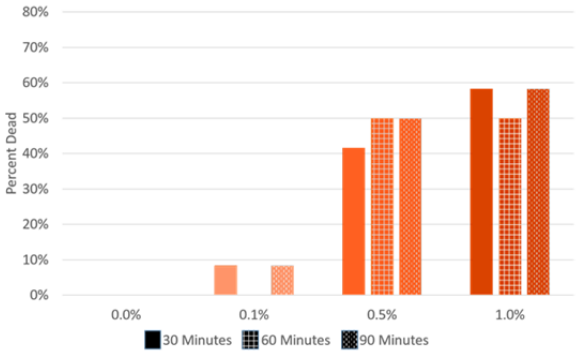

Figure 3. Survival rate of white clover plants 3 weeks after exposure that were subjected to three levels of Surflan for 30, 60, and 90 min.

Colchicine application

White clover plants that were subjected to 0.05% and 0.1% colchicine, whether for 48 h or 144 h, had greater than 50% mortality (Figure 4). As mentioned earlier, an LD_{50} is used as an indicator when mutations can be induced by a chemical. The plantlets that were exposed to colchicine at either doage time expressed minimal physical mutations in vitro. However, there was one physical mutation that produced a genetically stable six-leaf clover plant (Figure 5).

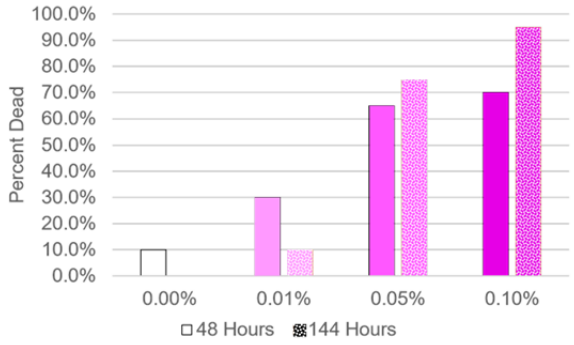

Figure 4. Survival rate of white clover plants 5 weeks after in vitro exposure to three concentrations of colchicine for 48 and 144 h.

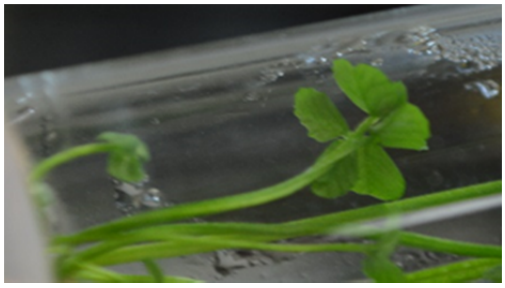

Figure 5. Colchicine-induced six-leaf white clover that was produced after being subjected to 0.1 g L^{-1} colchicine for 48 h in vitro.

Out of the two different approaches to mutagen exposures, the only one to produce a phenotypic mutation of significance was colchicine. The physical mutation induced was a six leaf clover (Figure 5). This mutation provides evidence that the 0.05 and 0.10% colchicine

concentrations in vitro were successful in the creation of a mutation in white clover with physical expression. It appears that the best LD_{50} concentration of colchicine lies between 0.01 and 0.05%.

CONCLUSIONS

The traditional propagation of clover plants by seeds was less efficient than their micropropagation in vitro. Traditional cultivation requires more resources, space, and handling time, whereas clover plants in vitro only require the confined space of a culture tube, the media in the tubes, and subculturing every 4-5 weeks. Propagation in vitro on media including the cytokinin BAP was more effective because a far greater number of clonal plants were able to be produced from one plant.

Propagation of clover plants by cross pollination produced genetically distinct plants and the mutation of clover plants with Surflan and colchicine also had limited success. The mutagens inflicted some degree of mutation to the plants that were exposed, but did not produce mutations that were aesthetically attractive. Plants that were subjected to Surflan had a stunting of growth and thickening of tissue. One attractive and interesting mutation, a multifoliate 6-leaf clover, was produced with colchicine.

Since the mutations that are induced by the chemicals are random, any aesthetically desirable trait being attained from the exposure would also be random. Further research with a greater number of plants would improve the chances of a successful mutation. Any unique plants that are produced, could then be propagated and bred for additional unique traits. This research has outline several techniques that can be used for this purpose.

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Non-grafted and grafted seedless watermelon transplants: comparative economic feasibility analysis©

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Abstract

The use of grafted watermelon (*Citrullus lanatus*) transplants is becoming increasingly popular as an alternative strategy to manage soilborne disease in the USA. The inherent challenges and costs of producing grafted watermelon transplants include: additional greenhouse space that is needed to grow the rootstock and to graft the plants; extra labor that is needed to perform the grafting; and special facilities that are required for the proper healing and acclimation of the grafted seedlings. These facilities range from relatively inexpensive modified greenhouses to state-of-the-art climate-controlled growth chambers. The objectives of this study were to provide a general guide for evaluating the feasibility of growing grafted greenhouse seedless watermelon transplants, and using grafted transplants to produce seedless watermelon in Washington State. Data on grafting supplies and labor were obtained from related studies at the Washington State University, Mount Vernon Northwest Washington Research and Extension Center. Greenhouse production costs were estimated from a composite of information gathered in 2014 from growers in eastern Washington and Oregon who produce non-grafted transplants. Data on crop yield resulting from field utilization of grafted transplants were obtained from WSU field experiments in eastern Washington. Enterprise budget analysis was employed to estimate the costs and returns of producing non-grafted and grafted transplants in a greenhouse. Data from Galinato, Miles, and Wimer (2014). "2013 Cost Estimation of Producing Seedless Watermelon in Eastern Washington" WSU Ext. Pub. FS150E were used and adjusted to reflect 2014 prices. A partial budget framework was used to calculate the net change in profit that can be expected from the field utilization of grafted transplants. Results suggest that the production of grafted watermelon transplants can be economically feasible for commercial greenhouse propagators if the transplants can be sold at more than $0.20/plant. The extra cost of grafted transplants can be acceptable to watermelon producers if using these transplants would provide a viable alternative to field fumigation and improve crop yield. From the watermelon producer's perspective, use of grafted over non-grafted transplants will be primarily based on the benefits gained from the effectiveness of grafted transplants as an alternative to chemical use in managing soil-borne disease. Benefits include reduced overall costs, improved yield, and maintained or augmented profit relative to using non-grafted transplants.

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Grafted tomato propagation and production: relative seedling vigor, graft compatibility, and on-farm yield of 23 cultivars©

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INTRODUCTION

Rootstock (RS) and scion cultivar selection is the first step in preparing grafted plants. Propagators must consider the relative vigor of seedlings before they are grafted and RS-scion compatibility. Ultimately, cultivars are chosen based on their performance on farms. Grafted plants will be made and used more widely and effectively when research-based information on seedling vigor, cultivar compatibility and plant performance is more abundant and accessible.

The hypothesis was that seedling vigor, graft success and/or grafted plant performance (yield) on farms differed among RS and scion cultivars and their combinations.

We tested this hypothesis by documenting: (a) the growth rates of seedlings of 18 RS and 5 scion cultivars, (b) the percentage of healthy grafted plants representing all 90 RS-scion combinations, and (c) their performance on farms.

MATERIALS AND METHODS

Cultivar selection

Tomato RS and scion cultivars were selected based on grower nomination and experimenter assessment of cultivar traits. Communication with growers was facilitated by organic certifying agencies, grower associations, farmer groups, trade publications, and digital media. Selection was made from commercially available rootstocks (66 total) developed by 19 companies and contained approximately 24 disease resistance packages.

Eighteen RS cultivars were chosen representing grower interest, 12 companies, and 12 disease packages. Five scion cultivars were chosen representing hybrid and heirloom and round- and oblong-fruited types.

The greenhouse experiment was repeated twice February-April 2014 at the OARDC in Wooster, Ohio to monitor seedling vigor and graft compatibility. The on-farm evaluation was conducted in April-November 2014 on 31 cooperating farms.

Seedling vigor

Forty-eight seed of each cultivar were sown in a half 96-cell tray as a unit with three units as three replications. Four plant and two environmental variables were measured from 4 to 26 days after seeding. Emergence was recorded daily from day 4 to 14 and day 4 to 13 after sowing in run 1 and 2, respectively (beginning from the appearance of at least one hypocotyl hook and concluding when counts did not increase for two consecutive days for all cultivars). Three representative plants from each unit were destructively measured 18 days after sowing. Aboveground dry weight was measured by a MS3002S Precision Balance (Mettler Toledo, Greifensee, Switzerland) after drying at 50°C for 2 days (Fisher Scientific™ Isotemp™ oven). Stem diameter was measured at 1 cm below the cotyledon by a Traceable® digital caliper (Control Company, Friendswood, Texas). Leaf area was measured by a LI-3100C area meter (LI-COR Biosciences, Lincoln, NE). These parameters were used to calculate cultivar-specific vigor values.

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$$\text{Vigor} = \frac{[\text{above ground dry weight (mg)} \times \text{stem diameter (mm)} \times \text{leaf area (cm}^2)]}{(T_{90} \times \text{GDD} \times \text{PAR})} \times 10^7$$

where T_{90} = the number of days to 90% emergence, GDD = growing degree days (10°C Tmin, 27°C Tmax), and PAR = photosynthetically active radiation. GDD and PAR represent these variables accumulated by 18 days after sowing.

Compatibility (graft success)

Eight grafters were recruited and trained, and then their success with all RS-scion combinations and self-grafted plants as the common control was recorded each day. A total of 2,904 plants were grafted using the cleft grafting method on 10 days. Plant survival was evaluated 2 weeks after grafting; plants with a completely wilted scion were rated as dead and others as living.

Grafted plant performance on farms

Growers nominated their farms as study sites. More than 1,000 grafted plants representing all 90 RS-scion combinations were provided to 31 growers in 13 states. Growers provided subjective and objective information on grafted plant performance.

RESULTS

Seedling vigor

Vigor 18 days after sowing varied significantly among cultivars (Table 1).

Table 1. Vigor results for cultivars.

Cultivar (listed from least to most vigorous)	Aboveground dry weight (mg)	Stem diameter (mm)	Leaf area (cm^2)	Emergence (T_{90})	Vigor value
Trooper	20.4	1.1	10.9	9.4	23.2
Shield	31.4	1.6	16.2	6.9	103.0
Aiboh	36.1	1.6	15.0	6.5	117.8
Estamino	34.9	1.5	23.5	8.5	124.3
Supernatural	42.7	1.7	18.0	7.3	151.1
Aooni	43.2	1.6	21.2	8.6	151.1
• Cherokee Purple	36.0	1.7	16.9	5.7	154.4
• Brandywine	44.3	1.9	22.4	6.2	263.2
• Better Boy	53.7	1.7	21.6	6.4	269.7
• Celebrity	50.1	1.8	24.5	7.0	276.3
RST-105	49.2	1.8	28.5	7.9	279.8
Resistar	44.8	1.9	24.3	6.4	282.6
RST-106	54.3	1.9	26.7	6.4	383.1
Akaoni	61.1	1.9	26.9	6.6	405.4
Cheong Gang	58.0	1.8	27.5	5.8	437.8
BB	58.3	2.1	27.7	5.9	515.5
Armada	72.5	2.1	27.1	6.2	596.1
Stallone	71.1	1.9	30.6	5.6	635.6
Beaufort	66.5	1.9	28.6	4.2	772.3
• San Marzano 2	79.5	2.3	32.2	6.5	794.6
Arnold	82.4	2.0	35.8	4.0	1312.3
Maxifort	93.3	2.1	40.5	5.1	1343.4
Kaiser	110.4	2.0	49.1	4.9	1942.3

• Are scion cultivars; others are RS cultivars.

Graft success

All combinations registered 92-100% grafted plant survival. Survivorship averaged 97% among all RS-scion combinations.

On-farm performance

Qualitative and quantitative information from growers revealed that grafted plant performance varied among RS-scion combinations and farms. Of the seven growers able to compare grafted and ungrafted plants, six growers concluded that grafted plants out-performed their ungrafted counterparts while one grower concluded that the yields of grafted and ungrafted plants were similar. All growers expressed interest in additional research-based information regarding the performance and use of grafted plants.

CONCLUSIONS

Grafting operations should account for variation in seedling vigor and grafter performance. Seeding dates may need to be set by RS-scion combination (e.g., to produce a high percentage of plants with similar stem diameters).

With important exceptions, high rates of grafting success (compatibility) can be expected among the many thousands of possible RS-scion combinations.

Coordinated, local-regional evaluations of grafted plant performance and complementary educational resources are required to enhance the wider and more effective use of grafted plants, perhaps especially among small-midsize organic farms.

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Towards improvement of *Impatiens*©

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Abstract

Common impatiens, *Impatiens walleriana*, have traditionally been the most popular annual flower used for landscaping. However, impatiens downy mildew (*Plasmopara obducens*), a pathogen which has recently become virulent against this species, leaves plants defoliated and commercially unviable. Research was started to identify other species, from a genus of over one thousand, which were more resistant to the disease. Screening identified many species with significantly higher resistance, as well as trends in which species were susceptible. Using a range of breeding and propagation tools, we explored different ways to improve common impatiens and integrate the resistance we identified. These included making efforts to better characterize the available germplasm, ploidy manipulation, tissue culture, and interspecific hybridization. Here we describe existing techniques for impatiens improvement, as well as the modifications we have developed for them.

INTRODUCTION

Impatiens is one of the most popular annual bedding plants and have traditionally been an important source of income for many American greenhouse growers. Unfortunately, in 2004, commercial plants of the most common impatiens species, *Impatiens walleriana*, were reported as being completely defoliated by a new race of impatiens downy mildew, *Plasmopara obducens* (Wegulo et al., 2004). By 2011 the pathogen had spread worldwide and become a significant problem in the landscape. This disease results in wilting, leaf and flower drop and ultimately death of this important bedding plant. As older samples of the pathogen have been identified on the native North American jewelweeds (Saccardo, 1888), *I. capensis* (syn. *I. fulva*) and *I. pallida*, there is also some concern about whether the pathogen could impact native North American ecosystems. However, since the genus *Impatiens* contains more than one thousand described species, an investigation has begun into the general degree of susceptibility to this disease, and whether factors correlated to resistance can be identified. Reports on the disease have already recognized that New Guinea impatiens, *I. hawkeri*, exhibit a high degree of resistance (Cunnington et al., 2008). However previous research has also shown that *I. hawkeri* has a very low success rates in crosses with *I. walleriana* even after embryo rescue (Arisumi, 1985) and that the few hybrids produced and which make it to maturity are abnormal and weak (Arisumi, 1987), limiting its use in resistance introgression.

The original goal of this research was to identify sources of resistance to downy mildew that were also cross-compatible with *I. walleriana*. As we accumulated more species and cultivars, it became clear that the genus *Impatiens* has a lot to offer beyond its most well-known representative. Therefore we expanded our research goals to also incorporate other methods of working with this diversity, and hopefully producing something commercially viable. Many of the same techniques to increase and harness diversity are applicable to all of the germplasm in our collection. By diversifying the commercial genepool, we hope to broaden people's concept of what an “impatiens” is and help bring them out of the shade and into the limelight.

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AREAS OF INVESTIGATION

Germplasm acquisition

The majority of the roughly 200 species we have are donations from private collectors. Descriptions and observations of some of the species used in our research can be found in Table 1. Through funding from a USDA SARE grant (#GNE13-063) we also have purchased other species from a range of commercial sources. Unlisted cultivars or species were purchased from online retailers and small, hobbyist garden centers. The USDA's Ornamental Plant Germplasm Center has also recently acquired a range of accessions, mostly representing the native North American jewelweed species, but also with a few species from further afield.

Table 1. Primary *Impatiens* species investigated during this research. Cytological data compiled from Goldblatt and Johnson (1979) and Yuan et al. (2004).

***Impatiens* species**	**2N**	**Description**	**Observations**
arguta	18, 20	Generally recumbent plants; flowers tubular, ranging dark to light purple, a white form is also available.	Reported hardy to USDA Zone 7.
balfourii	14	Plants generally 2-3 ft tall; flowers profusely, petals are purple and white; forms self seed readily.	1 month stratification; cuttings unsuccessful
balsamina	14	Plants 3-4 ft tall; many color forms; generally cleistogamous.	Fragrant, germinates within 1 week.
campanulata	20	Large, flat leaves; small white flowers with red spots, borne profusely.	Flowers year-round
capensis	20	Flowers generally orange with varying degrees of red spotting, unspotted forms are also common.	Native to North America; asexual propagation unsuccessful; 4-5 month stratification
cinnabarina	16	Plant 2-3 ft tall; leaves heart-shaped	Attractive form; flowers continuously
glandulifera	18	5 ft+ tall; tubular purple flowers.	1 month stratification; fragrant (melon).
grandis	20,36,40	Attractive flat leaves; flowers large and flat, generally white with red spots.	Difficult to flower; buds are very sensitive.
hamata	14	Small pink and white flowers.	Leaves always curled; a very weak plant.
hawker	16, 32	Many color forms available.	Slow to germinate.
hochstetteri	16	Small pinkish flowers borne in profusion; plant forms a trailing mound.	Might be a good filler plant; very pretty.
irvingii	14	Trailing; pubescent; flat; mauve flowers.	Seedpods resemble *I. balsamina*
laurentii		Plants forming sparse mounds 2-3 ft; flowers violet and flat.	Self-seed prolifically; seed may have a short dormancy requirement.
niamniamensis	32	Flower petals green with an enlarged spur.	Roots readily; free from most pests and diseases.
omeiana		Stoloniferous plant with short bouts of dormancy; many foliage forms; flowers yellow and tubular.	Flowering appears photoperiodic; reported hardy to USDA Zone 4.
pallida	20, 30	Different shades available in different forms, flowers yellow.	Native North America. 4-5 month stratification; asexual propagation unsuccessful.
pritzellii		Similar to *I. omeiana*, but taller. flowers yellow.	Likely hardy to USDA Zone 7. flowers more reliably than *I. omeiana*
repens	14	Prostrate, vining, red stems with small, rounded leaves; yellow flowers borne irregularly.	Very attractive foliage plant; good in green walls.
sodenii	16	5-7 ft tall; leaves whorled; flowers white or pink; several forms available.	Variability in self-seed set and floriferousness between forms.
usambarensis	16	Recumbent to slightly mounding; flowers red.	Leaves prone to thrip damage.
walleriana	16	Most common commercial species; many forms available.	Many forms not commercialized with different foliage and growth habits; rarely gets mite damage.

Interspecific hybridization

This approach, more than any other, has formed the basis for our improvement program. Toru Arisumi, a USDA scientist tasked with improving the genus *Impatiens* back in the 1970s, published extensively on his ovule-rescue techniques and the resulting hybrids (e.g. Arisumi 1973, 1977, 1980, 1985, 1987). We have modified our approach from his based on subsequent studies by other researchers. Han (1991) found that changing the carbohydrate source from sucrose to glucose resulted in better germination of rescued embryos. Later, Han (1994) demonstrated that the addition of glutamine to the medium resulted in higher survival rates of embryos. As browning persisted in our experiments, we also looked into using vitamin C as an antioxidant source. However, vitamin C as ascorbic acid tends to change the medium pH and can also deteriorate into an oxidant over time; something we have countered by using calcium ascorbate instead. These findings were incorporated into our medium recipe, although the substitution of glucose produced a softer medium that had to be amended with high concentrations of the gelling agent.

In addition to changes in medium, we have also explored changes to the actual technique of pollination. Initial crosses were done without emasculation, but resulted in self-pollinations in quite a few species. *I. balsamina* is particularly difficult, as it begins shedding pollen before the buds have developed color. The dissection required for bud emasculation also causes *I. balsamina* to release a range of browning chemicals; presumably phenolics. We now emasculate for almost all crosses, unless there is good evidence of sterility or self-incompatibility in the female parent. Pollinated flowers dropping before maturity has been a big problem, and we hoped to improve the retention rate by applying an anti-abscisic hormone to the base of the peduncle. We eventually decided on a commercial preparation for inducing tomato fruit set, "Blossom Set Spray," which contains kinetin. After comparing a few identical crosses done with or without hormones, the drop rate was roughly the same. However, the ovules from the treated flowers did appear quite a bit larger than those from the untreated ones, which may be an avenue worth pursuing in future breeding work.

The days-after-pollination (DAP) that a cross is rescued also appears to play a strong role in its chances of survival. While crosses rescued too early may not be developed enough to grow, crosses rescued too late may have already spontaneously aborted. Arisumi (1980) rescued ovules from a range of DAPs, but found variation in which of these ovules actually developed into seedlings. However, previous work by an esteemed colleague demonstrated that many crosses were not viable prior to 7 DAP (Kendra Hutchins, pers. commun.). We found that in crosses between two Himalayan species, ovules rescued at 10+ DAP showed a conspicuous brown, failed embryo within the ovule while crosses rescued at 7 DAP were uniformly white and appeared viable. In other crosses though, waiting for up to 14 DAP seemed to allow the ovule to develop further without mortality; the species combination in the cross is likely an important factor to gaging the appropriate age DAP.

Most of the crosses we have attempted have been informed by either previous publications on successful crosses (Arisumi, 1987), base chromosome number (conveniently listed for many species on http://www.tropicos.org/Project/IPCN (Goldblatt and Johnson, 1979), or phylogenetic proximity of the species (Janssens et al., 2009; Yuan et al., 2004). Publications list some crosses between rather distantly related species (e.g. *I. uguenensis* [syn. *I. sodenii*] × *I. campanulata* in Arisumi, 1987), but phylogeny and chromosome numbers suggest some potential combinations of closely-related species that have not been attempted yet. One of the groups in which we see great potential, and have had some measure of success with, are the Himalayan species. These are mostly re-seeding annuals, and we have also found that many of them have excellent downy mildew resistance. Little has been documented in attempted interspecific hybrids among these species, but they contain a great diversity of colors and forms. One concern with this group is the potential for invasiveness. Already, *I. glandulifera* is considered a noxious weed across much of Europe (Global Invasive Species Database, 2009), and *I. balfourii* has been suggested to have some potential in that area as well (Schmitz and Dericks, 2010). However, this is one instance where higher sterility levels in interspecific hybrids might be an advantage. Another group great potential are the species from Madagascar and the surrounding islands. Several

commercial series have already been produced from interspecific hybrids among these species, such as the African Orchid series popular among collectors and the recently released Downtown series from Fry Road Nursery. They have also been combined with *I. walleriana* to form the Seashell and Fusion series, created by Burpee and Ball Horticultural respectively (Pitman, 2004). These are popular for combining an *I. walleriana*-like plant with yellow flowers not normally found in the species. While we have found the Madagascar/*I. walleriana* hybrids to be very susceptible to downy mildew, Madagascar hybrids and species on their own seem to possess decent resistance. Many of the interspecifics not involving *I. walleriana* also have at least partial fertility, allowing more complex breeding projects. The major drawback to many of these species is their more cupped flower shape, which resonates with collectors but does not seem to appeal as much to consumers expecting the typical "impatiens shape" (read: *I. walleriana*). This is where *I. laurentii* and the closely related *I. lyallii*, with their flat flowers, might be very useful. Our crosses between these and other Madagascar species have produced hybrids with a flatter flower and wider range of colors. More advanced breeding, such as backcrossing, might improve appearances even more.

Interspecific hybridization has the potential to improve a wide range of traits beyond those explored in our breeding work. One often unexpected side effect of producing interspecific hybrids is the presence of double flowers in the progeny. Arisumi (1987) noted this in hybrids between *I. flaccida* and *I. repens* as well as *I. flaccida* and *I. walleriana*, and we have observed this phenomenon ourselves even between some of the closely related Madagascar species. Fragrance is something not typically associated with impatiens, but in surveys has been reported as the number one most consumer-desired trait in ornamentals (Clark et al., 2013). Quite a few fragrant species of impatiens are commercially available, perhaps *I. tinctoria* most famously, and crosses to bring this trait into a more manageable plant could be very interesting. Perenniality is another trait possessed by a range of impatiens species, with the best hardiness known being in *I. omeiana*, and which could be transferred to showier species. Hybridization with other species to expand the growing range of a species, such as *I. platypetala* has given to *I. hawkeri* for the appropriately named 'Sunpatiens' series or *I. flaccida* has contributed to *I. hawkeri* in the 'Fanfare' series (Guillen, 2002; Pitman, 2004), is another admirable goal. Several impatiens species are native to areas with drought, heat, and high sun, and would be great candidates for pushing the boundaries of impatiens cultivation.

Mutagenesis

Another technique which has great promise for improving impatiens is mutagenesis. While mutagenesis has a bad reputation in the edibles world, due to incremental changes in difficult-to-measure phenotypes (e.g., yield) and concerns about affecting consumers, these are non-issues in the ornamental world: our phenotypes are primarily visual and consumers generally do not actually consume our plants (Schum, 2003). Instead, mutagenesis has great potential to broaden the genetic base of a species without having to collect new populations of the plant from the wild; something that is increasingly challenging in the modern world. Also, mutagenesis allows retention of a given phenotype with modification of only a small handful of traits, rather than the sometimes larger-scale changes brought about by traditional breeding, and without the stigma associated with genetic engineering. This is not to say that exploration and introduction of new germplasm or traditional breeding are any less useful, just that mutagenesis adds another tool to the plant improvement toolbox.

Other researchers have previously published on induced mutagenesis in impatiens. Klozová (1962) used X-rays on *I. balsamina* and found changes in the type and quantity of anthocyanins produced. Bose and Basu (1967) applied diethyl sulfate to *I. balsamina* and reported plants without side-branches, as well as ones with fasciated stems. However, the experiment off of which we have based most of our work is Weigle and Butler (1983). They treated seeds of *I. platypetala* with a range of concentrations of ethyl methansulfonate (EMS) and found that a treatment of 80 mM for 24 h resulted in approximately 17.5% mortality. While this is lower than the 50% mortality they had hoped for in order to get saturated

mutagenesis, they still found several mutated plants including a dwarf form.

We tested a similar protocol on seeds of *I. balsamina.* However, we added a phosphate buffer solution to improve uptake (Kim et al., 2006) and a treatment with sodium thiosulfate afterwards to stop the mutagenesis reaction (Arnason, 1974). Pre-soaking the seeds in the buffer before treatment resulted in good germination but no leaf formation for any of the seedlings, suggesting a high mutation rate. Placing dry seeds into the buffer and treating immediately also lead to good germination rates, and 16 of the 120 treated actually produced leaves, in addition to all 8 of the control seeds. Of these 16, 6 matured into viable plants; although some of the loss here may have come from damping off during the slow maturation of the seedlings. These six showed distinctly different phenotypes from the controls (Figure 1). This method was also used to treat 240 seeds of *I. laurentii*, with 16 controls, but the germination rate of both the controls and the treated seeds was very low; leading us to believe that there may be some dormancy mechanisms in place that would need to be accommodated.

Figure 1. Plants of *Impatiens balsamina* grown from the same lot of seed, untreated (top) and treated (bottom) with EMS. The pink flower color is from segregation in the original population, other traits are presumed to be induced mutations.

Polyploid induction

Originally we started creating polyploid forms of different impatiens species as a way provide resources for our interspecific breeding work. Crossing two polyploid plants provides a complete set of chromosomes for each species, making meiosis smoother and sometimes increasing fertility of the hybrid. Arisumi (1973) documented that while most induced polyploid impatiens were less fertile than their diploid progenitors, inducing polyploidy in interspecific hybrids sometimes restored fertility. His best example of this was in crosses between a species from Java (possibly *I. platypetala*) and a species from New Guinea (likely *I. hawkeri*), where the diploid interspecific hybrids did not produce seeds but the induced tetraploids of the same plants set 11-23 seeds per capsule. While Arisumi (1987) also reported good results in getting interspecific hybrids between *I. walleriana* and *I. niamniamensis*, these hybrids were sterile, and there is no follow-up publication on whether inducing polyploidy might restore fertility. As *I. niamniamensis* is 2N=32 and *I. walleriana* is 2N=16, it would be interesting to recreate this cross with a 4N plant of *I. walleriana* and see if that hybrid had better fertility. If, on the other hand, sterility is desired in a cross, such as in the invasive species hybrids described previously, creation of odd-ploidy hybrids (e.g. triploids, etc.) that divide unevenly can be used to prevent proper

meiosis. Another advantage of polyploidy is larger and more rounded organs, such as flowers (Arisumi, 1973). The 'Bruno' series of *I. walleriana*, a tetraploid line released by Floranova (Uchneat, 2006), took advantage of this, with larger flowers and thicker leaves. Online reviews by gardeners praised the series' resiliency, claiming they grew in a wider range of conditions and tolerated abiotic stress better than diploid plants of *I. walleriana*. However, we have been unable to find a commercial or private source that still carries these.

As we were unable to find commercially available polyploid forms of common impatiens species, we endeavored to create our own. Arisumi (1973) described a technique of treating cuttings topically with a 0.2% colchicine solution, which we also employed. To improve penetration of the colchicine into the tissue we also diluted it with 2% DMSO, as we had previous experience with in other species. However, we found that the addition of DMSO seemed to increase phytotoxicity beyond the amount from the colchicine. Another modification was to use food-grade glycerin to dilute the colchicine in place of water, as it is thicker and less prone to running off the plant. It was unclear whether this actually improved transformation, but the treated surfaces did appear wet longer. One modification we have not extensively tested yet, but with excellent demonstrated potential in other species, is changing the anti-mitotic agent from colchicine to something less toxic and more potent, such as oryzalin.

Of the 15 species and cultivars we treated with colchicine, the best survival appeared to be in cuttings of the 'Xtreme' series of *I. walleriana*, seedlings of *I. balsamina*, and cuttings of *I. flaccida*. Flowers from treated plants showed a marked difference from those of untreated ones (Figures 2 and 3), except for in the case of *I. flaccida* where the two were indistinguishable and leading us to believe they escaped transformation. Some vegetative characteristics were also noticeable, such as thicker leaves in *I. walleriana.* Although the transformation of *I. balsamina* appeared to be uniform throughout the plant, likely due to the single growth point treated on the seedling, treated plants of *I. walleriana* appeared chimeric. Taking cuttings from visually distinct sections ameliorated this somewhat, but did not completely eliminate the variability. In one case, a cutting from a particularly thick-leafed section resulted in a plant which appeared incapable of setting flowers. The ploidy of this sport has not been tested, but we suspect it to be a higher-level polyploid (e.g. octoploid or above).

Figure 2. Flowers from plants of *Impatiens walleriana* treated (right) and untreated (left) with colchicine.

Figure 3. Flowers from plants of *Impatiens balsamina* treated (right) and untreated (left) with colchicine.

Tissue culture

Concurrent with our other work, we have kept an active tissue culture program of *Impatiens* species going. Originally we started this as a way to maintain large or difficult-to-grow species without having to allocate specialized greenhouse space. However, we also found that some species that normally deteriorate at the end of the growing season, such as *I. balfourii*, can also be grown beyond their normal senescence point through micropropagation. We are also hoping to use this as a way to efficiently apply mutagens to species that do not readily produce seeds, such as *I. repens*, but do not have conclusive results from this yet.

Fortunately, there have been several articles published on growing *Impatiens* in vitro. Many of these come from attempts to produce transgenic *I. walleriana* for a variety of reasons, such as resistance to impatiens necrotic spot virus (INSV). We contrasted four media recipes, based on the basal salts and vitamins published by Murashige and Skoog (1962) but with varying amendments (Baxter, 2005; Chou, 2000; Dan et al., 2010; Xiang and Wang, 2005). Each medium produced pronounced differences within each species, but these differences did not always follow from species to species. Routine preparation of media with co-autoclavable hormones, such as thidiazuron, was much easier than using partially heat-labile hormones, such as zeatin, that had to be sterile-filtered and added as the media cooled (Kyte et al., 2013). Based on the growth trends we have observed, we vary our media use to suit our current needs.

Another challenge we faced with tissue culture was that most of the articles published on impatiens use surface-sterilized seeds as the tissue source. This works well for species that readily produce seed and whose seed lack strong dormancy mechanisms, but did not fulfill our needs for other species that fell outside these criteria. Gunapala et al. (2008) tested several chemical formulations for surface-sterilizing plants of *I. repens* and found that mercury chloride yielded the lowest contamination rates. Unfortunately, mercury chloride is very toxic for humans. Instead, we tried a variety of other surface-sterilization techniques, using varying concentrations of several antiseptics. One discovery was that 70% ethanol, a standby in many surface-sterilization protocols, resulted in high phytotoxicity for the species we tested, especially when followed with a commercial bleach treatment. Instead, we began soaking the tissue in sterile water (amended with a few drops of 1N HCl to inhibit bacteria) on a rotary shaker for 2-3 h, following this with 10-15 min. in 3% hydrogen peroxide (to lift surface contaminants), a rinse with sterile water, 10-15 min. in 10-20% bleach, and 3 rinses

with sterile water. The rinse with sterile water between the hydrogen peroxide and bleach treatments was added after we observed vigorous bubbling when adding the bleach solution directly after the hydrogen peroxide, leading to be concerned about adverse chemical reactions. Following the above protocol produces plant material with low rates of contamination for most species. However, several species seem to be particularly prone to contamination even with the above protocol, possibly either due to coarse tissue surfaces or endophytic infection. Using all of these techniques, we have maintained up to 30 species in vitro.

CONCLUSIONS

There is a wide range of techniques available to both improve and utilize the diversity present in the genus *Impatiens*. We have outlined some of the ones that form the focus of our research, but this list is nowhere near exhaustive. Through our work, we hope to create germplasm that researchers with other skill-sets can build from. Interspecific hybrids provide a mechanism to transfer interesting traits between species, a process that can likely be aided by making crosses between individuals at different ploidy levels. Tissue culture allows us to preserve these hybrids, as well as other germplasm we have acquired, and multiply it so that we can create backups and share it more easily. Mutagenesis acts as a way to increase the diversity of species with narrow genetic bases, without the cost or environmental impact of seeking out wild populations. Drawing upon all of these, we have developed impatiens populations that are not only resistant to downy mildew, but also diverse and resilient enough to face new, unknown challenges from the environment, pests and pathogens, or even changing consumer trends. The genus *Impatiens* has a lot to offer already, and we hope that through our work we can make it more accessible.

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Grafted tomato propagation: effects of light intensity and temperature on graft healing and plant regrowth©

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INTRODUCTION

Grafting can improve vegetable productivity by combining desirable traits from two taxa into one plant. However, the grafting process creates severe wounds. Optimal healing of newly grafted plants requires careful light and temperature management (Lee et al., 2010). Grafted tomato plants are commonly healed in enclosed structures shaded to reduce light levels and moderate temperature (Rivard and Louws, 2006). However, what is the optimal combination of light and temperature conditions for efficient healing of grafted tomato plants is not clear. The hypothesis was that light and temperature affect the healing of grafted tomato seedlings separately and interactively.

Objectives were to (a) test the regrowth of grafted tomato seedlings under four levels of temperature and light intensities; (b) heighten the understanding of effects of key environmental variables on graft healing; (c) optimize conditions and management for grafted plant propagation.

METHODS

The experiment was conducted four times in April-June, 2015 at the OSU-OARDC in Wooster, Ohio. Tomato seedlings 'Cherokee Purple' and 'Maxifort' were grown in a greenhouse and grafted using the splice grafting method 3 weeks after seeding. Grafted tomato seedlings were healed under four temperature by light treatments arranged in two growth chambers as a split-plot design. One growth chamber was set at 30/25°C and the other at 25/20°C. Two zones differing in light intensity (50, 150 µmol m^{-2} s^{-1}) were created in each chamber by varying plant distance from the chamber lamps and using open frames covered with shade cloth. The light in both chambers were provided from one Metal Halide 400 Watt lamp (GE Lighting, Inc.) and one High Pressure Sodium 400 Watt lamp (GE Lighting, Inc.). Photoperiod was 12 h from 7 am to 7 pm in all treatments. Relative humidity was controlled at 90% in all treatments for the first 7 days, and reduced to 80% on the 8th day, 60% on the 9th and 10th days. Fifteen to eighteen grafted plants were used per treatment per repeat, placed on a layer of Kapmat. Plants and Kapmat were watered when appeared dry during the study period.

Plant growth was monitored non-destructively immediately after grafting and 10 days after grafting. Leaf area was measured based on digital images taken of five to six plants as a unit and analyzed by WinCAM, which separated the green colors of leaf from the background and calculated the percentage of leaf area out of the known area of analysis. Total plant length was measured from the soil line to the meristem and scion length from the graft union to the meristem by a ruler. Stem diameter at the rootstock and scion was measured by a caliper. Relative growth of the above parameters was calculated as (values 10 days after grafting-values immediately after grafting)/values immediately after grafting ×100%. Besides, plant growth was monitored destructively 10 days after grafting including leaf and stem fresh weight, leaf and stem dry weight after drying in an oven at 50°C for 2 days. Specific leaf area was calculated as leaf area/leaf dry weight. Compactness was calculated as aboveground dry weight/plant length.

Data analysis was performed in SAS. Separate and interactive effects of temperature and light were analyzed by ANOVA. Multiple comparisons were analyzed using the GLM procedure.

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RESULTS

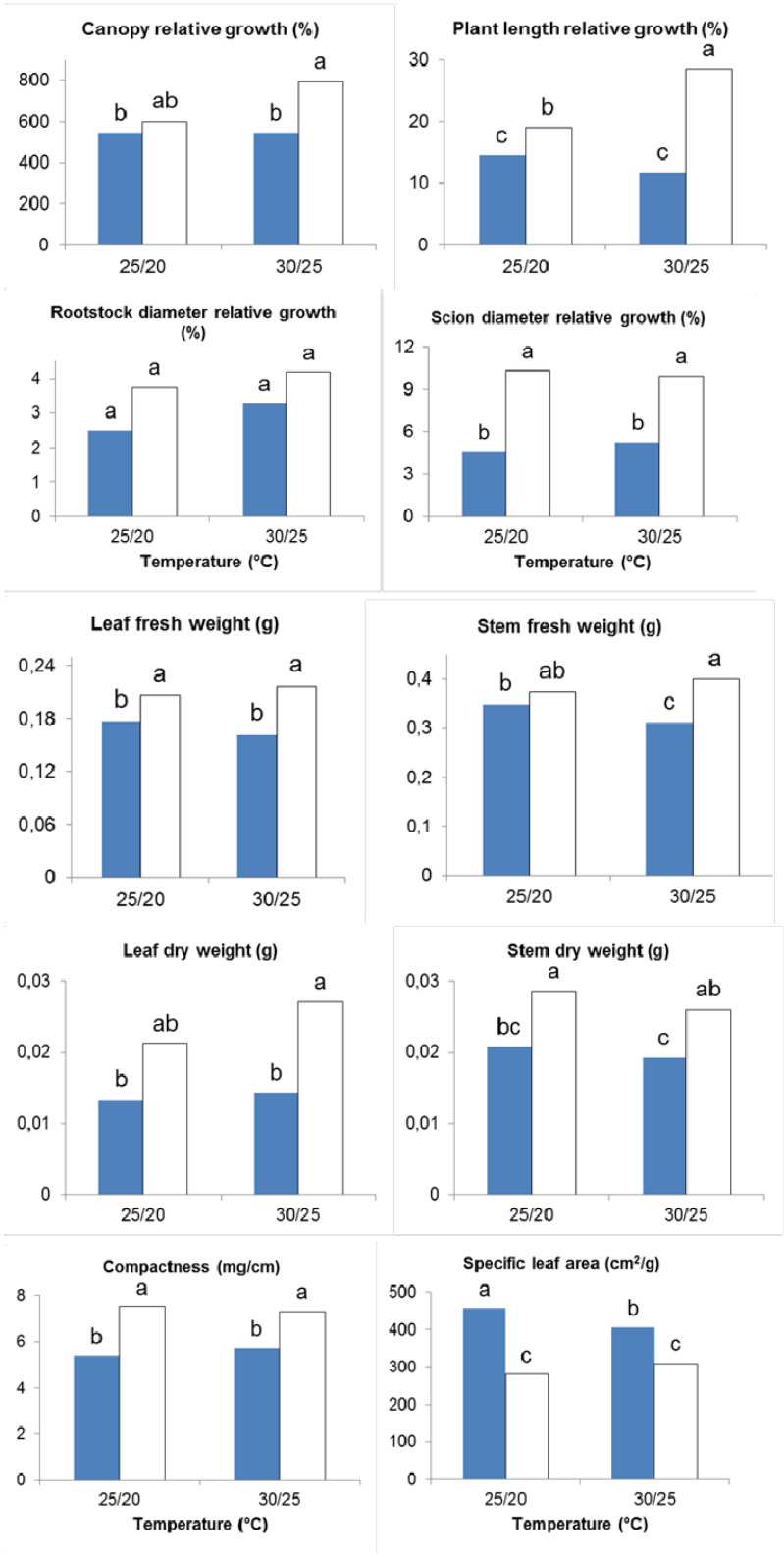

Figure 1. Growth of grafted tomato plants 10 days after healing under two temperatures by two light intensities. Different letters on bars represent significant difference at $p<0.05$. Note: shaded: 50 µmol m^{-2} s^{-1}, not shaded: 150 µmol m^{-2} s^{-1}.

Plant growth was generally unaffected by temperature. The majority of plant growth variables were greater under higher light intensity at 150 μmol m^{-2} s^{-1} compared to those at 50 μmol m^{-2} s^{-1}, while specific leaf area decreased under higher light intensity. The interaction between temperature and light intensity was significant in some healing variables. These results suggest that the conditions under which newly grafted tomato plants are healed warrant further study since increased efficiency in graft healing under optimal conditions is possible.

ACKNOWLEDGEMENTS

This material is based upon work that is supported by the National Institute of Food and Agriculture, U.S. Department of Agriculture, Specialty Crops Initiative under award Number 2011-51181-30963. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture. The work was also funded by SEEDS: The Ohio Agricultural Research and Development Center Graduate Research Enhancement Grant.

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The effect of 6-benzylaminopurine, a cytokinin, on bud-forcing of twelve oak species©

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INTRODUCTION

Oaks (*Quercus* L.) are globally iconic trees, prized economically, ecologically, and aesthetically. However, despite their importance, many species of *Quercus* are under threat from a wide range of global issues (Oldfield and Eastwood, 2007). One method of saving threatened oak species is micropropagation using young, newly flushed shoots collected immediately after emergence in the spring (Kramer and Pence, 2012). This is a narrow and somewhat unpredictable time window for obtaining explants. However, forcing bud break of cuttings can increase the time range to collect young shoot explants and allow for shoot development in a controlled, clean environment (Vieitez et al., 1994). The objective of this experiment was to determine the effectiveness of 6-benzylaminopurine (BAP), a cytokinin (hormone that promotes cell division), on bud break in 12 *Quercus* species.

MATERIALS AND METHODS

Dormant cuttings of 1- and 2-year-old wood were collected from 12 different species of *Quercus*: *alba, bicolor, cerris, falcata, imbricaria, macrocarpa, macrocarpa* var. *macrocarpa* (syn. for *macrocarpa*), *pagoda, palustris, rubra, texana,* and *variabilis*. Cuttings of 10-33 cm in length with 5-25 buds each (depending on species) were collected in Kennett Square, Pennsylvania, in mid-February. The experiment was a factorial design with 12 species, three BAP treatments (0, 100, and 500 ppm), and three replications, giving a total of 108 cuttings. The cuttings were placed into Erlenmeyer flasks with distilled water and placed in a greenhouse with a heat set point of 20°C and a cooling set point of 26.5°C. Cuttings were evaluated weekly and rated on a bud development scale of 0-4 with 0 = no development, 1 = slight bud swelling and elongation, 2 = moderate bud swelling and elongation with visible green coloration, 3 = bud break with partially visible leaf and/or inflorescence tips, 4 = at least one newly emerged leaf fully visible (target stage for shoot tip micropropagation).

RESULTS AND DISCUSSION

Results indicate that overall, the BAP treatment had significant effects on the *Quercus,* but responses varied by species (Figure 1). BAP treatment at 100 or 500 ppm significantly increased the rate of bud break and shoot elongation for four of the *Quercus* species: *imbricaria* (Figure 2)*, macrocarpa*, *pagoda*, and *variabilis*, while significantly decreasing the rate in *Q. falcata*. There was no significant effect from BAP application on the remaining seven species (Figure 3 for *Q. rubra*). All *Quercus* species except *alba*, *bicolor*, and *pagoda* reached Stage 4 with all treatments.

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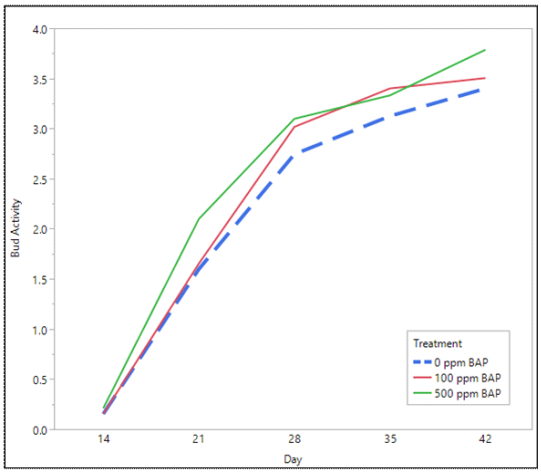

Figure 1. All *Quercus* species — mean bud activity.

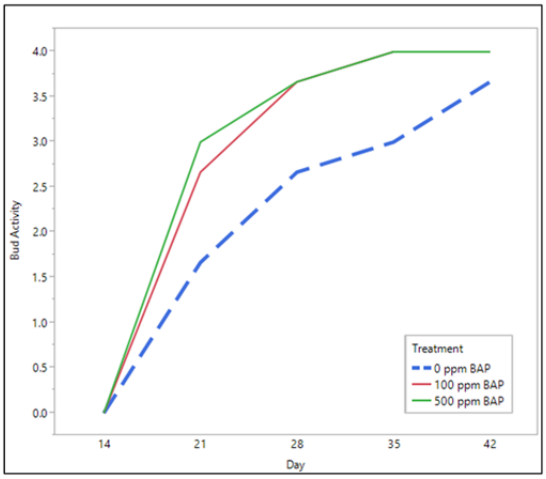

Figure 2. *Quercus imbricaria* (as an example species significantly affected by BAP treatment) — mean bud activity.

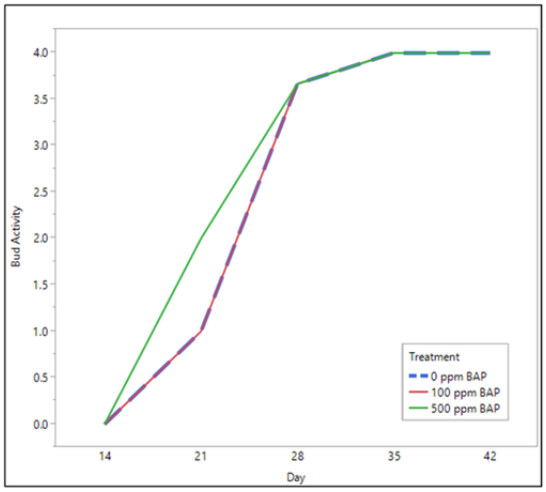

Figure 3. *Quercus rubra* (as an example species not significantly affected by BAP treatment) — mean bud activity.

CONCLUSION

The effect of the cytokinin, BAP, on *Quercus* bud-forcing varied by species and a majority of the species reached Stage 4 with all treatments. This indicates that forcing bud break without BAP application is a viable option, but the rate may be enhanced with some oak species by the application of BAP.

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Techniques for melon grafting©

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Abstract

Grafting as a cultural practice for controlling soilborne diseases and improving abiotic stress tolerance has been widely used in the production of solanaceous and cucurbits crops in many areas of Asia and Europe. Interest in vegetable grafting has been growing in the United States in recent years. By physically conjoining a plant with desirable fruit characteristics (called a scion) onto another plant with specific disease resistance or stress tolerance (called a rootstock), grafted plants combine the beneficial characteristics of both the rootstock and scion cultivars. The major obstacle of wide adoption of this technique in the United States is the high cost of grafted transplants. The production cost can be partially reduced by increasing efficiency of grafting techniques. Three methods are commonly used in melon grafting, i.e., hole-insertion, splice grafting, and tongue-approach grafting methods. The advantage of hole-insertion method is that it does not need grafting clips, but it has a narrow window regarding relative plant sizes of rootstock and scion. Splice grafting is easier to conduct compared with hole-insertion and tongue-approach methods, and it has less stringent requirement for the growth stage of rootstock and scion. Tongue-approach method may require more greenhouse space, while it often helps achieve a good graft survival rate. Plants grafted with hole-insertion and splice grafting methods require high relative humidity conditions for post-graft healing. Rootstock regrowth (sucker) can be completely eliminated by using tongue-approach method. To facilitate mechanical grafting, as well as long-distance shipping of grafted transplants, root excision at different grafting stages has been practiced. The diverse procedures of melon grafting techniques are presented in the project.

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Adventitious root formation in *Juglans nigra*: a time and place for everything©

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INTRODUCTION

Black walnut (BW; *Juglans nigra*) is an integral component of the Central and Eastern hardwood regions of the United States. Fine hardwood trees such as BW are used to manufacture high-end wood products such as veneer, cabinetry, gunstocks, and furniture, which are traded regionally and globally. Ecologically, BW serves an integral role as a riparian species, as well as providing food and shelter for wildlife. While BW continues to be cultivated commercially, significant effort and resources have been spent selecting for and breeding BW for improved timber characteristics.

As elite BW genotypes were developed it was quickly realized that clonal propagation was difficult. Traditional methods such as grafting requires a high level of skill, is time and labor intensive, has limited rates of success, and the resulting trees are not growing on their own roots, which can have an adverse effect on performance. Softwood cutting propagation is ideal for rapid multiplication of superior genotypes. However, the inability to predictably and reliably produce adventitious roots (AR) remains the greatest impediment to a routine BW propagation protocol. Recalcitrance to AR formation is common in most woody perennials such as BW. Adventitious roots formation is an extremely complex process controlled by many external and genetic stimuli; unfortunately, little is known about the underlying mechanisms controlling AR development.

Past attempts to improve adventitious rootability in BW have had limited success. Such gains were often difficult to reproduce, as rootabilty in BW is highly genotype specific, contributing to the inability to develop an optimized clonal propagation system.

RESEARCH OBJECTIVE

The objective of our research was to improve the frequency of AR formation in BW softwood cuttings, and investigate changes in anatomical patterning during root development. We tested multiple auxin types and concentrations, two environmental conditions during rooting, and two age classes of cuttings (easy-to-root half-sib seedling juvenile material vs. difficult-to-root mature grafted material).

CUTTING PROPAGATION

By using a fog system to maintain an elevated humidity, juvenile BW cuttings were successfully rooted as high as 72.2%. The use of high-density fog to root cuttings is commonplace in many horticulture systems, but has rarely been applied routinely to forest tree species. With this method, rooted cuttings were healthy and had well-developed root systems (Figure 1A, B). After transplanting to soil, cuttings continued to grow normally (Figure 1C). Cuttings rooted under intermittent mist however, often deteriorated as a result of superficial foliar salt accumulation and rooted less frequently than those in fog, independent of the auxin type used for root induction. Control cuttings without exogenous auxin application and cuttings taken from mature selections failed to root, regardless of rooting environment or auxin type and concentration.

After improving the rooting efficiency, we were then able to reliably study anatomical changes during AR formation in BW stems. By observing the timing and location of cellular changes during AR formation we hoped to better understand the underlying mechanisms

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regulating root formation. Stem tissue was collected on sequential days after root induction, and fixed in formaldehyde prior to paraffin embedding, serial sectioning, and staining. Histological analysis revealed that by Day 16, cell files were forming into what appeared to be root initials (Figure 2), and fully formed root primordia were evident by Day 18 (Figure 3). Adventitious roots were thought to originate from parenchyma cells located between gaps in phloem fibers. Location of root primordia formation and duration prior to emergence suggested an indirect pattern of root formation in BW cuttings. These preliminary findings are integral in the improvement of clonal propagation of elite genotypes, and will further allow for an elucidation of the molecular controls of AR formation in BW. Efficient and reliable propagation methods are also powerful tools for tree breeders and for conservation efforts.

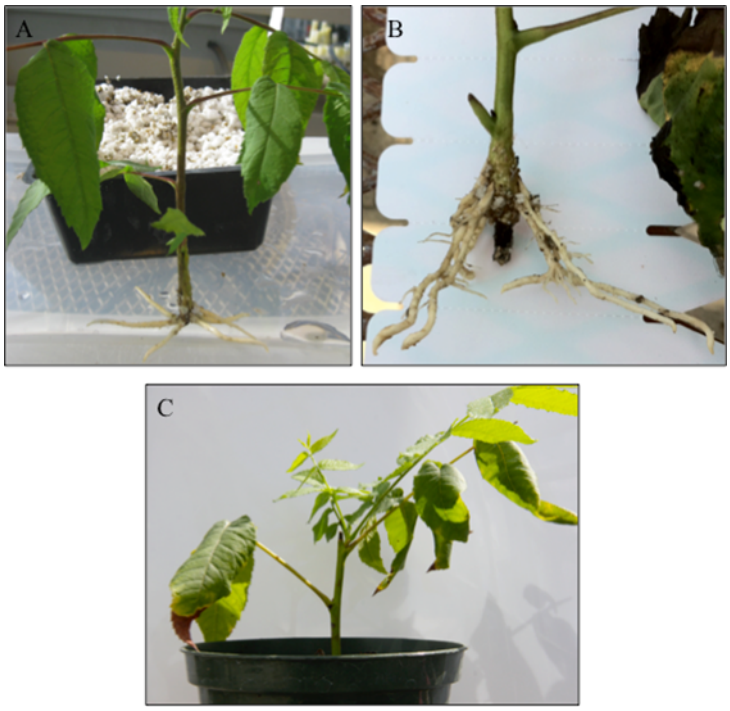

Figure 1. Successfully rooted black walnut (*Juglans nigra*) softwood cuttings. (A) Healthy rooted cutting 5 weeks after adventitious root initiation. (B) Close-up view of healthy root system of a rooted cutting. (C) Acclimatized rooting cutting transplanted in soil and continuing to grow normally.

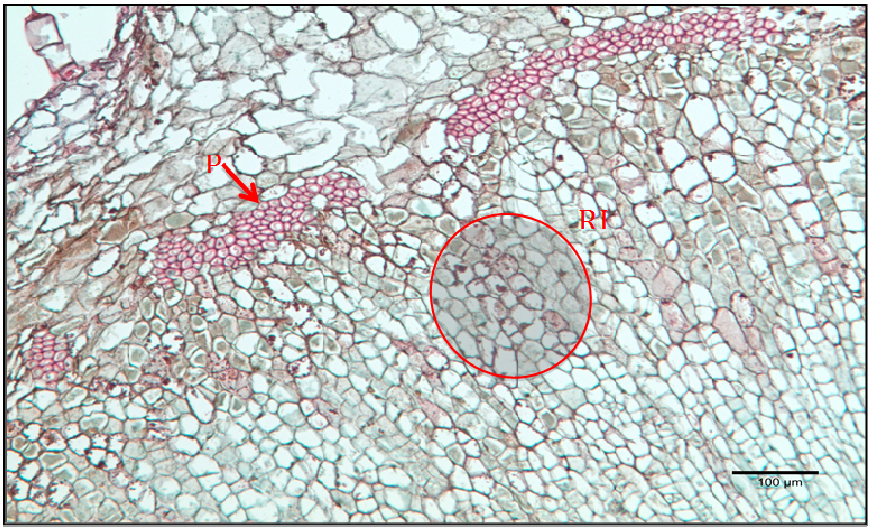

Figure 2. Juvenile cutting transverse section 16 days after auxin application. Organized files of cells forming root initials (RI) first evident in gaps between phloem fibers (PF).

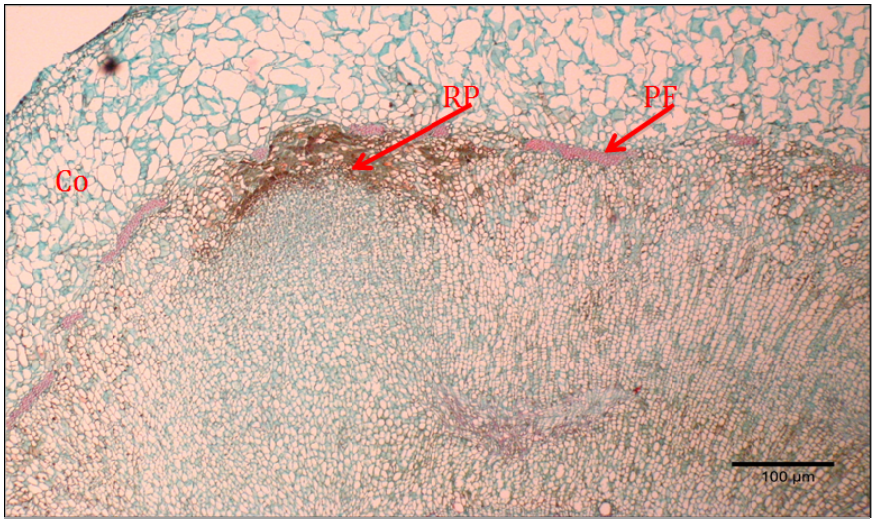

Figure 3. Juvenile cutting transverse section at Day 18. Root primordia (RP) is first visible with fully formed root cap. PF, phloem fibers; Co, cortex.

ACKNOWLEDGMENT

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Grafted watermelon transplants: a new business opportunity©

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Abstract

Grafting vegetable plants onto specific rootstocks which are resistant to soilborne diseases is a unique horticultural technology attracting interest among intensive vegetable crop producers as well as organic growers. In many parts of the world including the USA grafting represents the only feasible measure to control a diversity of problems such as soilborne disease and saline soil conditions. Cucurbit plants, particularly watermelon (*Citrullus lanatus*), are grafted using the one-cotyledon method. The optimal stage of growth for grafting watermelon is the 1 to 2 true-leaf stage for the scion and the 1 true leaf stage for the rootstock. A 9-day healing regimen was found to be successful for watermelon in western Washington conditions and had 90% survival for grafted watermelon transplants. Our current research studies are investigating how to further optimize the success rate for grafting watermelon transplants, such as applying antitranspirants to reduce water loss and utilizing the splice grafting method to eliminate rootstock regrowth. Additionally we are testing grafted plants to control verticillium wilt caused by *Verticillium dahliae* in Washington.

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The importance of USA National Arboretum crapemyrtles in the USA nursery industry©

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Abstract

The U.S. National Arboretum has bred and released crapemyrtles for over thirty years. In that time, Arboretum scientists have introduced genetics which have revolutionized crapemyrtle production in the United States. The greatest contribution to crapemyrtles was probably the introgression of powdery mildew (*Erysiphe lagerstromeae*) resistance via breeding with *Lagerstroemia fauriei*, which was brought to the USA from Japan by Arboretum staff. Almost all powdery mildew resistance in modern crapemyrtles has derived from that plant introduction. Additionally, the Arboretum was the first to develop and release dwarf crapemyrtles in the USA and almost all modern dwarves are derived in some way from Arboretum plants. A recent market review indicates that 30% of all available (from at least one nursery) cultivars of crapemyrtle are either USNA introductions or directly derived from USNA plant material. There are approximately 61 crapemyrtle cultivars that are more popular and more available (more than one nursery lists inventory). USNA introductions and plants directly derived from USNA plant material make up more than 50% (31 plants) of the most popular and available crapemyrtles in the USA nursery industry.

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Qualitative review of plant extract HB-101®©

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INTRODUCTION

Note: This presentation is not an endorsement or advertisement for the product reviewed herein. It is a qualitative product trial, in real world applications. Readers are welcome to use this paper as a guide to determine whether or not this product would be of use in endeavors.

Several years ago, I was visiting a large Japanese Supermarket in the Northwest suburbs of Chicago. In addition to the various groceries and dry goods from Japan and the United States, this store also has a large book store housed within the building. While I was looking over the various gardening and hardscaping books that dealt with the Japanese style of gardening, I also looked through the magazine section. There I found a number of gardening magazines and the subsequent garden themed ads.

I came across a number of fertilizer product ads, many of which I was familiar with. One product though, was new to me, and I investigated it further. I also had a friend who could translate for me, explain the product and its use, according to the advertisement. The product HB-101®, is widely used in Japan, and the subsequent pages in other publications, seemed to indicate it was a popular, or at least a well marketed product. After doing the Google® and Bing® search ritual, I ordered a bottle of this product for myself (via Ebay®).

To be clear, this is not a fertilizer per se, but a plant extract, which can be used as a foliar agent, used alone, or added to liquid fertilizer regime. I have used it as a standalone product and additive to various horticultural production and maintenance programs. I will relay those findings to you now.

MATERIALS AND METHODS

Firstly, this product was applied in the following ways, first as a foliar treatment. Either using a small hole rain wand sprinkler, or with a trigger sprayer. Another method was with a typical watering can, which was tipped over the media in the various containers, and poured into them. Containers where watered until the level of water neared the lip of the container, and then allowed to run into and drain out of the container. Sizes of these containers range from Herkuplast® cell trays, 3½ in. deep to 5 in. deep. Various sizes of Anderson Die and Manufacturing® band pots and nursery containers were also used. Several very large containers were used as well, 25 gal and larger. I also have begun to trial some of the grow bags for smaller specimen trees and shrubs.

As far as container media are concerned, I am currently using several proprietary blended soilless media from Old Castle Lawn and Garden™. These will typically have a starter charge of fertilizer, a wetting agent, and in some cases a slow release product with a release time up to several months. The components are bark, pine and or hardwood, with either peat, rice hulls and/or a compost component included. For long term container growing situations, I will include a “dry” fertilizer into the media via incorporation. The product that I have been using for some now is Nitroform® 38-0-0, small pearls, at a rate of 1 and 1½ lbs yard^3 of medium.

The manufacturer of HB-101, lists various dilution rates on the website, along with suggested application scenarios. Using these tables as a guide. In my trials, I came upon the following rates of HB-101 to incorporate into water. I use either 1.8 cc (mL) to 3.785 L, 2.5 cc to 7.57 L, or 3.75 cc to 9.46 L.

This depends upon the particular crop, time of the season, and the size of the plants. There may be at certain times of the season, an addition of typical water soluble fertilizer. I use several different types of the Jacks® fertilizers. I may also use spray grade ammonium

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sulfate, potassium nitrate, or Plant Marvel® fertilizer.

RESULTS AND DISCUSSIONS

Typically, foliar application and direct soil watering seem to be the most frequent methods used. When treating vegetables, tomatoes, eggplants, peppers, and various lettuce crops, there is a noticeable increase in the "vigor" of leaf growth. In the case of crops other than lettuce, secondary branching occurred, coupled with subsequent increase in flowering. This should result in more fruit bearing.

When treating herbaceous perennials while emerging in spring, the product was applied over the top of emerging leaves. While there was not an appreciable growth spurt. One observation was that during cold evenings, there was less damage to leaves than normally occurs.

In regards to tropical or temperennial plants, there were some different results. Plants observed for this trial were various aroids, philodendrons, monstera, *Alocasia, Colocasia*, also studied was tetrastigma, begonias, bromeliads, and assorted annuals.

There is a correlation to temperature and growth with these plants. The warmer the better. Many of these are in my personal collection and are overwintered either in my basement, or in a minimally heated green house. Temperatures are between 50°F in the night hours, to upwards of the mid 80s during the day, in the greenhouse. The basement temperature is constant at 75°F during the day, and 65°F at night. In April and for part of May, they may be outdoors during the day and be returned indoors at night. Once the night time temperature stays above 50°F, the plants stay outside.

I begin to apply the HB-101 in late April at the lowest rate. In several weeks, or when the temperature increases by 10°F, I begin to move the rate to the next higher concentration. By the end of May I am using the higher rate. I typically apply this product at 12 to 14 day intervals. The tropical plants show great vigor. Indeed, after the application of HB-101, 6 to 7 days later with either *Alocasia* or *Colocasia*, a new leaf or multiple leaves will emerge from the center of the plant. With plant maintenance, as leaves begin to yellow, they are removed, which encourages more growth. I have reduced the amount of liquid soluble fertilizer I have used previously. The amount was typically a table spoon, 14.8 mL, to 1½ tablespoons to 2½ gal., 7.57 L, of water. I now use about 60% less product, when incorporating the HB-101.

Temperate perennials appear to have better color in their leaves, and produce more roots when treated with this product correctly. The roots that are forming are finer roots, or more root hairs, in a container. I applied only HB-101 to several flats of *Asarum canadense*. The rate was the middle dilution, at 20-day intervals for 5 weeks. The ginger was dug dormant, from display beds, in late winter/early spring. The clumps were then transferred into Anderson® deep propagation flats. Whatever soil remained on the roots was left alone. This was done to retain any mycorrhiza present. Soilless medium was used to fill the flats. It is my contention, that this product may very well aide mycorrhizal growth as a secondary benefit. This is something I cannot scientifically prove at this time. I do believe by my visual observations though, the presence of the typical mycorrhizal white growth on the root hair tips with the use of this product. With regards to the leaves, they grew "normally". There were no larger than normal leaves. Twice a month, I would turn over the flats and observe the root and shoot growth. After the first several applications, roots were moving into the new media. They continued to grow at a constant rate through the season. At this time of the year, late September, fall is here in the upper Midwest, the trays are fully rooted. In late February or early March, these flats will be divided up into smaller divisions, which will be transplanted into 5- or 6-in. pots.

I admit these results were not obtained with strict scientific methods. None the less, this product does appear qualitatively at least to be a benefit to a number of different plants. I incorporate it in my garden maintenance business, and in growing plants in containers.

Thank you for your interest in this paper, it has been my pleasure to share this information with you.

The use of light-emitting diode systems for improving plant propagation and production©

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INTRODUCTION

The propagation stage of plant production can be challenging but the quality of the resulting seedling, rooted cutting or young plant is crucial to the performance of the finished crop. In protected horticulture many aspects of the crop environment such as temperature, humidity and irrigation, are carefully controlled to optimise plant performance. However, despite the importance of light to the process, many propagation systems rely on solar radiation which varies through the season and from day to day, resulting in crop variability.

High pressure sodium and other types of high intensity discharge lamps have been used to provide supplemental lighting. While these can improve plant growth they can also result in stretching due to the lack of blue light in their output spectrum. The introduction of light-emitting diode (LED) lighting systems for horticulture caused great interest due to the potential energy savings compared with traditional lighting systems (LEDs use 25% less electricity than 600W HPS lamps for an equivalent light intensity). However, there is a growing body of evidence that suggests LEDs provide many additional benefits beyond simple energy saving that may have a greater impact on crop production, for example by being able to "tailor" the output light wavelength to meet specific crop management requirements.

PLANT LIGHT RESPONSES

To understand why the spectral control provided by LEDs provides an advantage it is first necessary to understand how plants sense and respond to light. Chlorophyll pigments absorb light energy at wavelengths between 400 and 700 nm during photosynthesis but plants also possess an array of other photoreceptors (light sensing proteins) that can detect specific colours of light, using this information to change their morphology in response to the light environment they are exposed to.

In dark or low-light conditions these photoreceptors are inactive and plants stretch (become etiolated) as they attempt to grow towards light. When exposed to light the photoreceptors are activated and drive a process called photomorphogenesis. During photomorphogenesis plant stretching is inhibited, the leaves open, turn green and bend towards the light. In addition to the morphological changes, many aspects of gene expression and biochemistry are altered that help plants acclimate to the light environment. Photomorphogenic processes function throughout the life of plants and help them acclimate to changing light conditions and also control the transition from vegetative growth to reproductive growth.

There are several types of photoreceptors each of which is responsible for a specific set of photomorphogenic responses though some responses, such as stem elongation, are regulated by several photoreceptors working together. The photoreceptors can, in general terms, be grouped by the wavelengths or "colours" of light to which they are sensitive, blue, red/far-red and UVB ("ultra-violet") light.

Blue light photoreceptors include the phototropins and cryptochromes. The phototropins control stomatal opening, phototropism (bending to towards the light), chloroplast movement within cells, leaf flattening, and inhibition of hypocotyl elongation. The cryptochromes are involved in regulating pigment synthesis, the circadian rhythm, flowering, and inhibition of hypocotyl elongation.

Red and far-red light are sensed by a family of photoreceptors known as the

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phytochromes. They function to help plants detect the red:far-red ratio which changes when, for example, taller plants grow above, and cast shade on, shorter ones; and at sunset. They are important for germination, inhibition of hypocotyl elongation, apical hook straightening, leaf expansion, flowering time, regulating circadian rhythms, and chlorophyll biosynthesis.

The UVB light receptor is known as UVR8 and is very sensitive to small amounts of UVB. UVB causes plants to produce more pigmentation and tougher, more robust, leaves (Wargent et al., 2009), and can increase the concentration of essential oils in herbs (Kumari and Agrawal, 2011; Hikosaka et al., 2010).

EXPERIMENTS USING LEDS IN PROPAGATION

With LED lighting it is possible to select the colour and intensity of specific wavelengths of light used for plant production. This means that the light output spectrum can be altered to stimulate the different groups of photoreceptors in the plants in a crop and this allows plant morphology to be manipulated to produce plants that match customer specifications. The light spectrum can also be selected specifically to improve important stages of crop production.

Cuttings are often challenging to propagate as they are prone to dehydration. Selecting a light spectrum to minimise dehydration can improve cutting propagation. In the LED4CROPS experimental facility at Stockbridge Technology Centre, Yorkshire, UK, *Elaeagnus*, *Photinia*, and *Rhododendron* cuttings exposed to 100 µmol m^{-2} s^{-1} of light with different red:blue light mixtures (100% blue; 64% blue:36% red; 35% blue:65% red; 11% blue:89% red; and 100% red) showed a marked decrease in survival as the proportion of blue light in the spectrum increased (Figure 1). This effect was particularly pronounced in the *Elaeagnus* cuttings which shed most of their leaves within 2 weeks of exposure to 64 and 100% blue light mixtures. As blue light is associated with stomatal opening, higher intensities of blue light are thought to be causing cutting dehydration. Cuttings survival was best in light mixtures containing between 11 and 35% blue light.

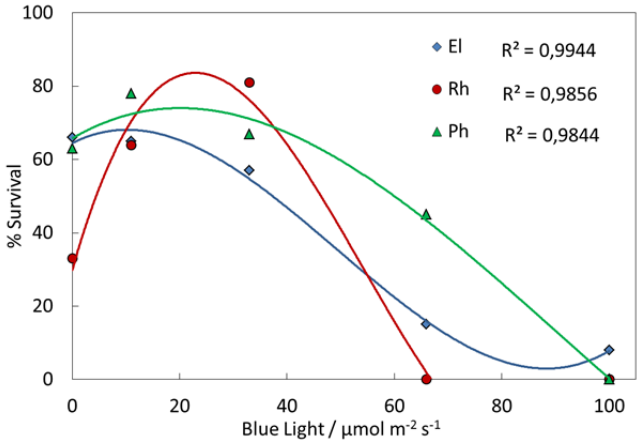

Figure 1. The percentage survival of *Elaeagnus* (El), *Photinia* (Ph), and *Rhododendron* (Rh) cuttings exposed to different mixtures of red and blue light. Lines show polynomial regressions, the R^2 values are shown next the figure legend. Total light intensity was 100 µmol m^{-2} s^{-1}.

As well as affecting cutting survival, light quality also influences rooting. Rooting was improved in grape *Vitis heyneana* subsp. *ficifolia* (syn. *Vitis ficifolia*) when illuminated with red light, compared to or blue light, or to light from fluorescent bulbs (Poudel et al., 2008). When Wu and Lin (2012) propagated *Protea cynarodies* cuttings under red LED light, 67% rooted compared to 7% under conventional fluorescent tubes; while 13% rooted under blue light or a red:blue (1:1) combination.

A second experiment in the LED4CROPS facility examined the influence of far-red light on *Elaeagnus, Photinia*, and *Rhododendron* cutting survival. Far-red light was found to reduce

cutting survival (Figure 2) and again this was most pronounced in the *Elaeagnus* cuttings. Currently we have no biological explanation for this response but it is possible that the far-red light is reducing the synthesis of some hormones that are important for root initiation.

However, even without a full explanation of the biology, information from these early-stage trials can be used to improve the light environment in production facilities, either through the use of LED lighting or spectral filter claddings or screens in glasshouses.

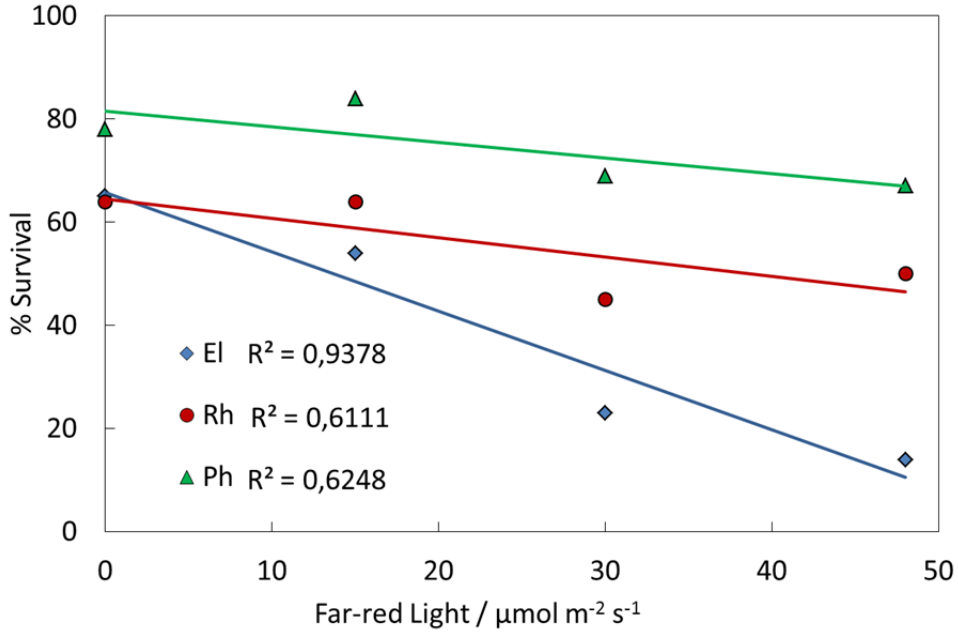

Figure 2. The percentage survival of *Elaeagnus* (El), *Photinia* (Ph), and *Rhododendron* (Rh) cuttings exposed to different amounts of far-red light in a PAR background of 100 µmol m^{-2} s^{-1} (11% blue:89% red). Lines show a linear regressions, the R^2 values are shown next to the figure legend.

LEDS TO CONTROL CROP MORPHOLOGY

Light environments can also be designed to control the morphology of crops during the vegetative stages of growth and to induce flowering. Both red and blue light are required to control plant morphology. In general, for plants grown under red:blue light mixtures with intensities of 200 µmol m^{-2} s^{-1}, compactness increases as the proportion of blue light increases in the mixture from 0 to 60% blue (Figure 3). If the blue percentage is increased beyond this, plants become increasingly etiolated. Careful selection of the light mixture for a crop species or cultivar can enable rapid growth and controlled morphology. The correct light spectrum may control morphology sufficiently to remove the need for chemical plant growth regulators.

While plants can be kept compact their rate of development may also be delayed if the spectrum is not optimised, resulting in delayed flowering. For bedding plants where advanced flowering is required prior to sale, far-red light may be used to induce flowering (Figure 3B). It is, however, necessary to add only just enough far-red light to induce flowering as too much will cause stretching and make it impossible to produce the compact plants required for the market.

A considerable amount of research into the uses of LEDs in different aspects of crop production is currently underway round the world. There are many examples of the use of spectral manipulation for improving propagation efficiency, crop morphology, pigmentation, flavor and aroma. Taken together these benefits have the potential to have a far greater impact on horticulture than the energy saving provided by LEDs.

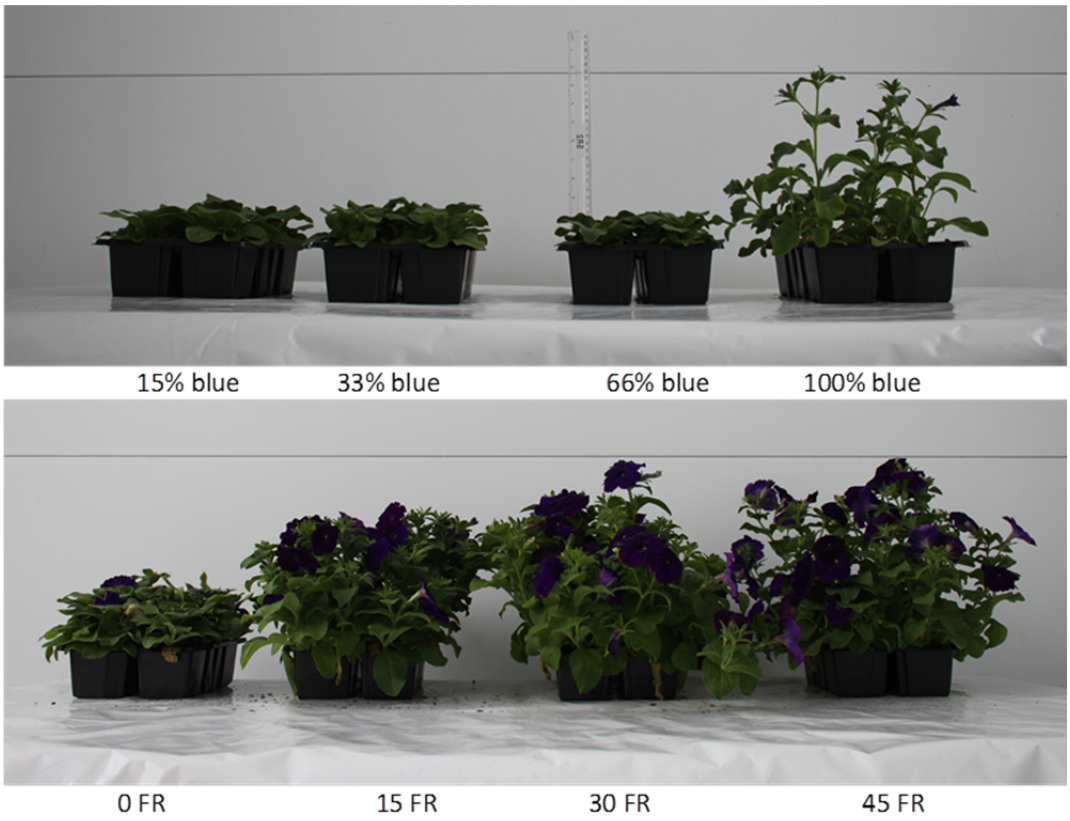

Figure 3. The influence of blue light percentage (top image) and far-red light intensity (bottom image) on the morphology and flowering of petunia plants when grown under PAR intensity of 200 μmol m^{-2} s^{-1}.

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Methods to simplify nursery operations and dispatch at Gunnar Christensen's Planteskole©

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INTRODUCTION

Gunnar Christensen's nursery has been in operation for 50 years. It was started on bare field site in 1962 by Gunnar and Nina Christensen and is now run by Henrik and Lotte, the second generation of the family. The nursery covers 17 ha, including 1.5 ha of greenhouses.

The business specialises in production for garden centres in Denmark and Sweden. Annual production is currently more than 1 million plants, including ornamental shrubs, perennials, herbs, fruit trees and soft-fruit plants, and other edible plants such as asparagus and rhubarb. Sizes range from small perennials in 11-cm pots to large specimen shrubs in 25 L pots.

An integrated approach, based on the use of biological control, is taken to crop protection and plant health. Plants are grown "hard" — in other words, fertilisers rates are used that are no higher than those proven in trials to be of direct benefit to the plant and irrigation is regulated to provide a mild drought stress both in the greenhouse and in field production. These approaches combine to produce robust, compact plants that will tolerate conditions during transport and at the garden centre.

The nursery employs up to 50 people in peak season, from April to June, reducing to just two in January. The workforce typically consists of about 20 regular Danish employees plus three or four Danish students; seasonally there will also be eight Ukrainian horticulture students and about 18 Polish workers, some of whom have been working at the nursery for more than seven seasons.

Management on the nursery aims to make complex or large tasks or operations simple and easy to do by breaking them down into manageable jobs. One reason for this is the number of different languages used by members of the workforce at different times through the year. The nursery's working language is English but even so, "English" can mean different things to different people. It can be difficult to explain Danish nuances in English so they can be understood in the English the Polish workers use. That is why is it important to have precise individual instructions and why simple single operations and deliberate planning is important.

METHODS FOR SIMPLIFYING OPERATIONS

The nursery uses Excel® spreadsheets to produce work schedules based on the combined knowledge and experience of all members of the team in the various areas of work. The format makes it easy for anyone to record their knowledge about actual tasks.

In propagation, for example, Excel records are kept of the different types of cuttings used, rooting hormones, biocides, fungicides, and any pest or disease problems. When a member of the team goes to harvest cuttings, they can use this knowledge base, for example to see exactly what time of the season to take the cuttings for best results, how the cuttings should be made, and how many of them to harvest for any given quantity of rooted cuttings required, and if there are special requirements with a specific crop.

The records also provide information on tray size, rooting medium mix, rooting hormone, biocontrols, or fungicides that may be required.

The beauty of the system is that it is continually being updated in the light of experience.

The Excel file can be used to print a label containing all the information we need for a

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particular crop of cuttings so that it can be seen by everyone working with that crop. Having this information readily accessible also makes it easy and simple when it comes to training new employees.

Production planning

The Excel information can be used for planning, management, and printing work cards and labels.

In the autumn and winter, for example, the nursery typically buys-in young plants for the coming season's production. The Excel planning sheet will include details of the taxa required and the numbers of each, from which a purchase order is derived and against which we can check deliveries. As the Excel data includes information based on the accumulated knowledge of production requirements and schedules for each crop — taking account of factors such as the potting machine capacity — it also enables delivery dates to be set.

From the Excel sheets estimated labour requirements can also be planned ahead before the start of the busy season, based on:

- Planned production quantities
- Plant handling and transport
- Propagation and production schedules
- Crop care requirements (nutrition, pest and disease control etc)
- Site maintenance

The result gives an overview of the expected need for staff week by week. The knowledge base is also used to guide where on the nursery each crop is to be placed, based on area required, irrigation and fertiliser requirements, dispatch logistics and care in general such as pests and fungal diseases.

From the Excel files we can print work notes covering, for example, potting, pruning, staking, and so-on. It is also possible to print a label that pulls information from the Excel sheet showing a crop's nursery location, its origin (as a cutting our bought-in young plant) young plant size, etc. All of this information is important in terms of managing the work being done to the plants.

For example, for each crop a potting work card will be printed from the production plan to inform the potting team. This shows young plant supplier, young plant size, potting depth, soil mix, plants per pot, pot size and field location.

In other words, all the information you need when you do the potting.

Overwintering logistics

Winters are cold in Denmark so it is essential to plan in advance how crops are protected over winter. The nursery has facilities to create six "climate zones" available for overwintering. including outdoors. The work is planned in August and September and is undertaken from October to December. Again all the experience accumulated over many years is collected in the Excel knowledge base and is used to plan over-wintering each year. Even for crops that can be left outside, it is important to know which need to be pruned or tethered and where to stand them for the winter.

The Excel sheets are used to plan labour requirements, space consumption and record this season's experience for use in the future. The knowledge base ensures the nursery's managers know that the labour is ready when it is needed, and the space is available in the right environment for each crop. It can also take account of where empty space will be required in the greenhouses for early production the following year.

Work management

The Excel-based system gives the nursery's managers an overview of the situation, and a plan enabling them to maintain control even during the busiest times of the season. Planning ahead means the work involved in sales, potting, propagation, keeping up to date with legislation, and dealing with customers is given full attention and so is less of a worry. When dealing with living plants and unpredictable weather or unpredictable customers

planning helps the business to react more efficiently. When 80% of the plans succeed, the remaining unpredictable 20% can be dealt with more easily so that in the quieter parts of the year it is possible to take well-considered and debated decisions so that the plan is ready when the busy times begin.

Being able to record the current season's experiences in a single place makes it possible to plan for things to run even more smoothly the following year.

The system ensures the business can make a deliberate plan that makes the best use of everyone's accumulated experience. That in turn helps to make all of the workers feel part of the team when they know their knowledge is being used in this way.

It is unfortunately necessary to have meetings and at Gunnar Christensens Planteskole these are kept as short as possible, with everyone is standing. A meeting is held every day at a fixed time late in the day, where the team leaders will present the next day's work programme in each department. Priorities for labour needs between departments are discussed. This helps to ensure an understanding of each department's challenges and needs and gives each team leader the opportunity to prepare their own department's work In good time to put their staff into individual tasks, so they also know the day's programme before they begin.

This process enables everyone to influence and understand the impact of their own work, leading to greater job satisfaction. The nursery is proud of its absentee record –in Denmark, the average is seven sick days per year for workers in the private sector, at Gunnar Christensens it is less than two.

IMPLEMENTATION OF A NEW DISPATCH SYSTEM

The nursery produces more than one million plants in eight different pot sizes, amounting to 1300 different product lines spread over 17 ha of production area. Its original dispatch system assembled plants together for despatch to a customer by gathering them on the Danish trolleys, two trolleys at a time, on the back of a tractor. An employee brought in one customer's order at a time, from the whole area and all employees in the dispatch area were required to choose the right plants from 1,300 taxa, with optimum growth, flower or bud. At the same time the plants were trimmed, labelled and generally prepared for despatch to each garden centre. Under this system it was hard for the individual to be good at the job and it took several years for a new employee to learn, because of the many different plants.

In the new system, which was put into operation from the beginning of the 2015 season, the plants are collected onto trucks. There are four mobile tables on each vehicle so each employee can pick plants for up to four customers at a time. Each nursery worker will now only collect plants in from a small area of the nursery representing 200 or 300 types of plant. When the plants come into the packing shed they are sorted into orders for each customer on roller conveyors to bring all the plants for a customer together in one line where they are prepared, trimmed, labelled and generally quality controlled before despatch to the garden centre.

This is easier and simpler for the workers because:

- Each member of nursery staff has far fewer plant types to become familiar with, which minimizes uncertainty when selecting the plants;
- If a mistake occurs, and it does, it is much easier to find out who is lacking information on what the customer expects for that type of plant;
- There are people in the packing hall who are skilled in making the plants ready for dispatch, and who are trained in watering and packing plants on the Danish trolleys.

Dividing the dispatch process into a series of simple tasks makes training easy and simple which means the nursery can use its unskilled foreign workers from their first day.

FUTURE WORK MANAGEMENT CHALLENGES

In Denmark, as in many other parts of Europe and indeed most IPPS Regions, fewer young people are seeking entry to the horticulture industry. It is increasingly important that those who do enter the industry can be trained not just in growing skills but as supervisors

and managers.

A greater proportion of the staff on Danish nurseries will be untrained seasonal workers, from Eastern Europe. This means it will be more important than ever to make the work simple, uncluttered, and with measurable targets, that are well planned in good time, before the busy season begins.

To achieve that our team leaders must be well prepared both for those immediate goals and those with slightly longer lead-in time.

Performance of plant protection products against *Thielaviopsis* on *Viola*©

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INTRODUCTION

Thielaviopsis basicola infects members of at least 15 plant families to cause black root rot resulting in uneven growth of seedlings and failure of establishment in newly potted nursery stock. There is usually a slow decline in plant vigour, until the plants are put under stress, for example in warm weather or drought. The roots develop dark brown speckled areas where long-lived resting spores (chlamydospores) are formed in the pale-coloured host cells. The fungus also produces abundant colourless endospores which are released outside the root and can be spread in run-off water.

In 2013, UK growers of pot and bedding plants and nursery stock recently became concerned about black root rot and the limited number of plant protection products that are available to them. In particular, Cercobin WG (thiophanate-methyl), can be used as a drench over ornamental plants, but only once per crop, and only if they are in containers in a permanent greenhouse. The UK levy-funded research body, AHDB Horticulture, agreed to fund a series of studies to find alternative treatments. An initial scoping study (Wedgwood, 2013) determined that other chemical active ingredients and biological products might be effective against the pathogen. This led to efficacy experiments (Wedgwood, 2014, 2015).

METHODS

Fungicide efficacy experiments with *Viola cornuta*

In total, 13 products, including conventional synthetic chemical products and biological or other non-conventional products (Table 1) were compared at the same time in two separate glasshouse experiments, with *Viola cornuta* sown on 9 May 2014. All products were used preventatively, a week before inoculation. In addition, in another set of plots, all except Cercobin WG, T34 Biocontrol and Trianum-G were applied again a week after inoculation.

Both chlamydospores and endospores of *T. basicola* were dispensed in a suspension over the top of the growing medium in module trays 4 weeks after sowing. Both untreated uninoculated and untreated inoculated plants 'control' plots were also set up.

Trianum-G granules were mixed into the peat-growing medium before tray filling and T34 Biocontrol was applied in liquid suspension to trays straight after sowing. All other applications were of liquids to the seedling trays at two-leaf stage and were made using an automatic pot sprayer. Products approved for foliar spray application, not as a growing-medium drench, were, for the experiment, given more water straight after application to achieve 1000 L of water ha^{-1}.

More *V. cornuta* were sown on 17 July 2014 to test the selected products in simple treatment programmes (Table 2), with *T. basicola* inoculation 4 weeks after sowing.

Following the analysis of the *V. cornuta* experiments, products were selected for testing on *Choisya* sp. in Experiment 4. The experiment was started on a nursery on 30 April 2015 with root rot due to be assessed in November 2015, and so no further details are given here.

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Table 1. Products tested on *Viola cornuta* in Experiments 1 (conventional plant protection products) and Experiment 2 (non-conventional) from May to July 2014 at ADAS Boxworth.

Product or experimental code	Active ingredient	Product approval status in UK (as at October 2015)
	Experiment 1	
Cercobin WG	Thiophanate-methyl	EAMU[1] for glasshouse use on container ornamentals
Signum	Boscalid + pyraclostrobin	EAMU for ornamental plant production
Switch	Cyprodinil + fludioxonil	Approved ornamental plant production
F173	Confidential	Experimental product
F174	Confidential	Only approved on other crops
F175	Confidential	Experimental product
F176	Confidential	Only approved on other crops
	Experiment 2	
Cercobin WG	Thiophanate-methyl	EAMU for glasshouse use on container ornamentals
Prestop	*Gliocladium catenulatum* J1446	Approved on protected ornamentals
Serenade ASO	*Bacillus subtilis* QST 713	EAMU for ornamental plant production
T34 Biocontrol	*Trichoderma asperellum* T34	EAMU for protected + container grown ornamentals
Trianum-G	*Trichoderma harzianum* T-22	Approved on protected ornamentals
HortiPhyte	Potassium phosphite	Plant nutrient
F178	Confidential	Not approved on ornamentals
F179	Confidential	Not approved

[1]Extensions of Authorisation for Minor Use (EAMUs).

Table 2. Programmes of one or two products applied at different timings for control of black root rot in Experiment 3. All except T1 were inoculated with *T. basicola* 4 weeks after sowing.

Timing	Treatment programme											
	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10	T11	T12
Wk 0									--------*T34 Biocontrol*-------			
Wk 3			----*Cercobin WG*----			*F174*	*F175*	*F178*		*F174*	*F175*	*F178*
Wk 5				*F174*	*F175*							

RESULTS

None of the products tested caused any visible phytotoxicity to *V. cornuta.*

In Experiment 1, compared with the 36% root rot severity in inoculated and untreated *V. cornuta* plants, seven treatments gave highly significantly ($P<0.001$) less root damage, with a mean 14% area affected (Figure 1). After use of the experimental products F174, F175 and F176 as either preventative alone or with curative application then root damage was similar to that of uninoculated plants (9.7%). Signum, applied preventatively also resulted in significantly less severe root rot than untreated inoculated plants, but with 20% damage.

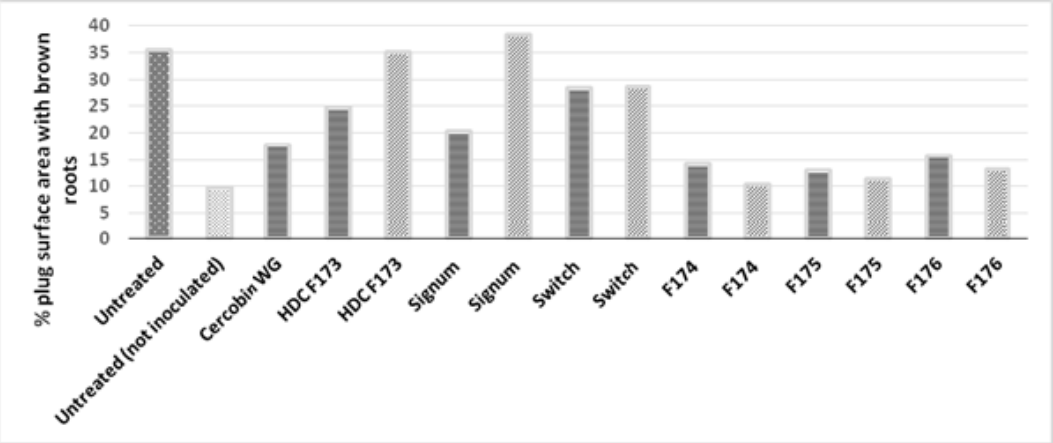

Figure 1. Experiment 1; conventional products. Mean percentage root area brown on the surface of *Viola cornuta* plugs on 11 July 2014 nine weeks after sowing (P<0.001, L.s.d. 13.964). All treatments with below 24% root browning differ significantly from the untreated inoculated. Key: Horizontal lines = product application before inoculation (preventative); Diagonal lines = product applications before and after inoculation (preventative + curative).

In Experiment 2, with non-conventional treatments to *V. cornuta*, root rot was significantly (P<0.001) less severe than for the untreated plants following the use of F178 preventatively plus curatively, with 6.3% root rot (Figure 2). There were no other significant differences in rot severity.

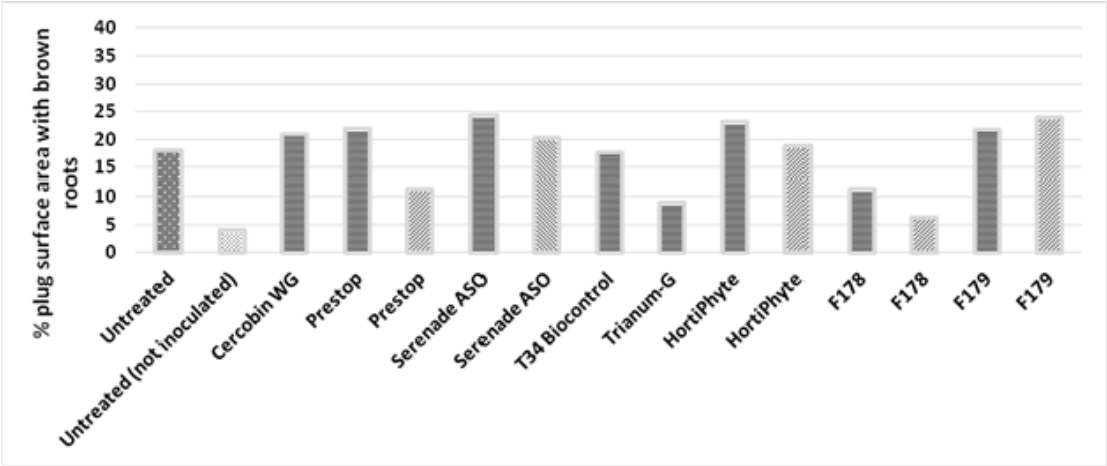

Figure 2. Experiment 2; non-conventional products. Mean percent root area brown on the surface of *Viola cornuta* plugs on 10 and 14 July 2014 9 weeks after sowing (P<0.05, L.S.D. 5.84). Only treatments with 6.3% or less root browning (only HDC F178 applied twice) differ significantly from the untreated inoculated. Key: Horizontal lines = product application before inoculation (preventative); Diagonal lines = product applications before and after inoculation (preventative + curative).

In Experiment 3, testing simple programmes on *V. cornuta*, two preventative treatments were significantly (P<0.001) less rotted (Figure 3), one with F174 (Programme T10) having 40% rot, the other with F175 (Programme T11) with 46% rot, both having received T34 Biocontrol at sowing. Use of any of these three products alone did not reduce root rotting.

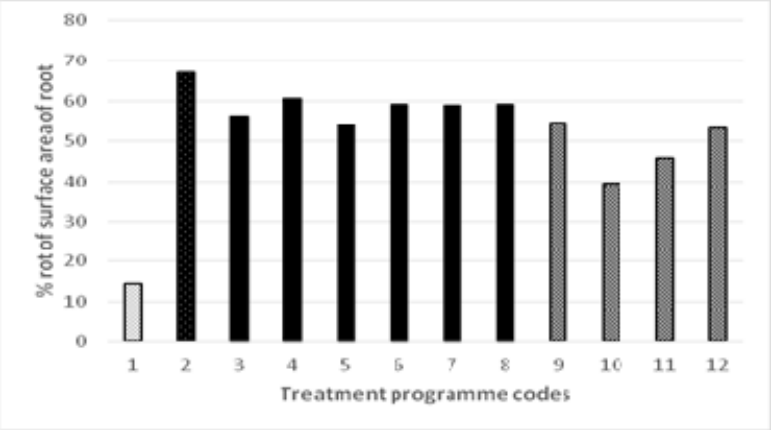

Figure 3. Experiment 3; Treatment programmes on *Viola cornuta* plugs (see Table 2 for details). The percentage of the root area showing rot on 24/25 September 2014 10 weeks after sowing.

CONCLUSIONS

Three out of the four conventional chemical plant protection products tested (codes F174, F175 and F176, not currently approved for use on ornamentals) reduced the severity of black root rot on *V. cornuta* whether applied preventatively alone or followed by curative application. Control was equivalent to that given by Cercobin WG.

The non-conventional chemical product code F178 applied both preventatively and curatively reduced black root rot severity on *V. cornuta*. No significant root rot reduction was shown from the microbial products tested, although both Prestop applied preventatively and curatively as a drench, and compost incorporated Trianum-G gave some reduction.

When short programmes were tested on *V. cornuta* the chemicals coded F174 and F175 did not reduce root rot severity when applied either preventatively, or curatively following Cercobin WG application. However, they were effective when used preventatively preceded by T34 Biocontrol at sowing.

The most promising products will be assessed by AHDB Horticulture for possible applications for EAMU authorisations to enable growers to use them.

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Technical sessions, Monday morning, 12 October 2015©

M. van der Giessen[a]

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OPENING PRESENTATION

The 40th Annual Meeting of the International Plant Propagators' Society-Southern Region of North America convened at 7:30 am at the Sheraton Tampa Riverwalk Hotel, Tampa, Florida with President Maarten van der Giessen presiding.

PRESIDENT MAARTEN VAN DER GIESSEN

President van der Giessen welcomed everyone to Tampa, Florida for the 40th Annual Meeting of the International Plant Propagators' Society-Southern Region of North America. He thanked Local Site Committee Chair, Shawn Steed and his committee and volunteers for the long hours in arranging the excellent tours, hotel, other planning activities and all their attention to detail. He welcomed IPPS International Board members, students, first time attendees, and new members, asking them to stand and be recognized. van der Giessen thanked the Executive Committee, and Kevin Gantt's Sponsorship Committee, which raised $30,000 in cash sponsorships and $2500 in-kind sponsorship; this was outstanding for the challenging economic times. van der Giessen encouraged the membership to visit and show their support of our sponsors during the meeting. He encouraged all members to make new members and first-time attendees feel welcome — share with them and seek from them. He pushed for good questions and enthusiastic participation at the Tuesday night question box.

van der Giessen announced that this is the 3rd year our region has participated with European Region (former Region of Great Britain & Ireland) in the *Young Propagator Exchange* program between the two regions. He recognized Ben Gregory from Great Britain, who was on the International IPPS tour of the Southern Region USA, led by International President, Dr. Patricia Knight. Adam Blalock of the Southern Region of North America was our designee to European Region. Both of these young professionals had an incredible exchange experience in our respective regions. This is the fourth year we are doing the *Vivian Munday Young Horticultural Professional Scholarship Work Program (former Vivian Munday Scholarship).* We currently have a five young professionals (Amny Rose, Judson Lecompte, Jeremiah DeVore, Connor Ryan and Drew Payton) who are making a strong contribution to this year's program. van der Giessen thanked Program Chair and 1st Vice-President, Laura Miller, for the excellent program and slate of speakers she assembled.

PROGRAM CHAIR LAURA MILLER

Program Chair Laura Miller welcomed all members, guests, and students. She thanked the membership for the opportunity to serve them, and then reviewed the scheduled program. The Question Box, scheduled for Tuesday evening, was to be co-chaired by Kevin Gantt and Tom Yeager. She then introduced the first moderator, Benjamin Berry.

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Hydrangea culture at Stephen F. Austin Gardens©

D. Creech[a]

SFA Gardens, Arthur Temple College of Forestry and Agriculture, PO Box 13000, Stephen F. Austin State University, Nacogdoches, Texas 75962, USA.

INTRODUCTION

There are approximately 23 species of *Hydrangea* but only five are widely grown in the USA. *Hydrangea macrophylla*, *H. quercifolia* and to a lesser extent, *H. paniculata* are well adapted to East Texas. While *H. arborescens*, *H. aspera,* and *H. petiolaris* survive, but are not well adapted. The hydrangea collection at Stephen F. Austin (SFA) Gardens dates back to the first Arboretum plantings in 1986, but only with the construction of the Ruby Mize Azalea Garden (1997) and Gayla Mize Garden (2011) did the collection expand to its present size. The most current inventory for the hydrangea collection can be found on the SFA Gardens website in three theme garden webpages (http://sfagardens.sfasu.edu): (1) Mast Arboretum (46 cultivars), (2) Ruby Mize Azalea Garden (232 cultivars), and (3) Gayla Mize Garden (39 cultivars). While there is some duplication of cultivars and the most recent plantings have yet to be added to the website database, the collection remains the most extensive in the southern USA.

SFA GARDENS

SFA Gardens comprises 128 acre (58 ha) of on-campus property at Stephen F. Austin State University, Nacogdoches, Texas. Tree, shrub, and herbaceous perennial evaluation at SFA Gardens is scattered across gardens and landscapes. Nacogdoches is in Zone 8b with an average annual rainfall of 1219 mm (48 in.). June through August is characteristically hot and dry. In recorded history, 1 Sept. 2000 was the record high, 44.4°C (112°F), and 23 Dec. 1989 was the record low -17.8°C (0°F). Soils are generally well drained, slightly acidic, and the native flora is dominated by pine, oak, river birch, sweetgum, sycamore, Florida maple, hornbeam, elm, hackberry, pecan, and hickory.

EXPLOSION OF NEW TAXA

Since Michael Dirr's hydrangea book, *Hydrangeas for American Gardens*, was released in 2004 (Dirr, 2004), there has been a virtual flood of new cultivars entering the market place, many patented and trademarked to one brand or another. In 2012, Dirr reported more extensive hydrangea breeding and advancement, and subsequent increase of cultivar releases (Dirr, 2012). The current flood of new plant materials is indeed bewildering. There is little doubt that the industry is moving increasingly to branded products (Scullin, 2014). This trend will continue. In early 2015, there are over 91 new cultivars of lacecap and mophead hydrangeas that tout reblooming as an attribute, and most cultivars are associated with major brands. Brands include: Endless Summer® (Bailey), Forever & Ever™, Edgy™, Everlasting ™ (Plants Nouveau), Mystical™, Hovaria® (Kaleidoscope®), Japanese Lady Series (Halo™, Frau™, and Angel™), Let's Dance™ and Cityline™ (Spring Meadow), Next Generation™ (Ball Ornamentals), and Showstopper Hydrangeas™, a series promoted by HGTV.

HYDRANGEA TAXA

Lacecap and mophead hydrangea evaluation at SFA Gardens

We have been planting *Hydrangea* at SFA Gardens since 1985 when there were very few cultivars available in the Texas market. In 1997, we initiated side by side trials at the Ruby Mize Azalea Garden. By 2005, we had accumulated over 250 cultivars. With our usual enthusiasm, students measured plant height and width, number of blooms, size of blooms

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and date of the bloom show. Over several years we have used groups of three visitors to rank the top picks. Those making the list of favorite lacecap and mophead hydrangeas included 'Amethyst', 'All Summer Beauty', 'Nikko Blue', 'Uzu Ajasai', 'Ayesha', 'David Ramsey', 'Penny Mac', 'Bailmer' (Endless Summer® hydrangea), Lady in Red', 'Dooley', 'Beauté Vendemôise', 'Souvenir du President Paul Doumer', 'Goliath', 'Badgers Choice', 'Red Ace', 'Kardinal Red', 'Tokyo Delight', 'Fuji Waterfall', 'Bluebird', 'Gori Otakga', 'Nachtigall' (syn. 'Nightingale'), 'Möwe', 'Blaumeise', 'Mousmee', 'Jogasaki', 'Taube', 'Lanarth White', and 'Peace'. In the variegated foliage arena, 'Maculata' (syn. 'Variegata'), 'Lemon Zest', and 'White Wave' have been given high marks, although the latter tends to burn in our high heat summers. With the recent transfusion of new cultivars, mostly branded product, we will repeat the process. Another focus over the years has been to maintain types that are rarely encountered, interesting wild collected specimens, and selections perhaps discarded from other programs but having merit.

Oakleaf hydrangea

Hydrangea quercifolia, oakleaf hydrangea, is a southern USA native that has performed well at the Pineywoods Native Plant Center. Drainage is critical. Dirr lists 27 cultivars, many of which are no longer available in the trade (Dirr, 2009). 'Lowrey' was selected by Lynn Lowrey years ago near Angola Prison in Louisiana. At the Pineywoods Native Plant Center, we have planted many seedlings from our original clone and they have naturalized along the banks of a small stream named Sara's Branch. Our collection includes: 'Brido', Snowflake™ oakleaf hydrangea; 'Sike's Dwarf'; 'Brihon', Little Honey™ oakleaf hydrangea; 'Alice', 'Alison'; 'Flemygea', Snow Queen™ oakleaf hydrangea; 'Ruby Slippers'; and 'Munchkin'. We will be planting 'Turkey Heaven', a selection by Hayes Jackson.

Peegee hydrangeas

Hydrangea paniculata, peegee hydrangea, performs well at SFA Gardens in full morning sun and needs good drainage. Dirr lists 34 cultivars (Dirr, 2009). The current inventory of SFA Gardens can be found on our website. There are over 60 cultivars available and we are on a mission to plant as many of these we can acquire. Images taken of northern grown plants suggest flower colors from dark pink to almost red, a trait we doubt we can duplicate here in the heat of the South.

Lesser known species

SFA Gardens has a number of specimens of *Schizophragma hydrangeoides*, which, if given enough time, can climb to the tops of our tall pine canopy. Flowering is superior with plants exposed to good morning sun. The species always elicits favorable comments. *Deucamaria barbara* is an underutilized durable evergreen native vine that has performed well for many years. With three genotypes represented, including one cultivar named 'Barbara Ann', we continue to promote the species as one of our top vines for the South. *Dichroa fibrifuga* is an evergreen small shrub with blue flowers and fruit - a species I thought would be more successful in the trade than to date. 'Yamaguchi Select' has paler flowers, almost lavender, and opportunities for further selection exist.

CULTURE

Good soil drainage is critical to hydrangeas in East Texas. Heavy rains and waterlogged soils often kill plants. Irrigation systems are required with our hot, dry summers. At SFA Gardens we utilize either sprinkler or drip irrigation. For plants utilizing sprinkler irrigation, the strategy is to apply water when plants show heavy wilt in the morning. For drip irrigation, the strategy is to apply ½ to 1 gal per day to plants utilizing a single emitter placed as close to the crown of the plant as possible. Oakleaf hydrangeas are particularly susceptible to poor drainage. At the Pineywoods Native Plant Center, we have observed that plants thrive along the edge of Sara's Branch, a stream that dissects the front of the property. In fact, it is here that we have found chance seedlings surviving into mature specimens, often germinating just above the water line at the base of the steep sloped banks. We believe that

this is the first example of oakleaf hydrangeas “naturalizing’ in East Texas.

CONCLUSIONS

Hydrangea remains a major commodity across a wide swath of the USA. In the South it is the standard, ornamental plant which reflects the culture and nature of southern gardening. The popularity of the genus is reflected in the number of books and articles written about the species. The surge of trademarks and brands has led to introductions of old cultivars under new names, an issue well covered by Tony Avent’s online article (Avent, 2007). In 2015, the industry is now ripe with an explosion of new cultivars, many extolling reblooming as a characteristic. While many gardeners have long noticed reblooming, particularly for cultivars cut back after bloom, the surge of new cultivars touting the trait is really only a decade old. The verdict on which cultivars are truly remontant has yet to be determined.

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Growth performance of container-grown flowering dogwoods with different shade intensity and color©[a]

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INTRODUCTION

Flowering dogwood (*Cornus florida*) is considered an aristocrat of native flowering trees of the USA and has a broad range extending through most of the eastern states and westerly through Iowa and south to Texas (Dirr, 2009). This species is one of the most beautiful and important small flowering trees utilized in the nursery and landscape industry. A multitude of species and cultivars of dogwood have been a staple in nursery cultivation. Today, the demand for container-grown dogwoods has increased as the demand for containerized trees has continued to grow over the last 20 years. However, dogwoods are a challenging crop to produce in container culture, especially when bare root liners are used as the initial transplant into containers; unacceptable levels of mortality and poor growth occur. Reasons for poor dogwood growth during the first growing season are anecdotally related to overwatering, underwatering, over fertilizing, poor root structure, environmental stress, or transplanting delay from the bare root harvest. Flowering dogwoods are considered an understory tree. Producers are successfully growing other native understory species under shade cloth (Phillips et al., 1991), but most producers continue to grow container-grown dogwoods in full sun.

Studies have shown that temperatures in black plastic containers can exceed 43.3°C (110°F) in full sun (Johnson and Ingram, 1984). Shade treatments of 40% black or white shade cloth were used to reduce root zone temperatures after transplanting dogwoods into containers and resulted in larger plants compared to plants grown in full sun (Montague et al., 1992). The objective of this research was to evaluate shade intensity and shade color on the growth of two cultivars of bare root dogwood liners after transplanting into nursery containers.

MATERIALS AND METHODS

Cornus florida L. 'Cherokee Princess' and *C. florida* 'Comco No.1', Cherokee Brave™ flowering dogwood PP 10166, bareroot flowering dogwood liners were obtained from a commercial nursery in Winchester, Tennessee. The size of the dogwood liners ranged from 41-61 cm (18-24 in.). Liners were potted into a #5 nursery container (Classic 1600, Nursery Supplies, Chambersburg, Pennsylvania) with pine bark substrate amended with 3.3 kg m^{-3} (5.6 lbs yd^{-3}) 19-5-9 (19N-2.2P-7.5K) Osmocote Pro 12 to 14 month controlled release fertilizer (Everris, Dublin, Ohio), 0.7 kg m^{-3} (1.2 lbs yd^{-3}.) Micromax (Everris, Dublin, Ohio) and 0.6 kg m^{-3} (1 lbs yd^{-3}) of AquaGro (Aquatrols, Paulsboro, New Jersey). Before plants were moved into their respective shade treatments, height and trunk diameter measured at 15 cm (6 in.) were recorded and used to grade plants into replications for small, medium, and large size. On 25 February, plants were moved onto a gravel pad in full sun or into one of three shade treatment structures [2.4×3.0 m in size (8×10 ft)]: a 50% black, 50% white, or 30% black shade cloth (Dewitt, Sikeston, Missouri). Each treatment was replicated four times and contained eight plants of each cultivar at an outdoor facility at the Nursery Research Center in McMinnville, Tennessee.

Cyclic irrigation was applied twice daily in early spring and increased to three applications during periods of increased heat throughout the summer. Water was applied using a 160° Spot-Spitter fan emitter (Roberts Irrigation Company, Inc., San Marcos, California). Leachate was collected bi-weekly from two plants of each cultuvar, among the

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four treatments (full sun, black 30%, black 50%, and white 50% shade). Electrical conductivity and pH (data not shown) were determined using a Myron L Agri-Meter (Myron L Company, Carlsbad, California) immediately after leachate samples were collected. The remaining leachate was then stored at 5.5°C (42°F) for further spectrophotometer analysis of nitrate nitrogen and orthophosphate.

Two plants from each replication were harvested on 7 July 2014, and two additional plants were harvested on 13 August 2014. Height, trunk diameter, leaf area, and internode length were recorded for each plant. Plants were severed at the substrate level, all tissue bagged and dried at 57°C (135°F) for 10 days to obtain shoot dry weight. The substrate was gently removed from the roots with compressed air and all roots were dried as described above to obtain dry weight.

The experimental design was a randomized block design with four replications of eight plants per cultivar per experimental unit. All data were subjected to analysis of variance with the GLM procedure of SAS (SAS for Windows Version 9.1, SAS Institute, Cary, North Carolina) and differences among treatments were separated by a Fisher's least significant difference, $P \leq 0.05$.

RESULTS AND DISCUSSION

Plant growth

Regardless of the shade treatment, plant height growth was similar during the growing season up through the July measurement date with Cherokee Brave™ flowering dogwood (Table 1). However, by August 2014, there was a significant difference in height with Cherokee Brave™ flowering dogwood among shade treatments compared to full sun and this continued until the end of the experiment. White shade cloth yielded the greatest height growth, but overall both 30 and 50% shade provided similar height growth. Plants grown under white shade were 48% taller and plants grown under black shade were 42% taller than plants grown in full sun.

However, even with this increased height difference with Cherokee Brave™ flowering dogwood in shade treatments compared to the full sun treatment, there was only a 25% increase in total shoot dry weight with plants grown under white shade and a 6% increase with black shade (Table 1). There was very little difference in root dry weight. The 50% white shade had the greatest root dry weight followed by 30% black, full sun, and 50% black treatments.

Mean trunk diameter was similar among treatments with Cherokee Brave™ flowering dogwood at the July measurement date with exception of the plants grown under 50% black shade cloth, which was significantly less (Table 1). This trend continued until the end of the experiment. Total trunk diameter growth (final measurement — initial measurement) was the greatest in 50% white and 30% black treatments. Full sun and 50% black treatments had the least trunk diameter growth with 50% black being the smallest.

Height growth with 'Cherokee Princess' was similar among treatments at the July and August measurement date (Table 1). By September, 'Cherokee Princess' showed a significantly greater increase in height with 50% shade cloth using either black or white shade. Plants grown in full sun were 15% shorter than plants grown under 30% black shade and 23% shorter than plants grown under 50% white shade. However, there was only a 7% (30% shade) and a 2% (50% shade) increase in shoot weight as compared to full sun treatments. Root dry weight was the greatest in 50% white shade followed by 30% black, 50% black, and full sun treatments (Table 1).

Plants grown in full sun had the largest trunk diameter on 7 July and was similar to plants grown under 30% black and 50% white, but significantly different from plants grown under 50% black. Total trunk diameter growth (final caliper — initial caliper) was larger in 50% white and 30% black followed by full sun and 50% black treatments (Table 1).

Table 1. The effects of shade type on height and trunk diameter growth of Cherokee Brave™ and Cherokee Princess dogwood.

Shade treatments	Total height growth (cm)			Total trunk diameter growth (mm)			Shoot dry weight (g)			Root dry weight (g)		
	7-Jul[1]	13-Aug[1]	14- Sept[1]	7-Jul[2]	13-Aug[2]	14-Sept[2]	7-Jul	13-Aug	14-Sep	7-Jul	13-Aug	14-Sep
Cherokee Brave™ flowering dogwood												
Full sun	24.7 a[3]	47.0 c	48.5 c	4.8 a	7.8 a	11.6 ab	115.0 ab	218.1 a	285.0 c	32.5 ab	68.8 ab	164.2 ab
Black 30%	32.5 a	68.1 ab	81.1 b	5.0 a	8.7 a	12.4 a	129.4 a	243.8 a	328.3 b	35.0 a	85.6 a	175.8 ab
Black, 50%	31.5 a	61.4 b	85.7 ab	3.4 b	6.3 b	10.8 b	87.5 b	172.1 b	279.6 c	22.5 b	61.9 b	144.6 b
White, 50%	35.5 a	77.4 a	92.9 a	4.9 a	8.9 a	12.4 a	128.1 a	245.6 a	383.3 a	30.6 ab	89.4 a	188.3 a
LSD	12.5	14.1	9.5	1.1	1.5	1.2	29.8	44.5	41.1	10.3	20.7	37.0
Cherokee Princess flowering dogwood												
Full sun	34.6 a	50.7 a	50.5 c	5.1 a	7.3 a	10.7 b	118.1 a	216.9 a	298.3 ab	33.13 a	76.9 ab	150.0 b
Black, 30%	37.1 a	58.7 a	69.5 b	4.2 ab	6.7 a	11.9 a	91.3 b	201.9 a	321.7 a	31.25 a	75.0 ab	198.3 a
Black, 50%	37.4 a	59.9 a	86.8 a	3.7 b	6.1 a	10.1 b	85.0 b	168.8 a	265.6 b	23.13 a	59.4 b	161.7 ab
White, 50%	43.5 a	64.9 a	83.5 a	4.7 a	7.1 a	11.9 a	108.8 ab	202.5 a	337.8 a	30.63 a	90.6 a	203.9 a
LSD	13.1	16.1	11.3	1.0	1.9	1.1	25.6	72.1	51.0	11.19	30.6	46.5

[1]Total height growth=Height measured on Sept 14, 2014-initial height measured on Feb 25, 2014.

[2]Total trunk diameter growth=Trunk diameter measured on Sept 14, 2014-initial trunk diameter measured on Feb 25, 2014.

[3]Means within columns followed by the same letter are not significantly different. Means separated using Fisher's protected LSD, α=0.05.

Light intensity

Phillips et al. (1991) reported that the light intensity with 20 or 55% shade cloth or shade color did not affect plant growth. However, our data shows that height, trunk diameter, and shoot dry weight were affected by the color of the shade and the percent intensity of the shade. Our data did agree with Montague et al. (1992) that dogwood under 40% white shade had some growth parameters that were larger than black shade at 30 or 50% and full sun and that any shade resulted in larger trunk diameters.

Root zone temperatures

Root zone temperatures during this study differed significantly between treatments. Root zone temperature for full sun treatments were recorded up to 41.1°C (106°F) and often exceeded ambient air temperature. Phillips et al. (1991) reported no difference in ambient air temperature. However, the root zone temperature was significantly less with plants grown under white shade than plants under black shade. Root zone temperatures were not greatly reduced until a 50% shade cloth was used. There were very few days that root zone temperatures exceeded 37.7°C (100°F) in both 50% white and black shade treatments (data not shown).

Container leachate

Container leachate collected from a subset of plants for both Cherokee Brave™ and 'Cherokee Princess' dogwood showed a similar response among treatments for electrical conductivity, and orthophosphate at most sampling dates with the following exceptions (Figure 1). On 21 May, the electrical conductivity, and orthophosphate levels were lower for full sun than the shade treatments; and orthophosphate had elevated levels on 16 July (full sun) and 28 August (full sun and 30% black) with Cherokee Brave™ flowering dogwood. The container leachate from 'Cherokee Princess' had similar levels of electrical conductivity, and orthophosphate at most sampling dates. As expected, electrical conductivity, and orthophosphate were initially high and remained so for about 12 weeks after potting; then stabilized around 0.2 to 0.3 dS m^{-1}. So with either cultivar, light intensity was not a major component of fertilizer release patterns in the container substrate.

Shade treatments regardless of color or density did have an effect on the plant growth of Cherokee Brave™ flowering dogwood and 'Cherokee Princess' dogwood. Plants grown under 50% black and 50% white had more height growth than plants under 30% black or plants in full sun. However, plants responded more dramatically from July to September than from February to July. This may be a result of the transplanting shock of the bareroot liners into container culture. Light intensity was not a major component of fertilizer release patterns in the container substrate. More research is needed to reduce the initial transplanting shock and refine the period and longevity of shade intensity of container grown dogwoods.

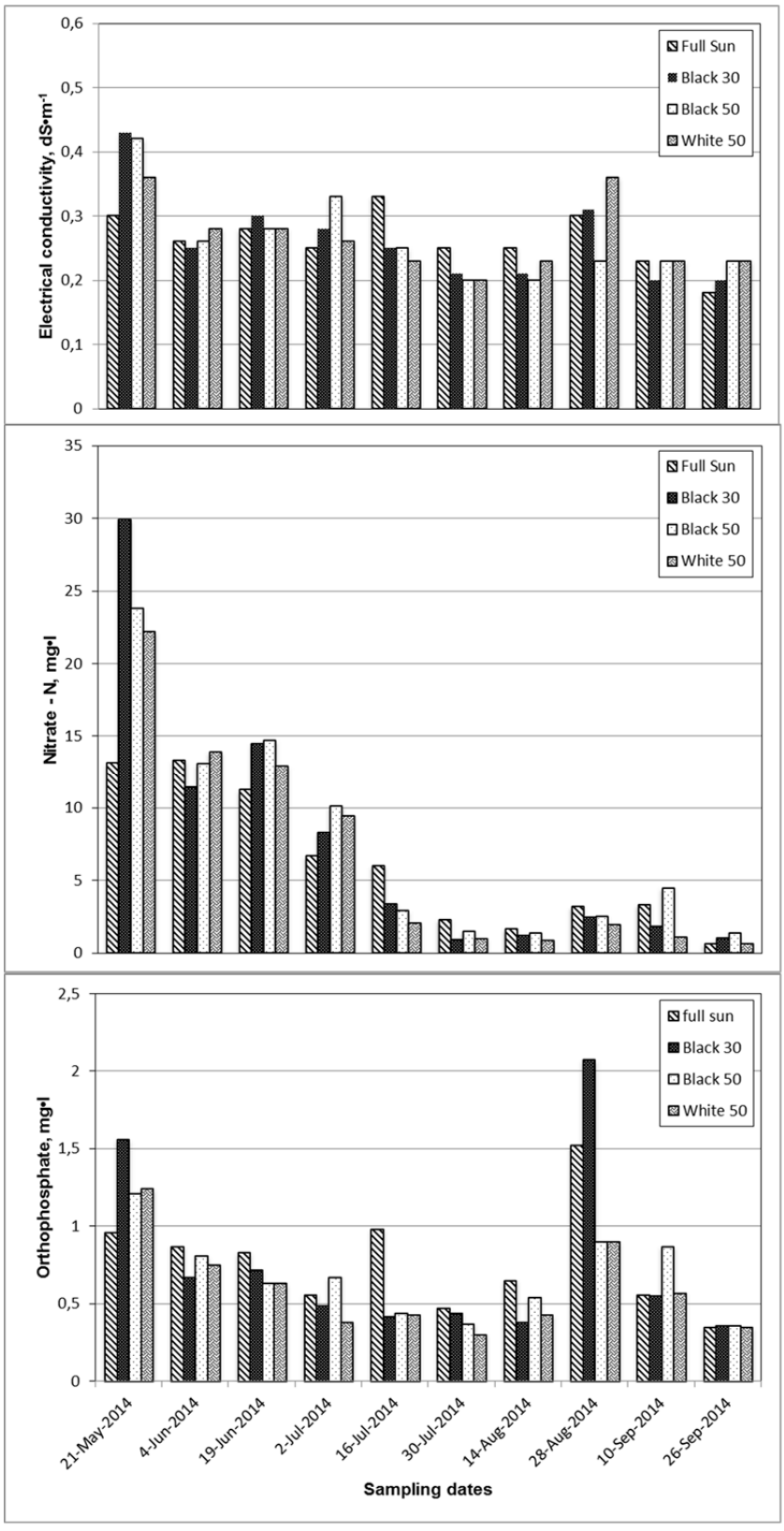

Figure 1. Electrical conductivity, nitrate nitrogen, and orthophosphate levels in leachate from container grown Cherokee Brave™ flowering dogwood. Note: top figure × axis = dS m^{-1}, middle figure × axis= mg L^{-1}, bottom figure × axis= mg L^{-1}.

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Amending pine bark with swine lagoon compost: is poo the answer?©[a]

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INTRODUCTION

Pine bark for use in the nursery industry is in short supply and at times not completely aged due to timber processing mills moving overseas (Lu et al., 2006). Growers are working to overcome these shortages, high prices, and quality issues by using other products (such as wood) or amending pine bark to stretch their supplies (Worley et al., 2008). Calcined clays can be used as an 8% (by volume) amendment to pine bark to increase buffering and water holding capacity as well as to reduce nutrient leaching in bark based substrates (Owen et al., 2007). Utilizing composted turkey litter as an amendment (at 4, 8, 12, 16% by volume) to pine bark increased available water but decreased air space (Tyler et al., 1993). Both Owens et al. (2007) and Tyler et al. (1993) emphasize the need to evaluate both the physical and chemical properties of an amendment to pine bark before adoption by the containerized plant production industries (nurseries and greenhouses). Therefore, before implementing a new substrate mix into an operation, impacts on plant growth, nutrient availability within the substrate, and changes to fertility programs must be considered. With many alternative substrates available, growers are looking for the most locally available substrate with the least increase in cost, and the ready availability of swine lagoon waste is an attractive option.

In North Carolina, production of hogs comprises $26,419,703 of North Carolina's $420,145,646 farm cash receipts (USDA, 2015). Incubation studies showed pelletized processed swine lagoon solids were an adequate source of phosphorus, but some plants, such as row crops, would require supplemental application of nitrogen (Duffera et al., 1999a). It has also been reported that the shoot dry weights of bermudagrass (*Cynodon dactylon* L. Pers.), sweet corn (*Zea mays* L. 'Silver Queen'), sorghum (*Sorghum bicolor* L. 'DK-54'), and field bean (*Phaseolus vulgaris* L. 'Blue Lake') in the Ap horizon of a Norfolk sandy loam soil mixed with processed swine lagoon solid were similar or superior to growth with a conventional inorganic fertilizer (Duffera et al., 1999b). The application of vermicomposted swine lagoon waste at 20% has been shown to increase plant dry weight in *Hibiscus moscheutos* L. 'Luna Blush' as much as 58% compared to 100% pine bark in a greenhouse setting (McGinnis et al., 2009). However, little research has evaluated the growth of herbaceous perennials in containerized plant production with swine lagoon compost (SLC) as the only source of nutrients. Therefore, the objective of this study was to evaluate the impact of increasing amounts of SLC to pine bark on plant growth.

MATERIALS AND METHODS

A study was designed as a randomized complete block with five replications to evaluate the impacts on plant growth of pine bark (PB) amended with varying rates (10%, 20%, 40%, 60%, and 80% by volume) of swine lagoon compost (pH 5.6) (n=25). Swine lagoon waste was dredged from a lagoon in Garland, North Carolina (Murphy Brown, LLC, Warsaw, North Carolina) and dewatered using a polymer (PT1051, PolyTec Inc., Mooresville, North Carolina) and a geotextile bag (TITANTube OS425/OS425A, Flint Industries, Metter, Georgia). The waste/polymer mix was pumped into the bags where the water filtered out and was pumped back into the lagoon. The bagged waste was allowed to drain for 2 years before use, resulting in swine lagoon compost (SLC). Once removed from the bag, the SLC was spread on plastic to dry with heat and forced air for a week, and then ground to 2 mm using a grist mill grinder (Molina Corona, Landers, Mora & Cia, LTDA., Medellin, Colombia).

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On 3 June 2015 seedling liners of *Musa velutina* H.Wendl & Drude, grown in 10.16 cm containers, were potted into 3.8 L (1 gal) (Classic 500, Nursery Supplies, Inc., Chambersburg, Pennsylvania) containers filled with pine bark (PB) amended with one of five increasing ratios of swine lagoon compost (SLC): 10:90, 20:80, 40:60, 60:40, and 80:20 SLC:PB (v/v). The plants were grown in a greenhouse (26°C day/18°C night temperature) with 50% shade (XLS Revolux Climate Screen, LivingShade, Hornsby NSW Australia), and natural irradiance and photoperiod. Plants were irrigated twice a day using low-volume spray stakes (PC Spray Stake, Netafim, Ltd., Tel Aviv, Israel). No supplemental fertilizer or lime was added.

Leaching fractions (LF = volume leached ÷ volume applied) were measured every 2 weeks (17 June, 25 June, 10 July) and irrigation volume was adjusted to maintain a 0.2 LF for each substrate. Irrigation water contained an average of 0.83 mg L^{-1} N, 0.21 mg L^{-1} P, and 3.44 mg L^{-1} K with a pH of 7.83. Additionally, substrate solution was collected every 2 weeks (10 June, 25 June, 14 July) using the pour-through nutrient extraction method (Wright, 1986). Substrate solution electrical conductivity (EC) and pH were determined via a combination EC/pH meter (HI 8424, Hannah Instruments, Ann Arbor, Michigan).

Total porosity (TP), airspace (AS), container capacity (CC), and bulk density (BD) analyses were conducted in the Horticultural Substrates Laboratory, Department of Horticultural Science, North Carolina State Univ., Raleigh, North Carolina. Substrate physical properties were determined initially at potting. Three replications of each substrate were packed into 347.5 cm^3 cylindrical aluminum 23 rings (7.6 cm dia, 7.6 cm ht) and they were used to determine TP, AS, CC, and BD according to procedures outlined in Tyler et al. (1993).

After 6 weeks, shoots were removed and roots were washed free of substrate. Shoot and root dry weights (dried at 60°C for 4 days) were determined and used for growth comparisons. The data were subjected to analysis of variance and regression analyses where appropriate ($P \leq 0.05$).

RESULTS

Total porosity and AS decreased linearly with increasing amount of SLC added to PB, while BD increased linearly with increasing amount of SLC (Table 1). Container capacity had a quadratic response to amount of SLC and was highest with 40% SLC added to PB. Substrates greater than 60:40 SLC:PB had AS that was below the recommended range (Yeager et al., 2007). Additionally, substrates with greater than 40% SLC (40:60 SLC:PB) had bulk density above the recommended range (Yeager et al., 2007).

Table 1. Effect of swine lagoon compost (SLC) additions to pine bark (PB) on total porosity (TP), container capacity (CC), air space (AS), and bulk density (BD) initially at time of potting on 3 June, 2015.

SLC:PB (v/v)[1]	TP	CC	AS	BD
	% vol.			(g cc^{-1})
10:90	84.05	52.39	31.65	0.19
20:80	74.51	54.75	19.76	0.20
40:60	77.88	64.16	13.72	0.28
60:40	74.90	63.48	11.41	0.32
80:20	55.05	49.11	5.95	0.44
ANOVA[2]	0.0016	<.0001	0.0030	<.0001
Linear[3]	0.0013	NS	0.0002	<.0001
Quadratic[4]	NS	<.0001	NS	0.0107
BMP Guidelines[5]	50-85	45-65	10-30	0.19-0.24

[1]The substrate consisted of: 10:90, 20:80, 40:60, 60:40, and 80:20 SLC:PB.

[2]Analysis of variance (ANOVA) effect of substrate ($P \leq 0.05$).

[3]Analysis of linear regression. NS=not significant, *P*-value given otherwise.

[4]Analysis of quadratic regression. NS=not significant, *P*-value given otherwise.

[5]BMP = Best Management Practices recommended ranges (in percentages) for substrates used in general containerized nursery production (Yeager et al., 2007).

The amount of SLC added to PB affected pH for each of the three sample dates (10 June *P*=0.0001, 25 June *P*=0.0001, and 14 July *P*=0.01) (Figure 1). On 10 June 2015 the substrate solution pH readings ranged from 5.9 to 6.6, while on 14 July 2015 it ranged from 4.8 to 5.8. Higher amounts of SLC did not always result in higher substrate solution pH. Two weeks after potting (10 June 2015), pH of the substrate solution increased quadratically as SLC in the substrate increased with a maximum at 60% by volume. Four weeks after potting (25 June 2015), there was again a quadratic response in pH to increasing SLC with the highest pH found again in the 60:40 SLC:PB substrate solution. At 6 weeks after potting (14 July 2015), there was again a quadratic response in substrate solution pH, but with a maximum at 10% SLC. All substrates maintained acceptable pH levels throughout the study (Yeager et al., 2007).

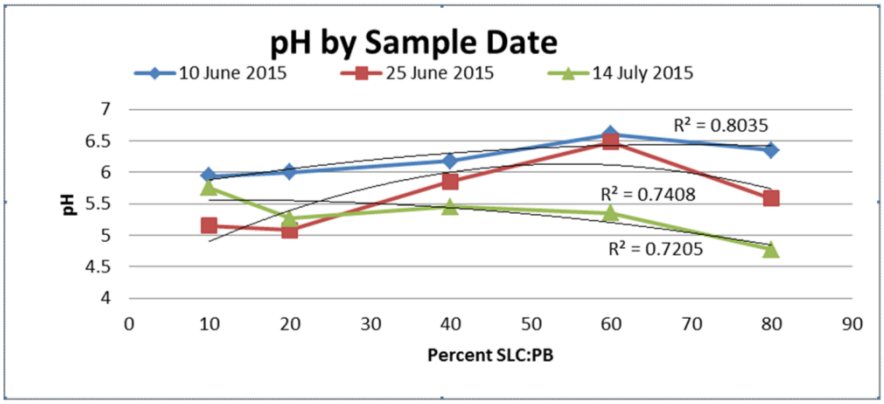

Figure 1. Effect of increasing amounts (10, 20, 40, 60, and 80% by volume) of composted swine lagoon solids added to pine bark on a substrate solution pH.

The substrate solution EC was also impacted by the amount of SLC added to PB at each sample date (10 June *P*=0.0001, 25 June *P*=0.001, and 14 July *P*=0.001) (Figure 2). At each sample date, EC levels increased quadratically as SLC in the substrate increased with the maximum EC found in the 40:60 SLC:PB substrate. While EC levels for substrates with less than 40% decreased over time, all substrates with greater than 10% SLC maintained unacceptably high EC levels (Yeager et al., 2007).

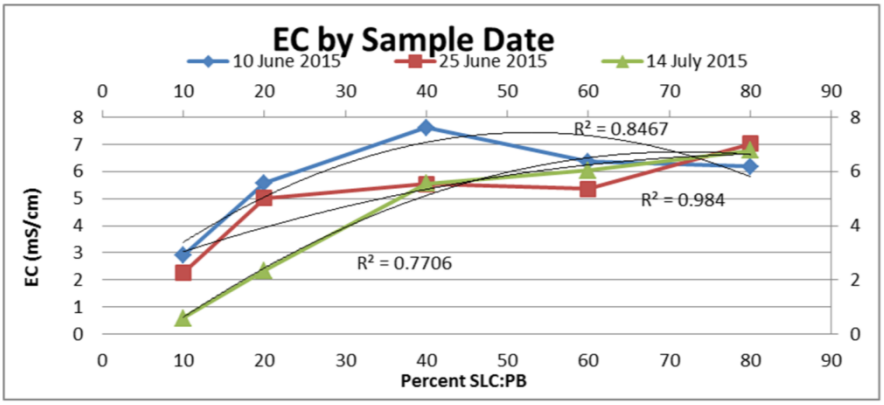

Figure 2. Effect of increasing amounts (10, 20, 40, 60, and 80% by volume) of composted swine lagoon solids added to pine bark on a substrate solution electrical conductivity (EC).

Musa velutina shoot and root growth were also both impacted by the amount of SLC added to PB (shoot *P*=0.0004 and root *P*=0.0008) with both root and shoot growth

decreasing quadratically as SLC amount increased from 10 to 80% (Figure 3). There was substantial growth reduction, particularly in roots, at rates of SLC greater than 20%. Additionally, roots of plants grown in 10:90 SLC:PB were nearly twice the size of those in the 20:80 SLC:PB substrate. Shoot growth showed reduction in dry weight at 20:80 SLC:PB and greater reduction at 40:60 SLC:PB.

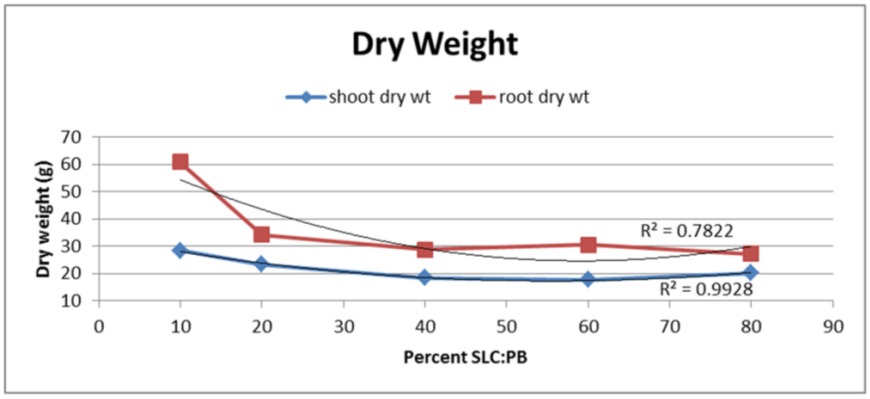

Figure 3. Effect of increasing amounts (10, 20, 40, 60, and 80% by volume) of swine lagoon compost (SLC) added to pine bark (PB) on root and shoot dry weights of *Musa velutina* using regression analyses ($P \leq 0.05$).

DISCUSSION

Pine bark substrate amended with SLC at all volumes (10, 20, 40, 60, and 80%) had TP and CC within the recommended ranges. However, when PB was amended with more than 60% SLC had AS that was below the recommended range and PB amended with greater than 40% SLC had BD above the recommended range.

Substrates with greater than 10% SLC (10:90 SLC:PB) produced smaller shoots and roots of *M. velutina*, in contradiction to results seen by McGinnis et al. (2009) with *H. moscheutos* 'Luna Blush', where shoot growth dry weight was consistently greater in pine bark amended with 20% (by volume) vermicomposted swine waste. Tyler and Warren (2000) also saw increase in shoot growth of *Rudbeckia fulgida* var. *sullivantii* 'Goldsturm' when pine bark was amended with 8% (by volume) composted turkey litter. Root growth was likely reduced by high substrate solution EC which damaged roots. Burned root tips were observed (visual observation). All rates of SLC maintained acceptable substrate solution pH throughout the 6-week study.

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IPPS young propagator exchange program©

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EXCHANGE PROGRAM — VISIT TO DENMARK

The 2014 International Exchange Program offered through IPPS Southern Region was hosted in Denmark 20-27 September. This opportunity is afforded to a Southern Region members age 35 and under. Thank you for selecting me to be the 2014 representative. This experience has enriched my career immensely.

This paper is dedicated to three IPPS Southern Region members who have influenced me greatly through active membership: Mr. Bob Black, Mr. Tom Saunders and the late Mr. Wayne Sawyer. I would like to express my sincere gratitude to my hosts Mr. Bent Jensen and Ms. Marianne Bachmann Andersen who welcomed me into their homes and made my experience authentic and engaging. The European Region members instantly welcomed me and have continued to serve as knowledge sources and inspiration for me.

My tour began in Copenhagen and over the 10 day stay I visited Køge, Sorø, Helsingør, Nyborg, Svendborg and Odense, Denmark and Helsingborg and Malmø, Sweden. Denmark is a fascinating country with a storied history dating back to the Vikings (Figure 1). Often held in the highest esteem for its environmental and energy policies, this society of just over five million residents is a global leader in design, architecture, farming, green technology and pharmaceuticals.

Figure 1. Map of Denmark.

The Danish strive to maintain healthy ecosystems by designing new and sustainable ways of living, environmentally friendly transportation, green infrastructure expansion and renewable energy sources such as wind turbine generated electricity. This ecologically sensitive approach to development has influenced Danish agriculture in many ways.

Denmark has the highest market share of organic products in the world with organic food making up 8% of the total food market. Some 7% of Danish land is used for organic

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farming with a goal of reaching 15% organically farmed land by 2020. Many horticulture producers are following suit by developing environmentally responsible growing systems and utilizing bio-controls for pest and disease management.

Retail garden centers of the Scandinavian region have many interesting innovations that could be applied to the independent garden centers and box stores in the USA. Cross merchandising for the outdoor lifestyle was a dominant feature. Furniture, garden art, grills and cooking supplies were all included in displays with traditional plant material. Halloween decor was highlighted including costumes, makeup, lights pumpkins and gourds. Haunted houses were created within greenhouse spaces attracting a broad demographic of customers including young families.

Plant material was consistently labeled thanks to an initiative of the Danish Nursery Association (Figure 2). The commitment to develop a unified marketing strategy includes education and community outreach from wholesale growers to garden centers ensuring that all horticulture professionals play a role in educating the consumer.

Figure 2. Consistent labeling of plant material.

Morten Sloth, production manager at Gunnar Christensens Plantskole and IPPS European Region member explains “It is a big job to market plants, but we are constantly trying to come up with little things that can bring attention to those plants which we produce.” Morten has created a pocket guide describing the many cultivars of strawberries, rhubarb and blueberries grown at the nursery. These are supplied to the employees at garden centers providing proper advice and a pleasurable buying experience for the customer.

Other noticeable trends in the garden center market included potting soil with a plant specific mixture of substrates. Organic fertilizers were featured through-out displays and ornamental edibles such as 1-gal pots of turnips (Figure 3), figs, blueberries and greens were incorporated with woody ornamentals as Foodscape vignettes. Through this cross merchandising technique consumers can visualize how a bio-diverse collection of plants will work in their home garden.

Figure 3. Ornamental edibles as foodscape vignettes — turnips in 1-gal containers.

One of the most striking products featured at garden centers and public gardens were the potted living willow topiaries (Figure 4). The tightly woven branches of a range of *Salix* species create high impact architectural detail through every season. These trained art pieces add dimension, novelty, and extravagance to the landscape while adding a unique interest to container gardening.

Figure 4. Topiaries of containerized living willows.

Coppiced deciduous trees and conifer specimens such as *Abies, Picea, Thuja*, and *Juniperus* were pruned into topiaries reflecting the manicured Danish design aesthetic. These stylized plants are ideal for consumers of any age and are well suited for small spaces

including patios and balconies.

Local produce, fresh cut flower bouquets, and indoor plants were heavily represented in retail outlets from small town markets to box store garden centers. House plants are an important market for Danish retailers and the creative displays of uniquely grown specimens and colorful selections make it a delightful section to browse through.

A diverse range of production facilities were highlighted during the IPPS Europe tour (Figure 5). Nursery innovations were featured at Gunnar Christensens Plantskole where a series of rotating presentations included strategies to reduce liverwort by using pre-formed wool top dressings. Bio-controls and beneficial insect applications were demonstrated. The use of conifer windbreaks and strategies to efficiently and sustainably develop nursery properties added great value to my experience.

Figure 5. Greenhouse production facilities during IPPS Europe tour.

Bio-dynamic farming and organic herb production were explored at Kiselgården a family run operation. This facility embraces a holistic approach to growing edibles with a diversified, balanced farm ecosystem that generates health and fertility from within the farm. Serving world renowned Michelin star restaurants like Noma and Geranium — the delicious produce is appreciated by the elite foodies.

New plant selections in an extensive trial garden at Gasa Young Plants gave insights to the future of global plant production. Innovations such as automated transplanters and high spectrum lighting were discussed while touring Gartneriet PKM, the world's largest producer of *Campanula.* Organic fruit production was the focus at Aqua Vitae Sydfyn, a Danish snaps distillery.

Møllegårdens Planteskole featured two brilliant growing strategies to make landscape installation more cost effective. Prima Færdig Bunddække® is a ground cover system consisting of plants that have been grown in a ready-made rolled mat, much like sod (Figure 6). These instant groundcovers are easy to install and reduce weed pressure by providing immediate ground coverage upon installation. Prima Færdig Hæk® is a high quality large hedge system developed to provide on the spot screening in new landscapes. With a wide range of plants in production these exclusive lines are very popular across Scandinavia.

Figure 6. Prima Færdig Bunddække® ground cover system of plants that have been grown in a ready-made rolled mat, much like sod.

The IPPS meeting revolved around the theme The Digital Nursery. The presentations provided many insights into the opportunities and challenges we face as plant producers in a time of rapidly changing technology. Anticipating the needs of the millennial consumer and adopting technologies to enhance the educational component of gardening is paramount.

Horticultural production is diversifying globally to include planting strategies that fulfill ecological needs, food production, urban infrastructure and a changing aesthetic value. The International Plant Propagators Society offers valuable resources for nursery professionals to develop their careers with collaborative influences. The International Exchange Program is an ideal platform to promote and expand membership with young professionals.

Evapotranspiration based irrigation at Saunders Brothers Nursery©

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INTRODUCTION

Saunders Brothers is a wholesale nursery located in central Virginia — in the foothills of the Blue Ridge Mountains. The company began in 1915 as a family farm, growing corn, tobacco, and raising cattle. In 1947, Paul Saunders stuck his first boxwood cuttings as part of a 4H project. In the 1980s as some of Paul's seven sons were finishing school and returning to the family business, the farm expanded to include an ornamental nursery. Like many folks in the industry, Saunders Brothers grew quickly in the 1990s and through the 2000s. We now grow over 500 different taxa of ornamental plants, 25 different cultivars of boxwoods, and 50 different types of fruit.

The company and the industry suffered during the recession of 2008. Supply outstripped demand and customers were reluctant to buy nursery product, as their expectations for quality increased. At the same time, the costs of our inputs were also increasing. This got us thinking: what can we do to decrease our input costs while also increasing quality?

OPPORTUNITIES TO IMPROVE IRRIGATION, ENVIRONMENTAL IMPACT AND PLANT QUALITY

At the 2010 IPPS Southern Region of North America Annual Meeting, we were presented with the opportunity to work with Tom Yeager and Jeff Million at the University of Florida to look at irrigation and plant water needs. Tom Saunders has always been fond of saying, "the person who controls irrigation does more to affect the quality of your crops than anyone else." So it seemed that this work had the potential to help with the challenges we were facing. Specifically, we were aiming to improve on the following areas:

- Plant quality: less disease, fewer losses, more repeat orders.
- Bottom line: the Saunders family has a very close connection to Nelson County and the Piney River area, and we strive to be good stewards of the land. But we are also practical, so anything we do needs to make financial sense for the company.
- Environmental impact and risk: even though we are fortunate enough to live in an area where water use is not heavily restricted, there are still obvious benefits to better utilizing and conserving resources. Our water supply can be unpredictable which impacts nursery production. We utilize the Tye River as our primary water source. During the past 100 years, Saunders Brothers has seen the river flood during Hurricane Camille and go dry during a hard drought year. Being less dependent upon water makes us more secure in the long term.

So that is why we were interested. But what is the big idea behind the project? What is driving this irrigation research? We like to call it the "Goldilocks Dilemma;" the idea that plants need a certain amount of water for optimal growth. Too much and they rot, too little and they are scorched. We want it to be "just right." If plants are at field capacity in a well-drained medium, they should be neither limited nor saturated. So the goal of the research and the system that developed from it was to quantify the amount of water a plant loses in a given day and to resupply that amount of water without excess.

COMPONENTS OF THE IRRIGATION SYSTEM

Our irrigation system has three major components that we categorize into two sections: the brains and the brawn. Basically, we want to be able to gather information about

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the environmental conditions the plants are experiencing each day, use that information to determine how much water our plants need, have the irrigation system process the information, create a schedule, and replace that water.

"The brains" are the science behind the system. The container irrigation (C-IRRIG) software program was developed to generate daily irrigation run times. These times are based on a couple of different pieces. First is evapotranspiration (ET) which is how much water a plant loses during the day. Second is irrigation rate and uniformity, which tells us how much water we are applying during an irrigation cycle. Lastly, leaching fractions (LF) which tells us how much irrigation water is actually making it into the container and available to the plant.

Evapotranspiration (ET) is measured by taking a weight of the plants in containers at the beginning of the day, after a normal irrigation cycle to get a wet weight. They are then again weighed at the end of day, around sunset after the plants are no longer transpiring and losing water, to get a dry weight. The difference between those weights enables us to find the volume lost and quantify how much water was lost in vertical inches. That is the plant's evaporative needs or what we refer to as the plant's ET for the day.

Irrigation rate and uniformity are how much water is applied and how it differs over a given area. This is an important component because it affects how long one can irrigate to get a certain, desired volume of water. Consider it much like calibrating something to know you are getting the volume you think you are getting. We measured this by setting up a grid of cups/containers in a house, ran a normal irrigation cycle, and recorded the volume in each cup. This data gives us a representation of how much water plants are getting relative to each other in the house. If you have variable results then the grower needs to improve irrigation uniformity and avoid the edge effect.

At Saunders Brothers we had a lot of challenges with this component. Our nursery is anything but level since it is located in the foothills of the Blue Ridge Mountains. There is a 61 m (200 ft) change in elevation from the river where water is pumped to the distribution pond to the highest point in the nursery. In order to put out the same volume of water, we may have to irrigate in the lower areas for 30 min [8 mm (0.3 in.) h^{-1}] and valves in the upper areas for 60 min [15 mm (0.6 in.) h^{-1}].

Leachate Fraction (LF) is measured much like ET, however we are starting with dry weight measured at the end of the day and taking the wet weight (the weight of the saturated plant and the collected leachate) the following morning. This is done after a typical irrigation cycle to determine how much water the plant received.

We spent the first 2-3 years of this process working with the University of Florida to fine-tune these measurements. The components and tests were a large part of what went into the research and development of C-IRRIG software.

THE CONTROL SYSTEM

The heart of C-IRRIG is the zone-editing page where one enters individual grower inputs for separate areas, houses or crops. This is where irrigation rate, uniformity and desired leaching fraction are entered as well as information about the plants. When all this information is combined with the weather station data, and the grower inputs — C-IRRIG will calculate the previous day's ET and determine how long you need to run irrigation in a given zone to replace the water loss.

One important aspect about C-IRRIG — it can be run independently. If one is happy with the controllers used and just want something that will give you run times based on ET, C-IRRIG will do that. There is no need for added wires or sensors, just the grower inputs. However, we quickly found ourselves adjusting and reprogramming Irritrol boxes daily. It became very time consuming and soon we realized we needed a better controller.

"The brawn," as we call it, is the FRALO control system. This portion manages and runs the irrigation schedule. It consists of two pieces: a software portion that allows you to edit the schedule and a hardware portion, and the control boxes in the field.

The software portion, based in Microsoft access is where the grower has the ability to build and edit the irrigation schedule. The grower has the option to enter and edit irrigation

run times and priorities. Using individual valves, one can set a priority (which could mean running X before Y), duration times, etc. The software also displays information about flow volume and pumping capacity, and total gallons used by zone. There are options for setting days to run and start times. The nice thing about the program is that it offers a lot of flexibility. One can separate different crops based on medium and irrigate crops differently. You can also set up cooling cycles or cyclic irrigation.

Once the schedule is set, it is sent wirelessly to the control boxes in the field. The controllers can be operated remotely from a PC, laptop, phone, or manually in the field. Each controller has a touch screen that shows information about flow, pressure, and run times for each valve. These boxes help to optimize system pressure and maximize efficiency.

Both C-IRRIG and FRALO can be used independently. However, we have found the magic happens when used together. At Saunders Brothers, we have spent the last 2 years working to combine "the brains" and "the brawn" to get an irrigation system that alters run times daily and is automated from start to finish.

We have covered a lot about the development of the system, but what does it actually take to get it set up and manage it on a day to day basis? It all starts with the configuration of the system, everything from installing infrastructure to doing the uniformity and rate tests. Obviously, there is a significant time investment at the beginning, but minimal time once the system is up and running. The majority of day-to-day time spent on the irrigation program is updating grower inputs and verifying that the program is working. It is definitely not a "plug and play" system, and requires active time in the field checking to make sure the system is functioning.

BENEFITS OF OUR IRRIGATION SYSTEM ON PLANT QUALITY AND COST REDUCTION

Our irrigation system has led to increased plant quality and a reduction in production costs. With the improvement of the irrigation system in conjunction with IPM practices, pesticide usage has decreased by 50% during the past 3 years. There are decreased weeds and reduced losses in overall plant production. This system has enhanced our ability to grow better plants that were difficult to produce in the past.

The system has impacted our financial bottom line. Keep in mind that these financial benefits will vary among nurseries. Our total annual nursery savings are estimated at $80,355 (Table 1). The annual labor savings are from the reduction of time that irrigation managers are in the field opening and closing valves, or changing Irritrol boxes in the field. With the automated system, all those daily changes are taken care of, freeing management for other tasks. Single crop quality savings is the opportunity cost to grow crops that we had previously struggled with. Overall plant quality savings comes from an estimated reduction in losses by 1% per acre of growing space. At our nursery this is an estimated 100 plants for every 15 houses. The fertilizer savings is an estimate of where we were able to reduce our fertilizer use. In a single year we saved more than $10,000. Hence, the estimated annual savings is $80,355 for the nursery. This creates a new bottom line and raises the bar for us as a company.

Table 1. Estimated savings and plant quality enhancement of irrigation system at Saunders Brothers Nursery.

Annual labor savings	$6,355
Single crop quality savings	$4,000
Overall plant quality savings	$63,000
Fertilizer savings	>$10,000
Total annual savings	$83,355

Lastly, the environmental benefits of reduced water usage were significant (Table 2). With an average rainfall in our area of 1194 mm (47 in.), we use some 630 million L (166.5 million gal) of water (Table 2). In 2012, which was considered a dry year, we were able to

reduce water usage by 51%. We have continued to do so in subsequent years, with an overall reduction of 56% during the past four years. Because we are using water more efficiently, we have been able to decrease fertilizer rates of some crops by 30%, since less is leached from containers. In addition, we can extend the time between herbicide applications because less herbicide is being washed away.

Table 2. Environmental benefits of reduced water and fertilizer usage of irrigation system at Saunders Brothers Nursery.

Year	Rainfall (in.)	Rainfall (mm)	Water Use (million gal)	Water Use (million L)
2009	48	1219	167	632
2010	46	1168	166	628
2011	57	1448	77	291
2012	37	940	81	307
2013	58	1473	70	265
2014	40	1016	67	254

All in all, this has been a great project to be a part of and we have been extremely fortunate to work with some fantastic people. We thank Russ Illig from FRALO control systems, and Tom Yeager and Jeff Million of the University of Florida.

Who wants to be a researcher? Getting meaningful results from on-site nursery research trials©

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INTRODUCTION

Many nursery growers have been conducting on-farm research trials for years, either independently in order to improve upon current production practices, or in cooperation with university, government or industry entities. By conducting research trials to answer specific questions, growers are able to develop real-world solutions based upon their specific needs at their location. Conducting a research trial in addition to managing normal nursery activities could seem like a daunting task. However, research trials can be designed and specifically tailored to meet a grower's needs in terms of time commitment, resources, space, or any other constraint.

The objectives of this paper are to outline the benefits of conducting on farm research, provide an overview of how to properly design research trials, and illustrate how to draw meaningful conclusions from research results.

WHAT IS ON-SITE NURSERY RESEARCH (OSNR)?

On-site nursery research (OSNR) is replicated, scientifically valid research conducted by growers — with or without the help of researchers. On-site nursery research is more than applying a new practice to a portion of your crops to make side-by-side comparisons or treating a single block of plants with a new herbicide to see how it performs. These types of activities would be classified as "demonstrations" which by definition are not valid experiments, but do offer value in observing how a new practice would work at your location. However, demonstrations do not have to be replicated or randomized, and do not sample the variation within a test area (Fishel, 2006; Veseth et al., 1999). It is not possible to make reliable comparisons using demonstrations only, so the best way to make management decisions would be to rely on well-designed research trials.

WHY CONDUCT ON-SITE NURSERY RESEARCH?

The purpose of conducting field research on nursery crops, or any crop for that matter, is primarily to try and help answer questions and solve production issues. Theoretically, applied nursery research is conducted on a small-scale in a somewhat controlled environment. One of the reasons most research is done on a smaller scale (besides funding limitations) is because smaller trials make it easier to reduce background "noise," which is also called experimental error. Background noise (or experimental error) are factors and variables that could influence trial results and may reduce or increase treatment effects such as: pest pressure, weather conditions, media, irrigation uniformity, or countless other factors. Treatment effects are evaluated under controlled conditions and then the results are used to predict outcomes on a larger scale.

Conducting your own research can also be used to confirm that research results and product claims are applicable to conditions and crops at your nursery (Nielsen, 2010). For example, a research report was published indicating that a new substrate amendment was shown to increase growth of *Hydrangea quercifolia* by 15% and reduce irrigation by 10% when added to pinebark and sand substrate. However, a nursery in a warmer climate may produce *H. macrophylla* and use a substrate comprised of primarily pine bark and sphagnum peat moss. In this case, before implementing major, widespread change in your production

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practices, it would be wise to conduct a small experiment to confirm that similar results can be achieved under your growing conditions or with different crops. The same would be true for field production nurseries as soil types can vary greatly from place to place. Overall, the major benefit of conducting research at your nursery to determine if a change in chemicals, substrates, growing methods, etc. is going to be effective for YOU – your crops, your environment, and your equipment.

PLANNING AND DESIGNING YOUR RESEARCH TRIAL

Research trials should follow a systematic approach — first, a question or hypothesis is developed that you need an answer to such as: Can I increase crop growth by using a different fertilizer ratio or timing? Can I apply this herbicide to my crops without causing phytotoxicity? Will this plant growth regulator increase flowering? Then the research trial is designed to answer the question/hypothesis at hand and data is collected, recorded, and analyzed without bias. Before conducting a field experiment, it is best write down the answers to the following questions in order to make the experiment more valid and useful to you:

- What are my objectives? (Reduce water use, increase crop growth or rooting percentage, utilize a new pesticide, etc.).
- What is the best way to design the experiment so that my results are most useful?
- What is the best/most efficient way to arrange my treatments and plots?
- What variables exist that could impact trial results (pest pressure, differences in field soil characteristics, weather, etc.)?
- What kind of data will be collected? How often?
- How will I analyze and use the data?

Once you determine your objective, you would first select one or more treatments to evaluate in addition to a control treatment. In most cases the control treatment would be your normal production practice. A control treatment is used to compare alternative methods (your treatments) to your standard method. Without a control it is difficult to determine if the treatment performed better or worse than your standard method. The most straight forward research goal would be to answer simple yes/no questions such as "Is herbicide "A" or herbicide "B" safer to use on my crops?" In this case, a simple trial could be conducted and treatments may only consist of herbicide A vs. herbicide B vs. a control (no herbicide). Depending on available space, you may also choose to investigate various rates of both herbicides to determine optimal rates and margin of safety. If you wanted to determine the optimum rate of fertilizer or a rooting hormone on a certain crop, it would be wise to include a wide range of treatment levels (rates), including a control.

There are many different ways to properly design experiments, but all include the basic components of replication and randomization of treatments. Replication and randomization both function to decrease experimental error, or "noise" and make data valid. A replication could be a large portion of a container growing area or field — or a single container-grown plant. A replication within a trial would be considered an "experimental unit." If treatments in a trial are not replicated, the results are invalid. Without replication there is no way of knowing if a treatment caused an effect, if the effect was due to some other factor or if the results are a due to merely chance. The number of treatment replications you need will depend on the question you need answered and also the magnitude of the differences you want to uncover. Detecting only major differences will usually require fewer replications. The more replications you have the greater the confidence in your results. However, as the number of replications increase so does time and expense. Often the number of replications will depend on available space, time, and resources. At least three or four replications are needed to be able to analyze the data, but 6, 8, 10 or more is preferred.

Randomization is needed for the same purpose, to reduce "noise". For example, if a field trial was designed with two treatments ("A" and "B"), and all of the "A" treatments were located on the west side of a nursery pad and all the "B" treatments were located on the east side of a nursery pad, we could not be certain of treatment effects because all the treatments

were grouped together (one treatment may have received more water, sunlight, pest pressure, etc.). By replicating and randomizing, we can be more certain of trial results.

EXPERIMENTAL DESIGNS

The simplest design is the completely randomized design (CRD). In a CRD, treatments (and controls) are assigned completely at random to a previously determined set of experimental units (plants, field plots, etc.). For example, if someone wanted to test four treatments (A, B, C, D) and a control (E) a completely randomized design could be set up (Figure 1). Completely randomized designs might be useful in testing a large number of treatments. A CRD is also appropriate when plant material is uniform and the environmental conditions are similar across the entire experimental area (such as in a greenhouse). While CRD are simple, they can create more "noise" than other types of designs, especially if it is conducted in nursery field soils (due to variability) or if there are differences in experimental units (plant size, health, etc.). In those cases, a randomized block design may be yield better results.

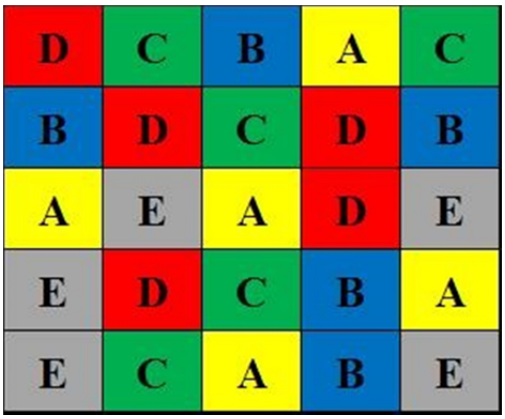

Figure 1. A completely randomized trial with four treatments (A, B, C, D) and a control treatment (E). Each treatment is replicated five times and treatments and controls are assigned at random.

A randomized complete block design (RCBD) is used to account for natural variability among treatments that might impact treatment differences. In RCBD, treatments are assigned at random to a group of plots (called blocks). Each block will contain one replication of each treatment (Figure 2). This design is useful in the field or if there is a lot of variability among plants used as experimental units. For example, if part of a field was poorly drained, plants in a research trial might also grow poorly which would impact trial results. One way to alleviate this issue would be to place a "block" of treatments in that area so that one replication of all treatments was in the poorly drained area (in addition to having other replications in more favorable areas). Another scenario where blocking would be useful is in an experiment that test the impact of a pesticide on crop growth, but your experimental units (plants) were not of uniform size. In this case, you could "block" the largest plants together and then have subsequent blocks of plants of similar sizes. Blocking according to plant size insures groups of plants with similar sizes received all treatments. There are several other ways to design experiments including split-plot designs, split-block designs, Latin square designs, and factorial designs, all with advantages and disadvantages. The easiest way to determine which type of design is best for your needs is to consult with university researchers, county extension agents, or others who conduct research regularly.

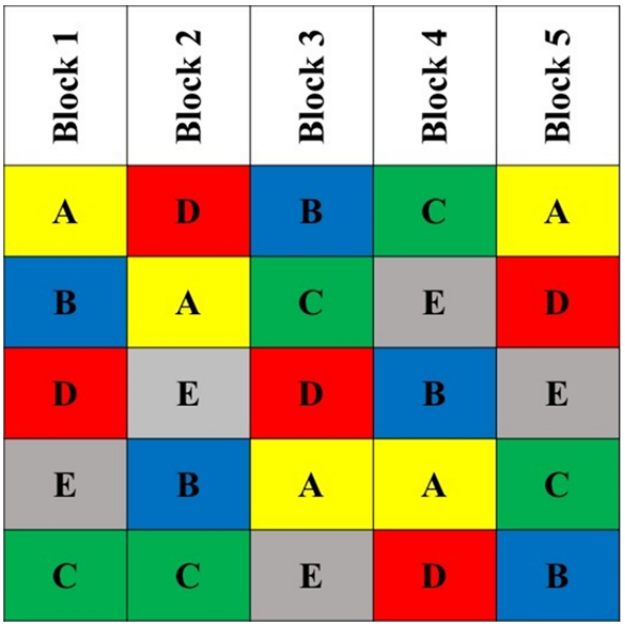

Figure 2. A completely randomized block design with four treatments (A, B, C, D) and a control treatment (E). Each of the five blocks contains one replication of each treatment and treatments are randomly assigned within each block.

ELIMINATING VARIABILITY

It is important to eliminate as many factors as possible that could influence trial results. Often times in weed science, we test different herbicides at different rates to determine if the herbicide causing injury or growth reduction to an ornamental plant. When controls are included, they usually receive no herbicide — and consequently may be filled with weeds within a few weeks which could impact crop growth, and thus trial results. A way to reduce this noise would be to regularly hand pull weeds from the controls so that any growth reduction could be attributed to the herbicide treatment, not weed competition. Treat all treatments as similarly as possible. If you had to move some plants from a shade house or greenhouse in order to treat them, move the controls also and not just the ones you are going to treat. Noise can be reduced by using the correct experimental design, using adequate number of replications, carefully selecting experimental units, and by treating all treatments as uniformly as possible.

DATA COLLECTION AND NOTE TAKING

Growth (height and width, caliper, etc.), flowering, substrate pH/EC, weed counts, and rooting percentage would all be forms of quantitative data — data that is measurable and recordable. Qualitative data, such as injury/phytotoxicity ratings, health ratings, or marketability ratings is subjective but can also be very valuable. The data that needs to be collected, and how often it needs to be collected will depend on the questions you need answered and what you are trying to achieve with the trial.

In addition to collecting data at set intervals, taking plenty of notes throughout the trial is invaluable. Pest pressure, unusual weather patterns, field operations, and other factors that could influence trial results should be documented throughout the trial. Regularly monitoring the trial would be ideal as you could correct any issues that may occur before they ruin the trial.

DATA ANALYSIS

Eliminating background “noise” or experimental error entirely is impossible, but statistical analysis allows us to identify background noise so we can more clearly detect these factors and better determine true treatment differences. The easiest way to analyze

data would be to compare averages across treatments using a program like Excel®. Statistical packages are available but are complicated, expensive and can take years to master. Some statistical software packages are available online for free, but are also difficult to use. A professional analysis using statistical software will provide you with more reliable results. Most university and extension personnel are happy and willing to collaborate with you on your trial and typically have access to statistical software. Most ONSR trials can be analyzed fairly quickly.

Is it always necessary to analyze your data statistically? Maybe not depending on your needs and the trial results desired. If your trial was properly designed and one treatment consistently outperformed the others in terms of size, flowering, or other parameter important to you, there is a good chance your results were statistically significant and you will know which treatment was most effective. Analyzing the data statistically just helps you to make your conclusion with more certainty. However, it should be noted here that poorly designed trials cannot be saved by statistics. If you are unsure if your design is going to provide useful results, do not hesitate to ask for help.

USING THE DATA

Before wide-scale recommendations are made, researchers typically repeat studies several times at different locations to validate results. OSNR is slightly different because these results are specific to your own situation. Repeating OSNR may be necessary if results are inconclusive due to unknown factors or noise. Repeating experiments may also provide further validation of previous results. Further validation is always important before making major and potentially costly production changes. Repeating trials may be limited due to time and resources.

CONCLUSION

Conducting OSNR can be enjoyable part of the nursery management process and may lead to significant improvements at your nursery. It can also be a time consuming, difficult (and possibly expensive) process. Do not hesitate to contact your local extension office or state extension specialist to ask for guidance. Most will be more than happy to assist you in any way possible and the process can be mutually beneficial.

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A simple and efficient method of germinating cycad seeds©

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INTRODUCTION

Cycads, an endangered group of plants from the world's tropics and subtropics, have been a mysterious and intriguing plant group to botanists since they were first documented more than 200 years ago. The number of described species continues to grow as subtropical and tropical regions are thoroughly explored; the latest count published in the World List of Cycads (*q.v.*) is 343. Interest in these plants has grown tremendously over the last 20 years, especially since accurate information has become readily available on the internet. The World List of Cycads, the Cycad Pages, the Cycad Society's Web site, and a number of other groups readily share information and photographs.

Many species of cycads are endangered, and both plants and seeds can be both difficult and expensive to obtain. The seeds of several species can be difficult to germinate and keep alive. The purpose of this paper is to explain and recommend the "baggie method" of germination, a technique that already is well-known in palms. It is not a new method for cycads by any means, but too many people are still unfamiliar with its ease and benefits. The method increases germination percentage and survivability of scarce and expensive seed. The information is especially useful to both the nursery industry and hobbyists; it will ultimately reduce the pressure exerted by poaching on indigenous cycad populations by making plants of the species easier to obtain. Indirectly, greater availability and ease of germination will reduce cost per plant, making cycad species readily available to those who wish to grow them.

Status of wild cycads

Unfortunately, at the same time as we continue to document new cycad species, habitat destruction and poaching continue to exact a heavy toll on wild cycad populations. Many species may become extinct in the wild. This is not new knowledge, with notable figures such as the late Cynthia Giddy, working as tireless advocates for the protection of cycad habitats in the 1960s. The IUCN Red List of Threatened Species can be accessed at: htpp: www.iucnredlist.org, and detailed information on each threatened to endangered species can be found. Unfortunately, lack of knowledge about cycads has led to some imprudent regulations prohibiting seed collection from the wild. Very few seed produced by cycads in the wild result in mature, fertile offspring. Making allowances for collection of some seed from wild populations would dramatically increase the number of living plants of a given species, and reduce pressure on wild populations. Ironically, the prohibition of wild seed collection has increased the amount of poaching and resulted in some species becoming more endangered, since it is almost impossible to protect every endangered cycad population in the wild from poaching. In fact, the IUCN Red List documents four *Encephalartos* species that are now extinct in the wild due to poaching.

Seed germination

While cultivating cycad species out of habitat is of limited use in preventing extinction, it can be of great utility in making many species available to those who might otherwise traffic in illegal collected plants. There are enough privately and publicly held cultivated specimens of many species to make seed available. The cost of seed is still high compared to many other groups of plants, but this cost is considerably less than the price of a germinated seedling or a plant poached from the wild. The value of providing someone with a plant that

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is legally obtained is inestimable. The relative availability of seed alone is an invitation to the horticulturally curious to attempt germinating their own seeds, with the great benefit of making the cost per plant reasonable for most collectors. The Cycad Society has a seed bank available to its members that routinely offers seed of fairly rare species at reasonable prices, and many members have developed formidable plant collections just by obtaining and growing Cycad Society seed bank offerings over the years.

The major problems in growing most cycads from seed (though there are exceptions to this generalization) are: (1) the seeds of most cycads have a fleshy sarcotesta (outer seed coat) with germination inhibitors that must be removed; (2) when the ripe female cones of many cycads disintegrate, dropping their seeds, embryos are often underdeveloped, requiring time, sometimes several months, for the embryos to reach maturity and germination to become possible; and (3) hard sclerotestas (inner, stony seed coat) of many cycad seeds resist penetration by moisture, thus slowing germination. The end result of these factors is that cycad seeds under normal greenhouse or shade house conditions, when they survive, germinate slowly and over a long period of time — a perplexing scenario to many nurserymen.

Three papers (Dehgan and Johnson, 1983; Dehgan and Schutzman, 1983, 1989) explain the relative impenetrability of a cycad seed coat and immaturity of the seed of some species at cone dehiscence. The drawbacks to the proposed method are twofold: the potentially dangerous and/or expensive chemicals to improve germination, notably concentrated sulfuric acid and gibberellin, and the fact that only three species, *Cycas revoluta* Thunb., *Zamia integrifolia* L., and *Zamia furfuracea* L.f., were tested, and optimal times and concentrations would have to be determined for other species. A skilled nurseryman, taking proper safety precautions, could use the acid and gibberellin method satisfactorily, but it is not feasible for a hobbyist or collector that may only want to grow small quantities of each species, seed of which can cost upwards of $5 each. In fact, anecdotal evidence suggests even nurserymen were not as successful with the chemical methods and laboratory exactitude that were used in the published papers. Anyone wishing to germinate species other than *Z. integrifolia* and *C. revoluta* would have to determine chemical concentrations and exposure times to produce optimal germination rates.

Having heard anecdotal evidence of great success growing cycad seeds with a simple method requiring only readily available materials and simple procedures, I investigated the "baggie method" and found it successful and gratifying. Two cycad species were available to test for a report to this conference. Many hobbyists have been discouraged by low germination rates when attempting to grow costly cycads from seed. Low percentage germination, first and foremost, can be related to seed viability, but attempting to germinate cycad seed in greenhouses or shade houses under mist can result in high attrition of the percentage of seed that are viable due to insects, microorganisms, and seed pilfering by rodents. Because the method considered here allows seed to be kept in protected locations until planting, a higher success rate can be achieved. The method is equally attractive because of the amount of space, money, and expertise necessary to establish a mist system and attempt chemical seed treatments. Success could be instrumental in rekindling the desire of many people to germinate cycads.

MATERIALS AND METHODS

Seed of two cycads became available in time for this trial, *C. bifida* (Dyer) K.D. Hill, and *Cycas revoluta* Thunb. (king sago) × *C. taitungensis* (emperor sago). *Cycas bifida* (fork-leaved cycad), from China and Vietnam (Figures 1 and 2) is little known in cultivation and quite rare, but a friend and I successfully pollinated a female plant and produced seed (Figures 3-6). A few seeds were sacrificed to look for embryos, and they were visible but very small, suggesting that a maturation period was most likely necessary. I also performed the pollination of a *C. revoluta* plant with *C. taitungensis* pollen in late spring of 2014. Both parents of the hybrid are known to have immature embryos in seeds at the time female cones either dehisce or the abscission layer between seeds and the megasporophylls are fully developed and seeds may easily detach.

Figure 1. Generalized distribution of *Cycas bifida* in China and Vietnam [Image credit to Wikipedia Foundation©].

Figure 2. Mature female plant of *Cycas bifida.*

Figure 3. Unpollinated female cone of *Cycas bifida* (forefront).

Figure 4. Female *Cycas bifida* cone a few weeks after pollination.

Figure 5. Mature female cone of *Cycas bifida* prior to dehiscence.

Figure 6. Cleaned *Cycas bifida* seeds.

The sarcotestas of all seeds were removed, and cleaned seeds mixed with slightly moistened sphagnum peat moss, and then put into freezer bags (Figure 7) and sealed. In the case of *C. bifida*, seed coat removal was easy because the sarcotestas scrape off with very little effort. The *C. revoluta* × *C. taitungensis* seed required repeated soaking and whisking in wet coarse sand with a cordless drill fitted with a wire wheel, and washing. This process was repeated over the course of approximately two weeks to completely remove the sarcotestas. As mentioned earlier, removal of sarcotestas was done to completely eliminate: (1) any germination inhibitors that might be present as well as (2) fleshy seed coat material that could decompose, potentially infecting and killing viable seeds. The amount of water required to moisten the sphagnum peat moss was approximately equal to the weight of the unmoistened sphagnum peat moss. Some 259 *C. bifida* seeds were put into freezer bags on 20 November 2014, and kept at room temperature on a desk in my office.

Figure 7. Cleaned *Cycas bifida* seeds in freezer bag.

RESULTS AND DISCUSSION

The first sign of germination in the bags was noticed in mid-February (Figure 8). Germinated seeds were taken from the bags (Figure 9) and planted nine times during the 4 month period from 28 February to 27 June 2015 (Figure 10). Each time, any ungerminated seeds were placed back into the baggies, and planting was done again each time emerging roots were seen at the extremities of the baggies. After the June 27th planting, the few remaining seeds were judged inviable and discarded. Cumulative germination of this seed batch was approximately 95%, and no decomposing seeds were seen during plantings. No attrition due to insects, microorganisms or rodents was experienced. It is also worth mentioning that the baggies were not routinely opened throughout the length of any of these experiments, and this seems not to have stopped germination.

Figure 8. Germinating seeds of *Cycas bifida* in freezer bag.

Figure 9. Germinating seeds of *Cycas bifida* ready for planting.

Figure 10. *Cycas bifida* germinating seeds planted in tree pots.

Because this species is known to possess a strong taproot, germinating seeds were planted in well-drained mix (2-1-1 Fafard 2P-fine pine bark-sand) in deep tree pots (Figure 10). Germination was rapid (Figure 11) and seedling growth appeared brisk (Figure 12).

Figure 11. *Cycas bifida* seedling producing its first leaf.

Figure 12. *Cycas bifida* seedlings several months after planting.

The other *Cycas* experiment was begun much later and has not yet been concluded. For the purposes of this paper, germinating seeds were deliberately left in the freezer bags to see if they would be damaged by remaining unplanted (Figure 13). The seedling roots of *C. revoluta* × *C. taitungensis* were tangled, requiring patience and time to separate without damage (Figure 13). However, the unplanted, germinating seeds were in good health at the submission of this manuscript.

Figure 13. *Cycas revoluta* × *C. taitungensis* seeds germinating and becoming tangled in baggie.

CONCLUSIONS

The success of the "baggie method" in germinating cycad indicates that it is worth trying on any available cycad seed. It seems to be a worthwhile way to optimize the percentage of viable seed brought from cone abscission to successful establishment in individual containers, and should be considered by nurserymen and hobbyists alike.

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Efficiency through innovation©

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INTRODUCTION

Today, labor is the most premium product and there are four factors that affect the cost of labor:

- Salary
- Training
- Insurance
- Accidents/injuries

Any monetary savings on these factors will help lower costs overall. Savings in one factor may be transferred to another factor as an expense.

One of the ways to minimize labor cost is to mechanize wherever possible. Mechanization may help, but it is not always the savings everyone assumes. When considering mechanization, keep these thoughts in mind:

- Speed: it may not be as fast but will be easier on employees. Will faster production affect quality?
- Safety: will it prevent injuries or accidents? Do you have the manpower to safely use the equipment? Will you be looking at much higher training costs?
- Labor: can it be done with less people? Will adding people dramatically increase production?
- Equipment costs: how soon will the equipment pay for itself? Do you have the manpower to do maintenance and in-house repairs?

PLACES OR METHODS FOR INNOVATION OR EFFICIENCY

When looking for places or methods for innovation or efficiency there are what I consider five options to consider: production efficiency, crop innovation, customer needs, order assembly, and equipment and materials

Production efficiency

Production efficiency is what I consider for all stages of production that lead up to the sale of plants. It may involve crops or materials. There are several methods we incorporate for production efficiency.

- We chip all plant trimmings for mulch. Benefits we receive include: less watering and better weed control on larger containers. We also save on dumpster fees.
- Pre-filling and palletizing flats. We palletize our CP, 3 in., 4 in. and quart flats. We identify soil mixes by wrapping the flats with different colored plastic/Saran™ wrap. Pallets are brought to the production area as needed. Pallets are always available for the production crew, so the crew never has to wait for pots to be supplied.
- We will jump container sizes on certain crops, i.e. specific trees will be transplanted from tree tubes to 5-gal or 15-gal containers, depending on the species.
- We are constantly fine-tuning production methods, such as discontinuing B&B production.
- Palletizing used pots.
- New potting machines. Look at new machinery for different potting methods.
- Vertical production space for baskets in greenhouses.
- Signage in houses for production crews to pot numbers by visual inspection of space available.
- Cutting and seed indention templates.

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Crop innovation

Crop innovation is very limited for our type of operation. We grow 4-in. material all the way up to 200-gal trees. Due to the varied crops, we are limited on some of the equipment we can use.

- Trimming machinery options include block mowers on wheels, handheld trimmers with short fixed heads, heads on poles, and articulation heads on poles.
- Tree anchoring depends on your soil types and wind situation.
    - o Cable system on posts.
    - o Short "T" post, which are low cost, easy to install and remove.
    - o Mobile home anchor-cage with a modified tool for installing.
    - o Other options — guy wire anchoring for palms.
- Tree rotisserie — rotate the tree in windy areas for straight growth. This is used to rotate trees 180 degrees so that prevailing winds do not force the tree to lean in the container.
- Water Truck to water block edges and for dust control on the roads.

Customer needs

Designing goods and services for customers is critical to our business. We spend considerable time on crop selection and efficiency to better serve customers. The type of customer base you want to service can dictate what crops you grow and/or how you grow those crops.

- We break our customers down into four categories: retail, wholesale, landscape, municipal/government/non-profit.
- Look at municipal entities for plant giveaways. Municipal entities are a good guaranteed way to move selected crops. We grow large numbers of trees for heavy culling and what we do not use, we pick through and sell for tree giveaways. This opens the option for possible free advertising.
- Contract growing — most of our municipal/government sales are contract grown sales. This entails guaranteed sales and payment up front. You set the terms and in many cases can charge a premium price for oversized specifications. Working with architects directly allows your input at the beginning of the project.
- Retail customers need high quality plants that may require specialized shipping or sleeves. It can be dependent on weekend weather and is very seasonal. There is also the yearly change in demand for new or patented plants.
- Wholesale and landscape sales are what we specialize in. You need high quality plants, but they do not always require as much special treatment for shipment. Sales are usually year round due to continued construction and weather will not affect commercial jobs as much as retail sales. Most of your landscape sales are common types of plants rather than the new hot plant of the year.

Order assembly

Order Assembly is for shipping and loading. This includes:

- Rolling ramps for loading onto trailers, from one trailer to the next, or even to move large trees the length of a trailer.
- Pallet forks for Vermeer® Skid Steers for loading plants onto trucks.
- Tarping methods — every nursery has their own method and so far we have found there is no best method.
- Hoist for tarps to be loaded onto the truck after delivery.
- Palletizing orders for shipping is becoming a very common method along with rack carts. These methods work if you are shipping only one or two sizes of containers and plants.

Equipment and materials

This can include anything from purchasing high-dollar equipment to utilizing scrap materials — to enhance work efficiency.

- PTO driven finishing mowers for pot in pot operation. There is one less motor to maintain, since we can use existing tractors. In the process, we eliminated two Great Dane mowers and now complete the same amount of work in half the man-hours. Maintenance of the finishing mowers are strictly for belts and blades.
- Synthetic oil vs regular oil. We have doubled miles between oil changes and that has saved on filters and oil used. We have also experienced fewer problems on small engines with the synthetic oil.
- Fuel additives have reduced small engine problems from ethanol/biofuels mixtures in gasoline. Costs added to fuel expense is only $.02-$.03 per gallon. For more than three years, we have not had any burned out engines from ethanol-gas mixtures.
- We no longer use bulk fluids. It is cheaper to buy oil by the drum rather than in bulk. Common carriers or pick up trucks can deliver drums. Whereas, a shortage of drivers for transporting bulk fluids requires a tanker, increasing shipping costs.
- For storage of filters — we assign a storage bin for each piece of equipment and the list the filters needed on the bin.
- Our supplier checks our bins, reorders and restocks the bins. Hence, there is less labor for us to locate and order filters.
- Multi-use equipment — we use a semi bed that can haul semi trailers or gooseneck trailers. Flat beds allow for transport of up to four pallets of material.
- Vermeer Skid Steers are used to load into trailers, pull large plants out of the drip rows, move pallets, and load customers' trucks and trailers.
- We use a manure spreader system as a field potting machine for large containers. After seeing a few nurseries that used manure spreaders for pot-pot production, we created our own portable potting machine.
- Skid steers — there are many attachments for skid steers that will allow you to expand your equipment selection at minimal cost. Some examples are trenchers, post-hole diggers, brooms, concrete mixers, tree shears, etc. The list is endless.
- We use iPhones for driver-tracking capability ("Find My iPhone" app), instant photos of accidents, broken plants, delivery site issues, etc.
- iPads inable enable management to communicate and keep availability list for others to access. Group texting among all management personnel is a huge asset for us. Instant pictures can be sent to customers or office staff, along with live data entry for production and chemical application.
- The Evernotes app is used on IPads and IPhones. his includes sales history, equipment info, crew info, employee info, quotes, shipping schedules, job scheduling, etc. Evernotes allows database type searches on any and all info saved into the program with the ability to email or fax the data.
- Some of our low cost innovations include:
  - Spray paint container sizes on pots to avoid confusion with employees or customers.
  - Spray paint color code on pots for trunk height of palms or caliper of trees.
  - Pot hooks for 5-gal pots and hooks to pull larger containers.
  - PVC pipe to protect mist tubes on sprinklers and avoid sharp edges on tables for water or spray hoses.

Recycling used container media with solarization©

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INTRODUCTION

Underutilization of used potting media for crop production in environmental horticulture wastes money and resources. In conversations with growers, it is estimated that about 10% of plants with potting media are culled and disposed of in the industry. Many nurseries dump culled plants and media on site and this waste is generally not reused. In an effort to recycle this waste, a series of methods were tested to solarize the used potting media. Solarization is a sustainable, inexpensive, and effective method to reduce pathogens, nematodes, and weeds. Solarization works using the light energy of the sun and transforming it into heat. When temperatures reach a certain threshold over a certain critical time, pests can be eliminated. Different pests (i.e., weed seeds, insects, fungi) have different thresholds for being heat killed. Research has shown that if container medium is held at temperatures of 70°C (158°F) or higher for 30 min or 60°C (140°F) or higher for 1 h, solarization can completely eliminate plant pests (Stapleton et al., 2008). Methods have been developed for treating smaller quantities of medium such as in nursery pots on pallets with a "double tent" method (Stapleton, 2000). Research has shown the effective use of solarization to treat small bags of potting soil on benches (Zinati et al., 2002). What was lacking was a larger scale method to treat higher quantities of spent potting media.

MATERIALS AND METHODS

Solarization

To develop a large scale method, a series of small-scale solarization treatments were conducted to arrive at a final protocol to be scaled up (Steed, 2014). A 0.8 m^3 (1 ft^3) method was tried with different configurations using double tent methods, different medium depths, hydration rates, types of plastic, heights of spacing between plastic sheets, and materials used to suspend the top plastic sheet until a suitable method was developed. This final method consisted of a layer of ground cover that was first placed on bare ground to keep weeds from growing through the plastic. A layer of four mil, clear, polyethylene plastic was placed over the ground cover to prevent nematodes or disease pests from moving up through the soil to reinfest solarized medium. The area to be treated was 7.3×7.3 m (24×24 ft). Next, used potting medium [pine bark and peat, (3:1, v/v)] was moved from a nearby pile with a front end loader to the treatment area. The medium was spread over the plastic to a depth of 5.1 cm (2 in.) with shovels and rakes and large plant debris was removed by hand. The volume of medium treated was 2.7 m^3 (3.6 yd^3). Enough water was added to the medium to moisten but not saturate, since fully saturated medium does not conduct heat to the bottom well. This medium was then wrapped in clear 4-mil plastic and sealed tightly around the edges so that the plastic laid flat on top of the medium with the medium touching the plastic. A series of ridges and valleys were created using 5.1 cm (2 in.) galvanized pipes resting on stands above this plastic layer to slope rain water towards one edge. Polyvinyl chloride (PVC) pipes were originally tried but melted due to the extreme temperatures that are generated. A four mil, clear plastic sheet was placed over the pipes and wrapped tightly at the edges and underneath the bottom layer sheet and medium. The final configuration looked like a bag of soil within a bag (Figure 1). The solarization process for this study ran for 14 days and was started on 20 Aug. 2013. Three soil probes were located within the treatment medium. Samples of solarized and untreated soil were analyzed for physical and chemical attributes. Medium was collected and placed in seedling trays measuring 30.5×45.7×6 cm (1ft × 1.5 ft × 2.5 in.) for a weed germination comparison. Three trays of

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each of the following were filled with 2.5 cm (1 in.) of medium: solarized, untreated and new potting soil. Trays were placed in a high tunnel with 30% shade. Trays were watered daily with overhead irrigation and grown for 2 weeks. Weed seedling numbers were counted after 15 days of growth.

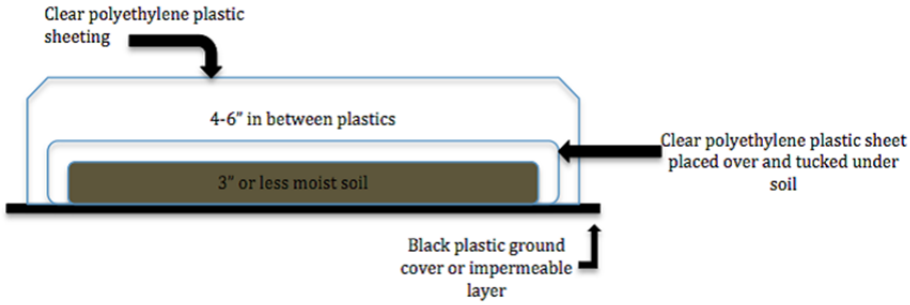

Figure 1. Schematic of solarization system. Illustration by Credit-Kallee Cook.

Growth study

Solarized medium was then tested in a growth study to find if there were any differences in producing plants or if the process might negatively affect plant growth (Steed, 2015). Fresh medium containing composted pine bark and peat (3:1, v/v) (Graco Fertilizer Co.) was used as the new medium comparison. Three different treatment soils were compared: 100% new soil as the control, and mixes of new and solarized soil at the proportions, respectively, 66:33 and 33:66. Treatments were replicated three times. The soil was added to #3 black plastic pots (9.5L). Time release fertilizer was added to the pots at 40 g of a 6 month time release, 14-5-11 (14N-2.1P-9.1K) with minors and 80 g of 12 month time release, 17-5-11 (17N-2.1P-9.1K) (Graco Fertilizer Co.). Rooted liners (60 cell) of *Viburnum suspensum* [10.2 cm (4 in.)] tall and *Lagerstroemia* 'Natchez' [25.4 cm (10 in.)] tall (ProGrowers, LLC, Plant City, Florida) were planted in pots on 30 Sept. 2013 and moved to the field. Irrigation was provided with overhead sprinklers. Weeds were hand-pulled. Plants were not pruned or staked during the growing season. The experimental design was a randomized complete block. Data was collected on 16 July 2014. Viburnum heights and widths were measured, while only 'Natchez' heights were measured. Means were compared with SAS JMP 11 Pro via Tukey's HSD test comparison.

RESULTS AND DISCUSSION

Solarization

The method developed worked exceedingly well with highest temperatures reaching 70.6°C (159°F), increasing the ambient outside temperature by 33°C (60°F). With this method, time and temperature thresholds were reached within 1 day to kill nematodes, plant pathogens, and most weed seeds. Most days exceeded the threshold unless there were afternoon rains for an extended time. This method demonstrates that medium can be sterilized and recycled with solarization at large capacities. It is only a matter of scaling-up to the size that can be effectively utilized. Our trials were done at a latitude of 36° 36' N, with daily temperatures that ranged between 20.7-33.6°C (69.3-92.5°F). It was not determined as to what range at higher latitudes the solarization would be effective, however, this could easily be tested in a small plot with the double tent method (Stapleton et al., 2000).

After 2 weeks in a greenhouse, the germination test of treated and untreated media produced some viable weed seeds compared to fresh, untreated soil. Three trays of newly purchased soil had zero weed germination. The solarized and non-solarized media averaged, respectively, 10.6 and 89 germinated weeds. Solarization reduced weeds by 88% compared to untreated, used media.

To enhance weed control an added step should be included which would hydrate used potting soil for about 14 days prior to solarization. Preferably, this should be done as a thin layer as in the solarization process. In fact, I recommend to prepare the soil for solarization then wet it for 2 weeks prior to wrapping the soil in plastic. This will allow for weed seeds to germinate prior to being solarized, thus eliminating weed seed that might be able to survive the heating process. Two small trials were done using this method with excellent results. All medium physical attributes did not change after treatment and soil chemical properties changed very little. Our test medium was a few years old, so fertilizer had long since been leached. This might not be the case if one uses fresh soil. Operationally, it appears that used medium will retain similar properties after the solarization process, except for pests. If using fresh soil with new, controlled release fertilizer — higher rates of nutrients can be released with elevated high temperatures.

Growth study

Among the different medium mix treatments 100:0, 66:33, and 33:66 (new soil: solarized soil) there were no significant statistical differences in viburnum height and width and 'Natchez' height (Table 1) (Steed, 2014). Hence, using solarized soil up to 66% of the soil mix caused no reduction of growth of these two woody plant species. This might not always prove to be the case depending upon the medium being used prior to solarization. Physical attributes of the medium are not greatly changed during the process of solarization so physical medium tests can be made on medium located in the pile to be treated. This will enable growers to determine the percentage of solarized soil that can be combined with new soil after the solarization process to grow plants. The on-farm cooperator used solarized soil and fresh soil (1:1, v/v) and had excellent results growing standard crapemrytle trees.

Table 1. Effects of large scale solarization on growth aspects of *Viburnum suspensum* and *Lagerstroemia* 'Natchez' (Steed, in press).

Treatment[1]	Measurement (in.)		
	Viburnum suspensum height	*Viburnum suspensum* width	*Lagerstroemia* 'Natchez' height
100:0	17.3 a[2]	29.5 a	66.1 a
66:33	21.1 a	26.9 a	66.8 a
33:66	19.9 a	24.9 a	65.6 a

[1]Proportion of new potting soil: solarized treated soil.

[2]Data are means calculated from three replications. Mean separation in columns by Tukey's HSD test, 5% level.

Cost of solarization

Costs for the large scale set up were $234 in materials and could be used for the entire solarizing season (Tables 2 and 3). Labor costs were $17 per solarization run, which included removing finished soil from the solarization pad. This also included costs of using a front-end loader at $65 h^{-1} as part of operating costs. Total costs per yard of soil was about $5 to treat used potting soil. This is a savings of about $39 m^{-3} ($30 yd^{-3}) of medium or about $108 per solarizing run with an estimated costs of $35 per yard used as the cost of fresh media. If soil was dumped directly into a solarizing pad material costs would break even in about 2.2 solarizing turns.

Table 2. Material costs for solarization project.

Materials	Costs ($)
3 plastic sheets	52.42
Pipes	150.00
Groundcover	31.20
Total	233.62

Table 3. Labor costs for solarization project.

Labor	Time (min)	Costs	Total vol.	Cost per yd^3
Tractor work \$65 h^{-1}	10	\$ 10.80		
hand labor \$10 h^{-1}	35	\$ 5.83		
		\$ 16.63	3.56 yds	\$4.67

A cable system could be used to suspend the top sheet of plastic, which would add an even greater cost savings to the system. In all likelihood, costs could probably be reduced to about \$3 m^{-3} (\$2 yd^{-3}) to treat media. We did not determine the longevity that the poly sheets could be reused with the system. We would be able get at least three turns of media and recoup the costs of all materials used each year.

MORE INFORMATION

To read more about this method here is a link to a factsheet. Methods and On-Farm Research Results 2013-2015. http://hillsborough.ifas.ufl.edu/documents/pdf/ornamental_production/A-Z_pubs/Soil_Solarization_Fact_Sheet.pdf

To watch a short presentation on the solarization process you can follow this link: http://hillsborough.ifas.ufl.edu/ornamental_production/videos.shtml.

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Rain gardens: understanding their benefits and their beauty©

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INTRODUCTION

Rain garden systems are one of the most commonly utilized stormwater control measures (SCMs) to capture and remove pollutants [such as nitrogen (N), phosphorus (P), zinc (Zn), copper (Cu), cadmium (Cd), lead (Pb), and total suspended solids (TSS)] from stormwater runoff (Davis et al., 2001, 2009; Hunt et al., 2012). They are constructed by excavating the existing soil within the landscape and refilled with 0.7-1 m of a sand/soil/organic matter engineered filter bed substrate (Davis et al., 2009). They are then planted with vegetation (Liu et al., 2014; NCDENR, 2009). Rain gardens can be placed in many different landscape scenarios. They function well for containing and remediating polluted stormwater runoff because of their two main components: (1) the engineered filter bed substrate (EFBS) and (2) the vegetation.

An EFBS has to have an appropriate infiltration rate (speed that water enters the EFBS) and saturated hydraulic conductivity (speed that water moves through the saturated EFBS) so that water can be conveyed through the system appropriately. Both infiltration and saturated hydraulic conductivity can be impacted by the surrounding (native) soil, which will impact exfiltration out of the rain garden, as well as by the different substrate components utilized and will change with time. Sand-based EFBSs [85-88% (by volume) sand, 8-12% (by volume) fines (silt and clay), and 3-5% (by volume) organic matter] are commonly recommended due to their suitable hydraulic conductivity and permeability (Hsieh and Davis, 2005; NCDENR, 2009). However, slate-based (MS-16 100% expanded slate, Permatill, Carolina Stalite Company, Salisbury, North Carolina) EFBSs have been shown to convey water well and may be a better choice for small rain gardens with high inflow volumes due to their higher infiltration and saturated hydraulic conductivity rates (Turk et al., 2014). Paus et al. (2014) found that the saturated hydraulic conductivity of rain gardens with either a sandy loam or a sand EFBS tended to increase with time near the surface of the system, possibly due to vegetation maturation, bulk density reduction, and freeze thaw cycles.

Engineered filter bed substrates also need to have binding potential for pollutant remediation. Hunt et al. (2008) reported that a rain garden with a loamy sand EFBS capturing runoff from an asphalt parking lot had effluent concentrations of total N, total Kjeldahl N, and NH_4-N that were 32.2, 44.3, and 72.3% lower than that of the influent concentrations. Also, total P in the effluent was reported to be 31.4% lower than that of the influent (Hunt et al., 2008). Turk et al. (2014) reported that a slate-based EFBS had better remediation of N (86% initially and 99% by the end of the 18 month study) than the sand-based EFBS. These researchers also reported that slate and sand had good P removal, 99% and 96% respectively (Turk et al., 2014). Aged pine bark (PB) is often used as the organic matter source in EFBSs; however, compost utilization as an organic matter source may provide many benefits, such as plant growth enhancement from nutrients, pollutant binding, and microbial support.

Arrangement of EFBS components within a rain garden system can also improve runoff retention and remediation. Layering of varying EFBS components can cause a saturated anaerobic zone within the rain garden system as shown by Hsieh et al. (2007b). An anaerobic zone within a rain garden system can promote the loss of N by the process of denitrification (Tiedje et al., 1984). A permeable sand layer over a less permeable soil layer increased stormwater retention within the EFBS and allowed nitrification in the well-

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aerated sand portion of the substrate and denitrification in the saturated, low permeable soil layer (Hsieh et al., 2007b). The less permeable bottom soil layer also increased contact time between dissolved P and the EFBS resulting in more effective total P removal (Hsieh et al., 2007a). Palmer et al. (2013) reported that utilizing a saturation zone within the rain garden system greatly reduced NO_3^- in effluent (71% compared to 33% without a saturated zone) when the EFBSs consisted of a 60% sand, 15% compost, 15% finely shredded cedar bark, and 10% aluminum-based drinking water treatment residuals mix. While the same was not true for O-PO_4, which was remediated better without a saturation zone (80%) compared to with a saturation zone (67%) (Palmer et al., 2013). However, the anaerobic zone needs to be located near the bottom of the rain garden system to prevent detrimental effects on plants such as root stress from anoxia or favorable environment created for root pathogens (Tiedje et al., 1984).

Vegetation in rain gardens can also have a positive impact on remediation and has been reported to improve the remediation of N and P from simulated polluted stormwater when compared to non-vegetated rain gardens (Read et al., 2008; Bratieres et al., 2008). Turk et al. (2014) reported that 176 days after planting plant uptake appeared to have a greater impact on remediation than EFBS composition. Gautam and Greenway (2014) grew a variety of Australian species in gravel, loam, and sand EFBSs. These researchers found that plants with the faster growth rates and larger biomass production retained greater amounts of nutrients in their roots and above ground structures (Gautam and Greenway, 2014). Plant parts accounted for 2.7-4.3% of the total P and 8.7-17.7% of the total N retained in the rain garden system (Gautam and Greenway, 2014).

Care should be taken when selecting plants to insure survival and functionality within the rain garden. Plants growing in rain gardens face two challenges: low nutrient levels in the influent (compared to typical fertility programs) and periodic drought conditions. The average total N ranged from 1.13 to 2.19 mg L^{-1} and average total P ranged from 0.07 to 0.33 mg L^{-1} for stormwater runoff from eight asphalt parking lots in North Carolina (Passeport and Hunt, 2009). These N and P concentrations are much lower than the N (50 to 100 mg L^{-1}) and P (10 to 15 mg L^{-1}) rates recommended for application during containerized nursery production (Bilderback et al., 2013). As rain gardens are non-irrigated landscape features, plants (within a rain garden system) need to be able to tolerate extended periods between rainfall while maintaining aesthetic appearance and maintaining transpiration. Several species have been evaluated and have proven to grow well and be aesthetically pleasing (Table 1). Vegetation in rain gardens also must be able to return water back to the hydrologic cycle through evapotranspiration (ET). Evapotranspiration is the process where water in the soil-plant system is transferred to the atmosphere and it includes both evaporation from the surface of the soil and transpiration from plant canopies (Hillel, 2004). The process of ET is critical in meeting long-term hydrology goals with rain gardens (Hunt et al., 2012). Low ET rates impact the water within and the water table below the rain garden system (Hunt et al., 2006). Increased ET from rain garden systems, may be achieved by utilizing types of vegetation that have long root systems increasing opportunity for storage by the media and for vegetation to take up water in between events (Hunt et al., 2012).

CONCLUSIONS

The EFBS, in combination with the appropriate vegetation make rain gardens functional and efficient at remediating pollutants and controlling volumes from polluted stormwater runoff. There are many different pollutants of concern and many different ways that rain gardens can be incorporated into the landscape. Plantings within rain gardens can be arranged so that they can divert and slow surface flow for filtration of sediments (Davis et al., 2009). Also, the plantings within a rain garden can be arranged so that they are aesthetically pleasing and support wildlife. Within the environment of a rain garden plant roots can aid in supporting the microbiological populations that may aid in degradation of pollutants and they should help in media permeability (Davis et al., 2009). Also, in order to most efficiently remediate pollutants and control the volume of polluted stormwater runoff, the size of impervious surface and the pollutants of concern, as well as the EFBS

composition, need to be thought of beforehand (Hunt et al., 2012; Riley et al., 2013; Turk et al., 2014).

Table 1. List of species that have been evaluated in rain gardens and have worked successfully.

Scientific name	Common name	Reference
Betula nigra	River birch	Turk et al., 2014
Betula nigra 'Duraheat'	River birch	Turk et al., 2014
Eutrochium maculatum 'Gateway' (syn. *Eupatorium purpureum* subsp. *maculatum* 'Gateway')	Joe-pye weed	Turk et al., 2014
Helianthus angustifolius	Swamp sunflower	Turk et al., 2014
Helianthus angustifolius 'First Light'	Swamp sunflower	Turk et al., 2014
Itea virginica	Virginia sweetspire	Turk et al., 2014
Itea virginica 'Henry's Garnet'	Virginia sweetspire	Turk et al., 2014
Juncus effusus	Common rush	Turk et al., 2014
Monarda fistulosa	Beebalm	Riley et al., 2013
Magnolia virginiana	Sweetbay magnolia	Turk et al., 2014
Magnolia virginiana 'Sweet Thing'	Sweetbay magnolia	Turk et al., 2014
Panicum virgatum	Switchgrass	Turk et al., 2014
Panicum virgatum 'Shenandoah'	Switchgrass	Turk et al., 2014; Riley et al., 2013

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How newcomers and millennials will succeed in the green industry©

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CHALLENGES IN THE NURSERY INDUSTRY

How will students, newcomers, and the young at heart succeed in the green industry? All that is required is to shift your mindset and proactively solve problems and seize opportunities. I will explain how successful men and women will grow in their careers and businesses in the green industry.

Starting with the major challenges before us — we produce live, green, perishable goods. Some people even think of us as luxury. I am one of them, but "luxury" is not a dirty word. Consider the multi-million dollar pet industry. In 2010, Americans spent $47.7 billion on pet products and services. In 2008, total U.S. sales in the green industry was $176 billion http://horttech.ashspublications.org/content/21/5/628.full.pdf+html. Yet, we are classified as a maturing industry. The green industry is in a period of hyper competition http://aggie-horticulture.tamu.edu/faculty/hall/publications/2010%2008%20Making%20Cents%20of%20Green%20Industry.pdf, and we are in a race to the bottom in the nursery business — competing on price. Academic opportunities and research funding can be scarce.

The nursery industry is also highly manual-labor intensive. Live goods are capital intensive. How are we ever going to overcome these tremendous challenges? I see opportunity, possibly more than ever before. The first and most important step is to change your mind set. The problems I described are framed by scarcity. We must change our mind set from scarcity to abundance. We are not going to solve these tough problems and create new opportunities without changing how we think.

DEFINING SUCCESS

How do you define success? It is a vague term and everyone has a different definition. I define success in terms of freedom and having creative control of my time. For green industry professionals, I define success as getting better every day. Your definition will be completely different and can change over time. It is important that you know what you want so that you can overcome adversity.

We are not going to solve our problems and create opportunities without trial and error. If you are new or young at heart, you will make mistakes. Losses are lessons, just try to not make the same mistake twice!

THE PARETO PRINCIPLE (80/20 ANALYSIS)

There are many problems to solve, so how do you focus and create success? Apply 80/20 analysis to your business, or career http://betterexplained.com/articles/understanding-the-pareto-principle-the-8020-rule/. Whether it is propagation, production, or research, 80% of your results are generated from 20% of your actions.

Now apply 80/20 to 80/20. The result is 64/4. Four percent of what you do produces 64% of the results! I believe the timing of when you take action is the 4%.

Several years ago I focused on improving irrigation timing and fertilizer rates. We were able to grow *Betula nigra* 'Cully', Heritage® river birch 100% faster by increasing the fertilizer rate per cubic yard and reducing the irrigation duration. That eliminated a year of crop production and it increased our growing area. It was like we just did a 300 socket expansion. Most importantly, the customer received a superior product and we became more profitable with less effort and inputs.

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FINDING YOUR STRENGTHS

Now that we know about the Pareto Principle (80/20 Analysis), what do we do with all this free time? Well what are you good at? I recommend everyone take a personality test if you have not already done so. I like *Strengths Finder 2.0* by Tom Rath (ISBN: 9781595620156). In 30 min you will know what your top five strengths are. Play to your strengths and create more success with your thoughts and actions.

MAKE A CHECK LIST

You might be thinking…but I am so busy already! I challenge you to replace yourself in your current situation. How do you do this? You create systems in your business and career. Start documenting the routine tasks you do. Make a checklist. Keep it to yourself for now. I took my irrigation job and formed it into a system. Now two other people are capable of checking irrigation daily and I focus on something new and more important.

You will do more in less time using a checklist or system. You will be able to incorporate more split testing. Test A is your control. It is how you grow and work now — your benchmark. Test B manipulates a variable, with the goal of doing better than your control. Once you have your work system you can start asking yourself: WHY are we doing this anyway? Does it make sense?

Most processes and work flows at Mountain Creek Nursery exist today because it was always been done this way. What does that mean? Well Mountain Creek Nursery was a soil-grown tree B&B nursery prior to its current container tree focus. Just because you pruned a certain way or harvested this way in the field does not translate exactly to the container yard. Start testing ideas and assumptions. In the tree business we get one crop turn a year. You must test assumptions. Your competitors are testing every day — and your nursery is getting farther and farther behind!

SERVICE-BASED BUSINESS

Now I want to focus on service-based businesses. The service based business has the opportunity for recurring revenue or income. Newspapers and magazines have been using the subscription model for decades. Anyone can market and sell information on the internet. You do not have to be picked anymore. Choose yourself and get started if you have something valuable to say or share.

ENTREPRENEURSHIP

Finally, I believe we are about to enter a golden age of entrepreneurship. Businesses are created when you get one customer. Test your idea by offering it to customers. Do not spend too much money creating a product or service nobody wants. Let your income lead your expenses and do not quit your day job until you reach 50% of your monthly expenses.

The best information and advice in the world is meaningless without taking action. Identify your challenges and apply your strengths and 80/20 analysis. Test ideas in your systems and remember: timing is critical to your success.

Can you change your mind set and continue to succeed? Yes, of course. Will you change how you think? I look forward to hearing about your success at next year's meeting!

Update on crapymyrtle bark scale©

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INTRODUCTION

Crapemyrtle (*Lagerstroemia*) is a $46M (farmgate wholesale value) crop. It is the number one deciduous flowering tree in the nursery trade. In the summer, crapemyrtle is one of the main flowering trees and flowers for 2-3 months. Except for minor problems such as crapemyrtle aphids and powdery mildew, the plant has been generally considered as low maintenance, and used extensively in landscape in the Southeastern United States and other regions. A new, highly unsightly pest, the crapemyrtle bark scale (CMBS), threatens to change the low maintenance reputation of this plant. The crapemyrtle bark scale (*Eriococcus lagerstroemia*) is a felt scale (*Coccoidea*: *Eriococcidae*) identified in 2014 through DNA work and morphological studies. It was first observed in 2004 in Richardson, Texas, a suburb of Dallas.

DESCRIPTION OF THE PROBLEM

In its native range in China, CMBS has been observed as north as Liaoning, Shanxi, Hebei and Beijing and as south as Sichuan, Jiangsu, Zhejiang, Guizhong, and Guangdong (Jiang and Xu, 1998; Luo et al., 2000; Chen and Zhang, 2011). There are as many as four generations per year for CMBS. In Guiyang, Guizhou Province, immature crawlers could be observed on branches in March before plants leaf out. The CMBS population fluctuates throughout the year, but the peak of nymph was observed in August and peak of pupae in June (Luo et al., 2000). In addition to crapemyrtle, pomegranate is also a host to CMBS.

Since its first sighting in 2004 in Texas, CMBS has been reported in 10 other states including Oklahoma, Arkansas, Louisiana, Tennessee, New Mexico, Georgia, Alabama, Mississippi, South Carolina, and Virginia. Sighting could be reported here https://www.eddmaps.org/cmbs/. At the end of October 2015, there were 85 county and 27 specific location reports (Figure 1).

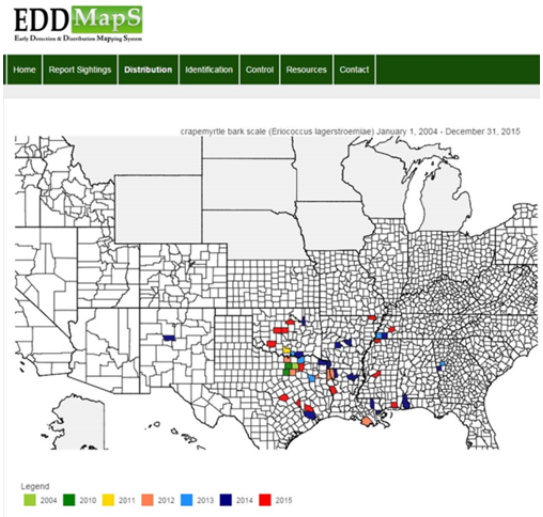

Figure 1. Reporting of crapemyrtle bark scale on early detection and distribution mapping system.

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Infested crapemyrtle trees often harbor overlapping generations of scale insects, including all life stages from eggs to adults (Figure 2). A good confirmation of CMBS presence is the "pink blood" (Figure 3) that oozes from the scale when crushed. Adult female scales look superficially similar to mealybugs, being white and fuzzy. Mature females are 2-3 mm long and can be found from the youngest shoot tips to the base of trunks (Figure 4), and frequently under peeling bark, a common trait of many crapemyrtle trees. The adult male scale has wings and may not be present. Scales produce honeydew which leads to growth of black sooty mold on the bark. Under severe infestations whole plants can be covered by black sooty mold, reducing the appearance quality of the plants (Figure 5). Overall impacts on plant health by CMBS have not been measured, but it appears that flower size and number are reduced with heavy infestations. Plants may leaf out later in the spring, and dieback of branches and entire plants has been observed on occasion.

Figure 2. Different stages of crapemyrtle bark scale found on this twig.

Figure 3. Infestation of crapemyrtle bark scale can be confirmed by the "pink blood" when crushed.

Figure 4. Crapemyrtle bark scale could be found any part of the trunk of a crapemyrtle.

Figure 5. Black sooty mold caused by crapemyrtle bark scale infestation was covering all parts of the crapemyrtle.

Adult female CMBS do not have wings, so long distance spread is thought to be due to the transportation of infested plants. Short distance spread could be due to wind, rain, birds, squirrels or ants. Ants have been observed on many trees when CMBS is present (Figure 6). For some scales, ants play a role in moving scales to fresh locations within a plant or to new plants.

Figure 6. Ants are often found at crapemyrtle infestation sites.

Crapemyrtle plants should normally be planted in full sun conditions. Observations suggest that levels of CMBS infestation may be correlated to shade levels, if other conditions were similar. This may provide additional support to the full-sun-planting recommendation for crapemyrtle plants.

Crapemyrtle bark scale could overwinter on trees, in forms of nymphs, pupae and adults. Activities of CMBS were seen as early as in February in Arkansas and Texas. Peak of crawler activity is has been seen in May, with additional peaks in March, June, July and August.

Natural enemies may have a significant impact on CMBS activity. We have observed formerly heavily infested plants with almost no trace of scale activity following high populations of predatory lady beetles. The two most common natural enemies we have observed include twice-stabbed ladybeetle (*Chilocorus cacti*) and *Hyperaspis* sp. No research has been conducted on using natural enemies to control CMBS in the United States.

Management of CMBS could also involve careful cultivar selection and use of chemicals. High infestation has been observed on 'Tuscarora', 'Lipan', 'Pink Ruffles', 'Tuskegee', 'Acoma', 'Velma', 'Choctaw' and 'New Orleans'. Neinicotinoids are effective in controlling CMBS when used as soil drench before the peak activity in May. However, bees are attracted to crapemyrtle pollen at certain times of year. Thus foliar application may not be a desirable option when crapemyrtles are in bloom. Physically removing or power-washing infested branches may reduce insect pressure, however the value of this tactic has not been evaluated. Generally speaking, a holistic management strategy for CMBS has not yet developed and is needed.

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Developing a risk assessment tool for evaluating potential invasiveness of ornamental plants©

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INTRODUCTION

This article summarizes *PRE Model Research*, published by PLOS ONE, March 2015, and led by C. Conser[1], L. Seebacher[2], D.W. Fujino[3], S. Reichard[4], J.M. DiTomaso[1] ([1]Department of Plant Sciences, University of California Davis, Davis, California, USA; [2]Washington State Department of Ecology, Lacey, Washington, USA; [3]University of California, Davis, Center for Urban Horticulture, Davis, California, USA; [4]University of Washington Botanic Gardens, Seattle, Washington, USA).

The nursery and landscape industry has introduced over 50,000 ornamental species in the United States (Gordon and Gantz, 2008). The total number of cultivars introduced increased from 29,000 in 1987 to 105,000 in 2008 (Levine and D'Antonio, 2003). Most of these species and cultivars do not cause environmental or economic problems. In fact, only a small percentage (between 0.1 and 1%) has become invasive.

However, of the species that are invasive in the USA, many originated from the horticultural industry. For example, in California, 60% of the 214 invasive plants impacting wildlands were intentionally introduced for human uses, and 47% of those plants are landscape ornamentals (Cal-IPC, 2014). Throughout North America, 82% of the 235 invasive woody plants are horticultural in origin (Reichard and Hamilton, 1997) and in the estimates of invasives originating from the nursery industry range from 34 to 83% (Bell et al., 2003).

The most cost effective way to avoid establishment of new invasive ornamental plants is to prevent their introduction at the beginning of the nursery supply chain. This can be achieved through risk assessment tools. Weed Risk Assessment (WRA) is a systematic process that uses available evidence to estimate the risk of a plant species becoming invasive in a given region. While there are many WRA tools that have been developed for a variety of applications, including evaluating plants in botanical gardens, none were specifically designed to screen ornamental plants prior to being released into the environment.

The most widely used WRA tool was developed in Australia (Pheloung et al., 1999) for import screening purposes, and has since been adapted for use in other parts of the world. The tool consists of 49 questions. It has been shown to be 90 to 100% accurate in correctly identifying invasive plants, but results varied dramatically from 21 to 75% accuracy in categorizing known, non-invasive plants. As a result, the tool is considered by the horticultural industry to be too conservative in predicting invasiveness, with far too many non-invasive species categorized as invasive. This will likely reduce its practical application by the industry.

The United States (US) also has a WRA tool used by USDA-APHIS to prevent the importation of invasive plants (Koop et al., 2011). Unlike the Australian WRA, this tool has high accuracy in classifying both major-invaders (94% accuracy) and non-invaders (97% accuracy), but it is not designed for evaluating potential invasiveness on a regional scale or for determining invasive risk with plants in the early pre-marketing stages.

For the nursery and landscape industry to consider a WRA tool useful, it must be: highly accurate in predicting potential invasiveness and non-invasiveness, easy to use, and not require a long period to complete the assessment process. Thus, we initiated a project using a science-based and systematic process to develop a highly accurate (for both invasive and non-invasive plants) Plant Risk Evaluation (PRE) tool specifically for screening

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ornamental plants.

MATERIALS AND METHODS

We assessed questions from existing WRA tools and developed the PRE tool with the most predictive and statistically relevant questions for ornamental plants. The ultimate goal of this project is to provide the horticultural industry with a voluntary screening tool that prevents new, high-risk plants from being introduced or sold in regions where the plants are likely to become invasive.

The initial step in developing the PRE tool required an evaluation of several existing WRA screening tools to determine the most appropriate and highly predictive questions, contributing to model accuracy for ornamental plants. From the various tools available we identified 56 questions that were commonly used to evaluate a set of known invasive and known non-invasive plants. These questions covered invasive history, climate match, difficulty of control, environmental impacts, reproductive and dispersal strategies, and growth rate.

Using the 56 questions, we evaluated a total of 35 plants, 21 known invasive and 14 known non-invasive plants. The invasive plants were selected from the California Invasive Plant Council's (Cal-IPC) Invasive Plant Inventory and the non-invasive species were chosen from the Plant Right's Suggested Alternatives for Invasive Garden Plants (PlantRight, 2014). As many questions as possible were answered using available literature, as well as searches of online databases and species' fact sheets.

For each plant species evaluated, we calculated the total score and the percentage of questions that were answered. To determine which questions contributed most to the predictability of invasiveness and non-invasiveness, we used a two-tailed Fischer's Exact Test, which statistically compared the answers for each question between the known invasive and non-invasive species. In addition, we calculated the percentage of times each question was answered for all known invasive and non-invasive plants. The scores for known invasive plants ranged from 21 to 44, with an average score of 31. The scores for known non-invasive plants ranged from 5 to 14, with an average score of 10. For each plant species screened, the percentage of questions answered for known invasive plants ranged from to 80% to 98%, with an average of 90%. The percentage of questions answered for known non-invasive plants ranged from to 86 to 95%, with an average of 89%.

The Fischer's Exact Test identified a total of 31 questions that had a greater than 95% probability of separating invasive from non-invasive species. The percentage of times each of the 56 questions was answered for known invasive plants ranged from 5 to 100%. The percentage of times each of the 56 questions was answered for known non-invasive plants ranged from to 0 to 100%. Of the 56 questions evaluated, 17 were eliminated because they did not provide statistically significant predictive power to separate known invasive from known non-invasive plants. Other questions were eliminated because they could not be answered at a high enough frequency (only 0 to 19%), they were irrelevant to evaluating ornamental plants or new plant introductions (mostly environmental impact related questions), or the question was inherently biased. For example, the question was only known and answered when the phenomenon was studied, which was nearly always with known invasive species (i.e., allelopathy, palatability to animals, impacts on grazing).

After removing or merging questions, we were left with a PRE tool that contained 19 questions (Table 1). We tested the 19-question PRE tool by screening 94 additional plants, 57 known invasive and 37 known non-invasive plants. Similar to the 56 original questions, we used a two-tailed Fischer's Exact Test to compare the predictability of each question and calculated the number and percentage of times each question was answered. From the analysis, 16 of the 19 questions showed statistical significance between the known invasive and known non-invasive species.

Similar to the same questions in the 56-question evaluation, each question was answered at a high frequency, ranging from a low of 54% for non-invasive plants to 100% for most other questions. An average of 97% of the questions were answered for both invasive and non-invasive plants for the 94 species evaluated. For individual species, this

ranged from 85 to 100% of the questions answered.

Table 1. PRE tool questions and their statistical predictability in separating known invasive and non-invasive species. Fisher's Exact Test compared the 57 invasive species against the 37 non-invasive species for each question. Percent of each question (Q) answered is also included. Brackets after question indicate citation were question is included in WRA model.

Question	Question in PRE tool	Fisher's exact test (2-tail)	% Q was answered for invasive plants	% Q was answered for non-invasive plants	Point values Yes/No
1	Has the species become naturalized where it is not native (Koop, et al., 2011; Pheloung et al., 1999; Brunel et al., 2010; Caley and Kuhnert, 2006)?	*P*<0.0001*	100	100	1/0
2	Is the species noted as being invasive elsewhere in the US or world in a similar climate? (Reichard and White, 2001; Koop, et al., 2011; Virtue et al., 2008; Caley and Kuhnert, 2006)?	*P*<0.0001*	100	100	2/0
3	Is the species noted as being invasive elsewhere in the US or world in a similar climate (Reichard and White, 2001; Koop, et al., 2011; Virtue et al., 2008; Brunel et al., 2010; Caley and Kuhnert, 2006)?	*P*<0.0001*	100	100	3/0
4	Are other species of the same genus invasive in other areas with a similar climate (Reichard and White, 2001; Koop, et al., 2011; Virtue et al., 2008; Caley and Kuhnert, 2006)?	*P*<0.0001*	100	100	1/0
5	Is the species found predominately in a climate that matches those within the region of introduction (Koop, et al., 2011; Pheloung et al., 1999; Brunel et al., 2010)?	-	96	100	2/0
6	Dominates in areas this species has already invaded (displaces natives) (Koop, et al., 2011; Virtue et al., 2008; Brunel et al., 2010; Caley and Kuhnert, 2006). Can overtop and/or smother surrounding vegetation (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999; Caley and Kuhnert, 2006).	*P*<0.0001*	100	100	1/0
7	Is the plant noted as being highly flammable and/or promotes fire and/or changes fire regimes (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999; Caley and Kuhnert, 2006)?	*P*<0.0001*	79	97	1/0
8	Is the plant a health risk to humans or animals/fish (Toxic tendencies) (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999; Brunel et al., 2010; Caley and Kuhnert, 2006)? Has the species been noted as impacting agricultural/grazing systems (Koop, et al., 2011; Pheloung et al., 1999; Brunel et al., 2010)?	*P*=0.0001*	100	100	1/0

Table 1. Continued.

Question	Question in PRE tool	Fisher's exact test (2-tail)	% Q was answered for invasive plants	% Q was answered for non-invasive plants	Point values Yes/No
9	Does the plant produce impenetrable thickets, blocking or slowing movement (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999; Caley and Kuhnert, 2006)?	*P*=0.0002*	93	100	1/0
10	Reproduces vegetatively via root sprouts/suckers (Reichard and White, 2001; Pheloung et al., 1999) or stem/trunk sprouts/coppicing (Reichard and White, 2001; Koop, et al., 2011).	*P*=0.0314*	98	100	1/0
11	Plant fragments are capable of producing new plants (Reichard and White, 2001; Pheloung et al., 1999).	*P*=0.0002*	100	100	1/0
12	Does the plant produce viable seed?	*P* =0.0001*	100	100	1/0
13	Produces copious viable seeds each year (>1000) (Reichard and White, 2001; Pheloung et al., 1999).	*P*<0.0001*	86	78	1/0
14	Seeds quick to germinate (Reichard and White, 2001; Pheloung et al., 1999).	*P*=0.1296	75	68	1/0
15	Short juvenile period. Produces seeds in first 3 years (herbaceous) or produces seeds in first five years (woody) (Reichard and White, 2001; Pheloung et al., 1999).	*P*=0.0078*	89	54	1/0
16	Long flowering period with seeds produced for more than 3 months each year (Reichard and White, 2001; Pheloung et al., 1999).	*P*=0.2320	86	86	1/0
17	Propagules dispersed by mammals/insects or birds or via domestic animals (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999).	*P*<0.0001*	100	97	1/0
18	Propagules dispersed by wind or water (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999).	*P*<0.0001*	98	97	1/0
19	Propagules dispersed via agriculture, contaminated seed, farm equipment, vehicles or boats, or clothing/shoes (Koop, et al., 2011; Virtue et al., 2008; Pheloung et al., 1999; Caley and Kuhnert, 2006).	*P*<0.0001*	100	94	1/0
		Average	97	97	Range of 23/0

RESULTS

The results showed scores for known invasive plants ranging from 12 to 21, while the scores for known non-invasive plants ranging from 2 to 13. Based on the separation in scores among the known invasive and non-invasive species, the scoring scale for the 19-question PRE tool was established to be: <11 as an "Accept" (low invasive risk); 11 to 13 as "Evaluate Further"; and, >13 as a "Reject" (high invasive risk) (Figure 1). Plants which fell into the "evaluate further" category may need additional assessment by an expert panel.

For the 57 known invasive plants evaluated through the 19-question PRE tool, no species were classified as accept. When species within the "evaluate further" category were excluded, the accuracy of the PRE tool in prediction invasiveness was 100%.

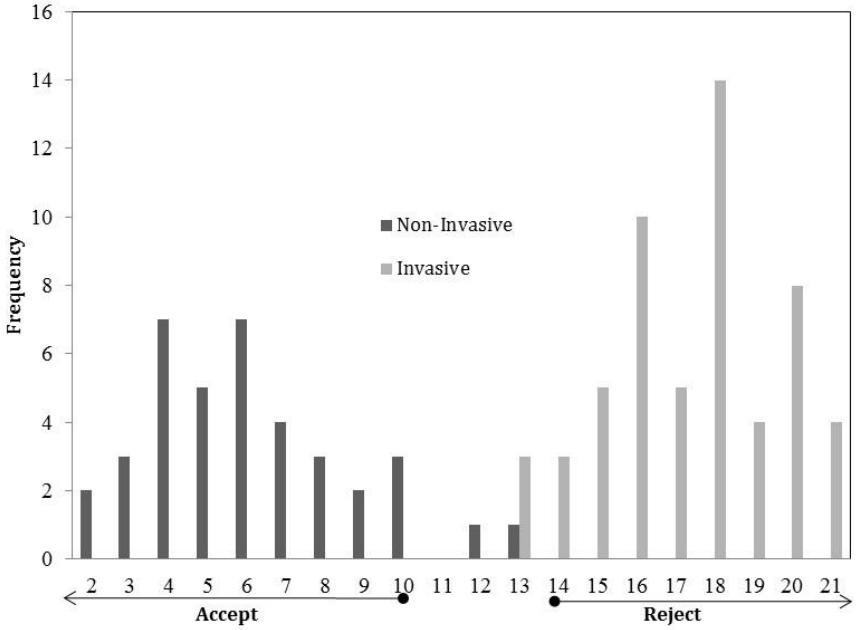

Figure 1. Histogram of scoring frequencies for 19-question PlantRight (PRE) tool. Scores: Invasive >13; Non-invasive <11; Evaluate Further = 11-13.

Even when the four species listed as "evaluate further" were considered false positives (invasive species incorrectly accepted as non-invasive) the accuracy and sensitivity was 93%. For the non-invasive species, the 19-question PRE tool gave no false negatives (non-invasive species rejected as invasive), but the tool did classify one species in the "evaluate further" category. Thus, the percent accuracy of the model when plants classified as "evaluate further" are excluded is 100%. Even when the "evaluate further" species are considered as false negatives, the accuracy is still a very high 97%.

When considering both known invasive and non-invasive species, the overall accuracy of the PRE tool model was 100% when "evaluate further " species were excluded and 95% when they were included.

NEXT STEPS

The next steps in the development and validation of the PlantRight PRE tool will be to: 1) test the consistency of the tool by different users (industry, academia, and conservation); 2) test the accuracy of the tool in evaluating invasive risk on a national scale (to demonstrate that it can be used beyond California, and at different scales); 3) incorporate climate matching capabilities; 4) develop an online PRE tool and database (https://pre.ice.ucdavis.edu) in partnership with UC Davis; and, 5) encourage voluntary nursery industry adoption. The ultimate goal of our PRE efforts is to equip members of the horticultural industry with a practical screening tool to prevent potentially high-risk plants from being introduced or sold in regions where they are likely to become invasive.

CONCLUSION

The PRE tool can be used preventatively by the nursery industry to screen ornamental plants for potential invasiveness prior to introduction to the marketplace. PRE can also predict the risk of invasiveness (low or high) for a given species or cultivar in a designated region.

The tool is expected to provide the industry with a variety of benefits, including: 1) a practical, efficient tool to accurately assess invasive risk, by region, early in the evaluation process (before making a significant economic investment); 2) a decision support tool to stay ahead of local and/or regional regulatory threats; 3) additional information regarding

taxonomy, reproductive characteristics, culinary and medicinal uses; and, 4) optional services including an online PRE database (tiered access and password protected), and access to maps of climate-matching results under various assumptions (e.g., drought tolerance) and scenarios (e.g., irrigation, climate change).

Because invasive plants represent only a small percentage of the horticultural inventory (~1%), screening plants for invasive qualities should not present a major economic hardship to the industry. Pre-screening of potential introductions would be expected to categorize the vast majority of species as possessing low (or no) invasive risk, while identifying relatively few as having a high probability of becoming invasive.

More importantly, because development of new cultivars represents a significant economic investment for nursery growers throughout the US, pre-screening would prevent nurseries from spending important research dollars to develop new cultivars with high invasive potential. Rather, the tool could help industry promote exclusively non-invasive plants in regional markets.

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Mulch type affects degradation and weed control potential in container production©

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SIGNIFICANCE TO THE INDUSTRY

Weed control practices in container production primarily consist of two methods, hand pulling and herbicide applications, but these are not ideal for larger container production. Mulches have been proven to be an effective alternative method of weed control in large containers (Richardson et al., 2008; Bartley et al., 2014). Due to the abundance of fertilizer and water in the nursery environment, degradation rates of available mulch species and types could drastically vary (Altland and Krause, 2014). This research, conducted with the use of litter bags, shows that of five readily available mulch species, pine bark mini-nuggets, Eastern red cedar, and loblolly pine followed by sweetgum and Chinese privet showed the best weed control potential determined by elemental composition, particle size distribution, and degradation rates.

INTRODUCTION

Much like the promise of the United States Postal Service jingle, "Through rain or shine, snow or sleet," weeds consistently deliver a multitude of problems to container nursery growers through spring and summer, fall and winter. Many of these problems are attributed to the ability of weeds to competitively affect the desired ornamental due to the limited amount of space, water, and nutrients within a container (Berchielli-Robertson et al., 1990). Numerous researchers have reported that only one weed in a small container (trade gal. or 1-gal.) could affect the growth of a container grown plant (Berchielli-Robertson et al., 1990; Fretz, 1972; Walker and Williams, 1989) but this is highly variable depending on both the crop and weed species.

The necessity to control weeds in container production has driven two practices in container production, hand pulling and herbicide applications. Hand weeding is increasingly expensive due to increasing labor cost (Gilliam et al., 1990) and further complicated by new immigration laws. Problems associated with herbicide applications in container production include non-target herbicide loss (Case and Mathers, 2006). This problem is further convoluted with increased container spacing at the time of application (Porter and Parish, 1993; Gilliam et al., 1990), such as that required for large container production (7 gallon and greater).

More recent research has shown that tree derived mulches, such as chipped cedar, pine-bark mini-nuggets, and Douglas fir, may reduce the need for hand weeding and herbicide application (Case and Mathers, 2003; Richardson et al., 2008; Bartley et al., 2014). Pine-bark mini-nuggets, as with other tree-derived mulches, create an environment that is not conducive to weed germination due to low fertility, large particle size, and hydrophobic properties (Richardson et al., 2008). This alternative method of weed control has also been shown to be very effective in large containers where the space in the container could be more easily allocated to a mulch layer instead of growing medium (Richardson et al., 2008; Bartley et al., 2014).

For a mulch to be deemed effective, at least in container production, the mulch must offer an inhospitable site for weed seed germination, and be able to maintain its integrity for an extended period of time. Growing large container plants warrants different growing practices due to the longevity of the plant growing in the container, in some instances, up to 18 months or more (Hunter Trees, LLC, pers. commun.). The problem with most organic

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mulches is they don't provide long-term weed control because of degradation (Altland and Krause, 2014). As the mulch degrades, it becomes an excellent substrate for weed germination due to decreasing particle sizes, barrier depth, and increasing water holding capacity.

Research has been conducted in landscape trials utilizing two different methods to determine mulch degradation rates. Duryea et al. (1999) developed the use of plastic rings to contain mulches on the surface of a plowed, open field and was able to determine decomposition rates by taking an initial dry weight and collecting the mulch within the rings at 1 and 2 years' time. Skroch et al. (1992) established landscape trial plots (4×4 ft) mulched with either pine bark, hardwood bark, cedar chips, longleaf pine needles, or shortleaf pine needles at a depth of 9 cm (3.5 in). Decomposition rates were collected by determining the amount of mulch it took to replenish the plot to the initial depth after 230 and 630 days. However, results from these studies may not be applicable due to the fertilization and irrigation abundance found in nursery and greenhouse production. The nature of this production is highly conducive for organic matter decay (Altland and Krause, 2014).

In order to establish best management practices when using alternative means of weed control, such as mulches, degradation rates of readily available tree species mulches must be recorded in a nursery production environment. These rates will ultimately determine mulch's weed control potential over time. In order to analyze mulch decomposition over time in a nursery production environment, litter bags, which allow for easy recapture of the mulch, were utilized. Litter bags, more commonly implemented in forestry and agronomic research, consist of an inert mesh or screen material resistant to decay such as nylon, woven polypropylene, or fiberglass with mesh spacing recommended based on the objective of the research (Robertson et al., 1999).

MATERIALS AND METHODS

This study is currently being conducted at the Paterson greenhouse complex of Auburn University in Auburn, Alabama. The experiment was initiated on 8 June 2015 when eastern red cedar, loblolly pine, Chinese privet, and sweet gum trees were harvested. Harvested trees measured 10-20 cm (4 to 8 in.) in diameter measured at 30.5 cm (12 in.) from the soil; only the trunk portions of the trees were utilized to provide mulch. Trees were chipped with a Vermeer BC1400XL brush chipper on 12 June 2015. Along with these four mulches, pine bark mini-nuggets were included (pine bark mini-nuggets landscape, Garick, LLC. Cleveland, Ohio) to provide a commercially comparative mulch treatment. All mulches were sifted through a series of screens to determine particle size distribution (Figure 1) and analyzed for elemental composition. Particles greater than 12 cm (4.75 in.) were discarded. All mulches were subjected to drying at 79°C (175°F) for 10 days to insure consistent moisture levels between mulch species.

Litter bags were made from 2 mm (0.08 in) fiberglass screening (New York Wire, Hanover, Pennsylvania). The 2-mm spacing size was preferred due to reports advising the use of at least 2 mm to allow for the loss of fine particles, macrofauna, and megafauna while maintaining sufficient contact with the substrate or growing medium (Robertson et al., 1999). Litter bags were 30 cm by 20 cm (12×8 in.) with the sides secured with marine-grade nylon thread to withstand constant moisture and the degenerative effects of UV light. With one side left unsecured, litterbags were filled on 23 June 2015 with 1400 mL (6 cups) of the designated mulch species. After the mulch was placed in the bag, the bag was gently shaken for 5 seconds to allow small particles to pass through the screening. The unsecured end was rolled and fastened with a binder clip and then initial weights of all bags were recorded. After the initial weight of the bag was recorded, the mulch bag was placed in a BP167 Lotus Pan (Nursery Supplies Inc., Kissimmee, Florida) filled with substrate that was pine bark and sand (6:1, v/v) amended per cubic yard with 2.3 kg (5 lbs.) dolomitic lime, 6.4 kg (14 lbs.) of Polyon® 18-6-12 (Pursell Technologies, Sylacauga, Alabama) and 0.7 kg (1.5 lbs.) Micromax® (Scotts Co., Maryville, Ohio). The lotus pan width allowed the mulch bags to be placed fully prostrate on the media surface without the need for an overabundance of unutilized medium. Drain holes were drilled into the containers to allow for adequate drainage.

Containers, with one mulch filled litter bag per container, were placed on a nursery pad and irrigated with 0.5 in. of water twice daily from impact sprinkler risers.

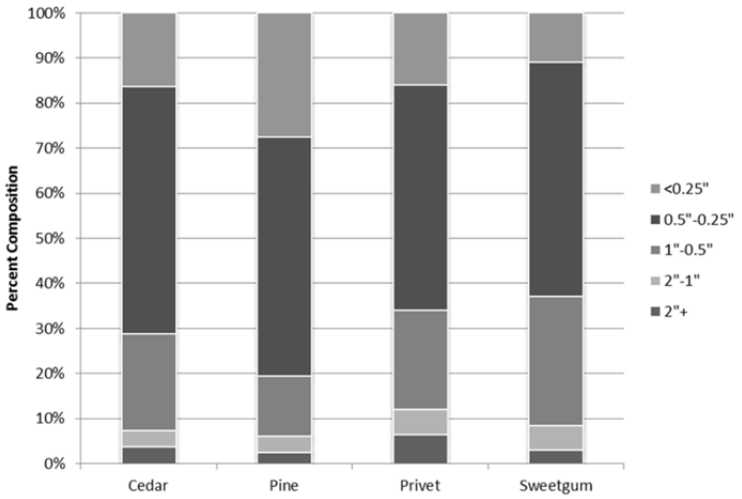

Figure 1. Particle size distribution by mulch species.

The study is a completely randomized design with five mulch treatments (eastern red cedar, pine, pinebark, privet, and sweetgum) with 48 reps per mulch treatment. In monthly intervals, eight reps per mulch treatment were collected, bagged, dried, and weighed. The ultimate objective of the study is to have sufficient data to allow for mulch degradation to be regressed over time. Currently, degradation has been evaluated at only 2 dates (July 27 and Aug 27, 2015) since study initiation. These data were subjected to ANOVA and individual difference between mulch species within each collection date were separated by Tukey's test at a significance level of 0.05.

RESULTS AND DISCUSSION

Initial data was taken on all mulch treatments to determine the elemental composition of each mulch species. The elemental composition of mulch is important because research has shown that those mulches with high carbon-nitrogen (C:N) ratios are favored over those with low ratios (Herms et al., 2001). In general, mulches with ratios greater than 30:1 have ratios high enough to prevent microbe colonization and exhibit nitrogen deficiencies, inhibiting weed growth (Herms et al., 2001). Analysis of each mulch species revealed all mulches had a C:N ratio of 183:1 or greater and trace amounts (typically less than 0.3%) of macro-nutrients (Table 1).

Table 1. Mulch composition analysis.

	Mulch type				
	Cedar[1]	Loblolly pine	Pine bark	Privet	Sweetgum
C:N ratio[2]	183:1	202:1	211:1	211:1	317:1
Nitrogen	0.26[3]	0.24	0.23	0.22	0.14
Phosphorus	0.02	0.02	0.02	0.02	0.02
Potasium	0.16	0.13	0.08	0.20	0.13
Calcium	0.66	0.18	0.18	0.28	0.31

[1]Only the trunk portions of each species were used and analyzed for composition.

[2]Carbon: nitrogen ratio

[3]Figures expressed for macronutrients are a percentage of the composition.

On 27 July 2015, eight replications from each mulch species were collected, bagged with paper bags, and dried for 5 days at 79°C (175°F). Each tagged mulch bag was weighed in the paper bag to insure no further loss of mulch particles occurred. Weights collected were subtracted from the initial weight taken on 23 June 2015 to yield the total weight lost due to degradation. Means comparisons between the five mulch treatments revealed significant differences in degradation rates after just 34 days of exposure in a nursery environment. Pine bark retained its integrity greater than any other mulch with a mean loss of just 3.8 g or 1.6% of the initial weight. Loblolly pine and cedar mulches each had a mean loss 8.5 g or 4.2% and 3.8%, respectively. The greater percentage loss of loblolly pine than cedar is attributed to the lower bulk density when compared to cedar. Sweetgum degraded more than pine bark, loblolly pine, and cedar but less than privet with a mean loss of 18.8 g or 7.2% of the initial weight. Privet, having the greatest initial bulk density, also degraded the most in the first month losing 31.7 g on average or 10% (Table 2). At this time, fungal growth (mycelium) was visually seen only in litter bags containing sweetgum mulch.

Table 2. Degradation of mulches over two months[1,2].

Mulch	Bulk density	Weight lost[3] (g)		Percent lost[4] (%)	Weight lost (g)		Percent lost (%)
Eastern red cedar	224.7[5]	8.5	C[6]	3.8	10.5	c	4.4
Loblolly pine	212.6	8.5	c	4.2	9.6	c	4.4
Pine bark mini-nuggets	229.8	3.8	d	1.6	5.9	c	2.3
Privet	324.5	31.7	a	10.0	43.7	a	13.5
Sweetgum	259.8	18.8	b	7.2	27.3	b	10.6

[1]1 month degradation rates were taken on July 27, 2015, 34 days after treatment.

[2]2 month degradation rates were taken on August 27, 2015, 64 days after treatment.

[3]Average weight lost = Initial weight - weight at the time of collection.

[4]Percent lost = (weight - initial weight / initial weight) x 100

[5]Bulk density measured in g 1400 cm^{-3}

[6]Means followed by the same letter are not significantly different according to the Tukey test (p=0.05).

n=8

On 27 Aug 2015, eight additional per mulch treatment reps were collected and analyzed in the same manner as the month previous. After 64 days, means comparison between mulch species revealed significant difference, expanding upon the results recorded in July. Pine bark, with a mean loss of 5.9 g or 2.3%, showed the least amount of degradation but was not significantly different than loblolly pine or cedar. Loblolly pine and cedar lost 9.6 and 10.5 g, respectively. Both loblolly pine and cedar lost 4.4% of their initial weight after 64 days in a nursery environment. Sweetgum, after two months, weighed 27.3 g lighter on average with an average percent loss of 10.6%. This loss is 3.4% greater that the percentage lost after just 34 days. Privet continued to show the greatest degree of degradation losing an average 43.7 g or 13.5% of its initial weight (Table 2). At this time, fungal growth (mycelium and reproductive structures) were seen in litter bags container sweetgum and privet mulches.

Due to the initial data presented, pine bark mini-nuggets have shown great potential to control weeds effectively due to its larger particle size distribution, minimal degradation rates over 64 days, and very high C:N ratio compared to the other mulches evaluated in this test. Other research results indicate that the ability of pine bark to withstand degradation may also be attributed to high lignin and low carbohydrate levels (Duryea et al., 1999). Loblolly pine and Eastern red cedar have shown good weed control potential due to good C:N ratios and degradation rates that are statistically equivalent to pine bark mini-nuggets after roughly two months. Loblolly pine and cedar's ability to resist decay may be attributed to phenolic chemicals or hydroxylated aromatic compounds which may negatively influence

decomposing organisms (Swift et al., 1979). These compounds were found in very high concentrations in a mulch blend containing both pine and cedar in a study conducted by Duryea et al. in 1999. Because of the high degree of decomposition recorded in both sweetgum and privet mulches, these data suggest that these mulches may provide weaker weed control efficacy than that of the three aforementioned. As this study progresses, it is our hope with this research to be able to study and plot degradation rates of these readily available mulches in order to better equip the container plant industry to effectively utilize mulch as an alternative method of weed control where current practices fall short.

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Effects of maintained substrate water contents from transplant to early stage of potted *Impatiens*©

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INTRODUCTION

In the first few weeks of bedding plant production, growers rarely irrigate so as to bring the substrate up to container capacity (the point at which the substrate can hold no more water against gravity). Research has been carried out to determine effects of substrate water content on bedding plant growth. Van Iersel et al. determined effects of substrate water content on petunia (*Petunia* ×*hybrida*) (2010). The substrate was maintained at or above substrate volumetric water contents (VWC; cm^3 water cm^{-3} substrate) of 5, 10, 15, 20, 25, 30, 35, and 40%. Shoot dry weight increased quadratically with VWC. There was a little increase in shoot dry weight between 25 and 40% VWC. All plants were well-watered uniformly for the first 9 days after being transplanted into the containers, and it took 9 days after irrigation treatment initiation for the substrate to reach the 5% VWC target. The substrate water content maintained during the first 9 days is not reported. Furthermore, substrate water content at container capacity was not reported. Therefore, it is not known how 40% VWC (the wettest treatment) compares to container capacity in the substrate and container used in the study.

Although growers do not regularly irrigate to container capacity in early stages of bedding plant production, growers differ on the ideal substrate moisture content for early stages of bedding plant production. It is commonly assumed that drier substrates cause roots to grow deeper into the container.

The objectives of this research were: (1) to determine the effects of substrate water content on *Impatiens walleriana* Xtreme™ Red in early production stages when not thoroughly watered in at potting and (2) to determine depth of root growth within the container at varying substrate water contents.

MATERIALS AND METHODS

This study took place at the Paterson Greenhouse Complex on the campus of Auburn University. On 9 July 2015, Fafard 3B was amended with 3.6 kg m^{-3} (6 lbs yd^{-3}) of 3-4 month Osmocote Plus 15-9-12 (15N-3.9P-10K, Scotts Co., Maryville, Ohio). Six 15.2 cm (6 in.) containers (Dillen Products, Middlefield, Ohio) were filled loosely to the brim and dropped 5 times on a table to settle the substrate to the lip 1.6 cm (0.625 in.) below the brim. The containers were then weighed and minor adjustments were made to bring the mass of added substrate to each container to 322 g (11.7 oz). After the six containers were filled, remaining substrate was sealed in a container to maintain the moisture content. Containers were watered until the substrate held no more water and were allowed to drain for 1 h in a dark room to container capacity. Containers were weighed, placed in a forced air drying oven at 60°C (140°F) for 2 days, and weighed again. It was determined that the containers held 136 g (4.8 oz) dry substrate, and the average gravimetric water content (GWC; grams water/grams substrate) at container capacity was 81%. On 14 July 2015, forty 15.24 cm (6 in) containers were filled with 332 g (11.7 oz) of the Fafard 3B substrate that had been sealed. *Impatiens walleriana* Xtreme™ Red that were sown on 15 June 2015 in a 200-cell plug tray were acquired from Young's Plant Farm, Inc. (Auburn, Alabama). Ten plugs were randomly pulled from the flat to determine the average fresh weight per plug (4.1 g). One plug was transplanted into each container. Containers were placed in a temperature controlled greenhouse maintained between 18°C (64°F) and 34°C (93°F) for the duration of the experiment. Each container was weighed to determine the volume of water needed to

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bring the container to a target GWC of 80, 76, 72, 68, or 64%. Although substrate GWC at container capacity was 81%, a target GWC of 80% was selected as the highest target GWC in order to prevent leaching. The weight of each container at its target GWC was calculated by dividing the dry weight of Fafard 3B in each container by 1 minus the target GWC and adding the empty container weight and the average plug fresh weight [136.2 g ÷ (1-Target GWC) + empty container weight + 4.1 g]. The volume of water needed to bring each container to its target weight was slowly distributed evenly across the surface using a 60 mL (2.12 oz) syringe directly after placing containers in the greenhouse. This process was repeated daily between 8:00AM and 10:00AM to bring each container to the Target GWC. Each treatment had eight replications. The experimental design was a Completely Randomized Design, and containers were spaced 20 cm (7.9 in.) center to center on a greenhouse bench. Plant size was recorded weekly by calculating size indices for all plants [(height + widest width + perpendicular width) ÷ 3)]. Four plants per treatment were harvested three weeks after potting (WAP) on 4 August 2015. Shoots were harvested and placed in a forced air drying oven at 60°C (140°F) until dry to determine shoot dry weights. The harvested containers were hand watered until substrate could hold no more water and left to sit in a dark, cool room for 2 hours. The containers were then weighed to determine the weight at container capacity. Substrate pH and electrical conductivity (EC) were measured using leachate samples collected using the pour-through method. Containers were placed in a freezer at -2°C (28.4°F). On 6 August 2015 the average shoot weight for each treatment was calculated and added to the formula for the target weights in place of the initial plug weight. The remaining plants were harvested in a similar manner 6 WAP on 25 August 2015. Once root balls were thoroughly frozen, a machete and rubber mallet were used to divide root balls in half top from bottom. Root balls averaged 8 cm (3.2 in.) in height from top to bottom. Roots were washed and dried in a forced air drying oven at 60°C (140°F) until dry and weighed to determine root dry weights. All data were analyzed using regression analysis within JMP statistical software (SAS Institute Inc., Cary, North Carolina).

RESULTS AND DISCUSSION

Size index increased linearly and quadratically with target GWC 1 and 2 weeks after potting (WAP) (Table 1). Size index increased linearly at the 0.05 α level with target GWC 3, 4, and 5 WAP. However, by 6 WAP there was a strong quadratic relationship (α=0.001) between size index and target GWC. Size indices 6 WAP ranged from 20.0 to 21.9 among plants in target GWC treatments between 64 and 76% and sharply increased to 27.2 among plants in the 80% target GWC.

Table 1. Size indicies of *Impatiens wallerina* Xtreme™ Red as affected by target gravametric water content (GWC).

Target GWC (%)	1 WAP[1]	2 WAP	3 WAP	4 WAP	5 WAP	6 WAP
64	4.0	6.7	10.9	14.4	19.3	20.0
68	4.4	7.5	11.3	15.5	19.5	20.4
72	4.6	7.8	11.1	16.2	20.7	21.3
76	4.5	8.1	11.8	16.4	20.8	21.9
80	4.9	9.1	12.8	17.4	22.6	27.2
Significance[2]	L***Q**	L***Q***	L*	L*	L*	L***Q***

[1]Weeks after potting.

[2]Regression response non-significant (NS), linear (L), or quadratic (Q) at the 0.05 (*), 0.01(**), or 0.001 (***) level.

Shoot dry weight 3 WAP ranged from 0.65 to 0.75 g among plants in target GWC treatments between 64 and 76% and sharply rose to 1.18 g in the 80% target GWC treatment (Table 2). Roots of all plants were present only in the top 4 cm (1.57 in.) of the root ball 3 WAP. As a result, only total root dry weight is presented. There was no regression response between root dry weight and target GWC 3 WAP.

Table 2. *Impatiens wallerina* Xtreme™ Red shoot and root weights as affected by target gravimetric water content (GWC) 3 weeks after potting.

Target GWC (%)	Shoot dry weight (g)	Root dry weight (g)
64	0.65	0.90
68	0.73	0.55
72	0.75	0.50
76	0.68	0.40
80	1.18	0.55
Significance[1]	L*Q**	NS

[1]Regression response non-significant (NS), linear (L), or quadratic (Q) at the 0.05 (*), 0.01(**), or 0.001 (***) level.

Shoot dry weights 6 WAP responded linearly and quadratically to target GWC at the 0.001 α level (Table 3). The average weights increased from 4.35 to 6.50 g between 64 and 76% target GWC treatments and sharply rose to 9.33 g at the 80% target GWC treatment. There was no response for top dry root weight or total dry root weight to target GWC. However, bottom dry root weight increased linearly and quadratically with target GWC at the 0.05 α level. There was significantly greater root dry weight in the bottom half of containers in the 80% target GWC treatment compared to all other treatments.

Table 3. *Impatiens wallerina* Xtreme™ Red shoot and root weights as affected by target gravimetric water content (GWC) 6 weeks after potting.

Target GWC (%)	Shoot dry weight (g)	Roots		
		Dry weight total (g)	Dry weight top (g)	Dry weight bottom (g)
64	4.35	5.35	5.20	0.15
68	4.63	4.30	3.53	0.78
72	5.67	7.27	6.60	0.67
76	6.50	5.30	5.08	0.23
80	9.33	8.33	6.23	2.10
Significance[1]	L***Q***	NS	NS	L*Q*

[1]Regression response non-significant (NS), linear (L), or quadratic (Q) at the 0.05 (*), 0.01(**), or 0.001 (***) level.

Varying target GWC levels also affected the pH and EC levels in the substrate 3 WAP and 6 WAP (Table 4). At 3 WAP there was a strong linear and quadratic response in respect to pH and target GWC at the 0.001 α level. The pH decreased linearly from 5.62 at the 64% target GWC treatment to 5.14 at the 76% target GWC level and sharply decreased to 4.73 at the 80% target GWC treatment. There was no response between EC levels and target GWC 3 WAP. There was a quadratic relationship at the 0.05 α level between pH and target GWC in the 6 WAP. Substrate pH ranged between 4.48 and 5.25. There was a quadratic response at the 0.01 α level between substrate EC and target GWC. EC ranged from 5.12 to 6.56 mS cm^{-1} between 64 and 76% target GWC treatments, while substrate EC measured only 1.85 mS·cm^{-1} in the 80% target GWC treatment.

Table 4. Substrate pH and EC as affected by target gravimetric water content (GWC) 3 and 6 weeks after potting.

Target GWC (%)	3 WAP[1]		6 WAP	
	pH	EC (mS cm^{-1})	pH	EC (mS cm^{-1})
64	5.62	5.55	5.25	5.44
68	5.53	6.17	5.04	6.56
72	5.47	7.98	4.76	5.21
76	5.14	5.95	4.48	5.12
80	4.73	6.73	5.09	1.85
Significance[2]	L***Q***	NS	Q*	L*Q**

[1]Weeks after potting.

[2]Regression response non-significant (NS), linear (L), or quadratic (Q) at the 0.05 (*), 0.01(**), or 0.001 (***) level.

Total irrigation volume applied per plant increased linearly and quadratically at the 0.001 α level (Table 5). Irrigation volume applied per plant increased 12% between 64 and 68% target GWC, 13% between 68 and 72% target GWC and 5% between 72 and 76% target GWC. However, irrigation volume applied per plant increased 30% between 76 and 80% target GWC. Leachate volumes of 4 mL or less were collected in the 80% target GWC treatment during the first ten days of the experiment (data not shown).

Table 5. Total average irrigation volume applied per plant as affected by target gravimetric water content (GWC) from 17 July 2015 to 24 August 2015 on *Impatiens wallerina* Xtreme™ Red.

Target GWC (%)	Irrigation volume (mL)
64	2134
68	2392
72	2696
76	2843
80	3690
Significance[1]	L***Q***

[1]Regression response non-significant (NS), linear (L), or quadratic (Q) at the 0.05 (*), 0.01(**), or 0.001 (***) level.

Size index 6 WAP, shoot dry weight 3 WAP, shoot dry weight 6 WAP, and bottom root dry weight had significant increases between 76 and 80% target GWC. As stated earlier, growers typically do not irrigate to container capacity. Although containers in the 80% target GWC treatment were irrigated to a level close to container capacity daily, the substrate dried considerably between irrigation events due to high daily temperatures. Results may differ at cooler temperatures. A moisture characteristic curve was developed for Fafard 3B using the modified long column method in order to relate GWC to VWC (Altland et al., 2010). Target GWC levels of 64, 68, 72, 76 and 80% relates to VWC levels of 7, 12, 20, 32, and 47%, respectively. In the study by Van Iersel et al. (2010), shoot dry weights increased little between 25 and 40% VWC, while shoot dry weight in our study increased significantly between target GWC levels of 76 and 80% which relates to VWC levels of 32 and 47%. While plants in our study were irrigated once per day in order to bring substrate up to the target GWC, plants in the study by Van Iersel et al. were irrigated on-demand when substrate water content dropped below the set VWC level (2010). As a result, substrate in the 76% target GWC treatment dropped to a GWC as low as 72% between irrigation events which is equivalent to a VWC of 25%. Substrate water contents at this level resulted in significantly less plant growth.

In this study, maintaining low substrate moisture contents directly after transplanting resulted in significantly smaller plants as soon as 2 WAP. Lower substrate water contents did not result in a higher percentage of total root growth in the bottom half of the container. Results may differ in cooler temperatures and with more frequent irrigation events.

Literature cited

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