

Oregon Department of Agriculture and Oregon Association of Nurseries
Nursery Research Project Final Report 2016

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Title: Developing novel, disease resistant forms of nursery crops

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Background

The Ornamental Plant Breeding Program at Oregon State University takes a broad and long term approach to cultivar development. We breed a wide variety of plants including shade trees, flowering trees and shrubs, conifers, and herbaceous perennials in an attempt to address the needs of the robust and diverse Oregon Nursery Industry. Below is a brief explanation of eight (8) projects we are conducting to develop new cultivars that have improved phenotypes and/or disease or insect resistance. These projects are within the larger program I direct, are ongoing, and have previously been funded. I believe we have made promising steps toward improved cultivars on every front and in one case we have already released an improved cultivar of flowering currant.

Ribes sanguineum. Flowering currant is native to the Pacific Northwest and is commonly recommended by native plant enthusiasts as a replacement plant for butterfly bush or other non-natives that may have weedy tendencies. It is drought tolerance once established, has attractive flowers, and encourages native pollinators in gardens. Unfortunately, it is leggy and also has issues with powdery mildew and aphids. In 2014, we released ‘Oregon Snowflake’ as an improvement to ‘White Icicle’ due to its improved growth form and novel dissected leaf. As part of our mutagenesis program we have identified 6 plants that did not have either powdery mildew or aphids during 2015. **Our next goals include developing varied flower colors of compact, cutleaf forms and starting to breed for powdery mildew resistance.**

Galtonia candicans. Cape hyacinth or summer hyacinth is an herbaceous perennial propagated from bulbs with large, strap-like leaves that generally grow 2 to 3 ft high and 2 in wide. It produces a loose raceme of 20 to 30 pendulous white flowers; each 1 to 2 in long, which can have great impact due to the large size of flowers but the length of the scape over which flowers are dispersed is undesirable. A shorter inflorescence that concentrated the flowers over a shorter area would be a significant improvement. Additionally, this species produces an extremely large amount of seed and may end up being weedy in the garden, so we are also interested in reducing fertility. To my knowledge, there is only one cultivar of this species (‘Moonbeam’ = double flowers). A superior form has the potential to establish dominance in the market. We have developed a number of improved selections by mutagenesis that are being propagated for replicated trials. Data from 2014 demonstrate the effectiveness of our treatments to induce mutations and alter seed viability (Fig. 1). **Our goals are to confirm our initial selections maintain compact habit, continue observations of our second generation, and propagate improved selections by tissue culture. We hope to begin trials with participating nurseries in 2016.**

***Philadelphus* spp.** Mockorange has lost favor in recent years and I believe this is, in part, due to sub-par cultivars being released. Some of the newer cultivars have little fragrance, which should be a selling point for all mockorange. We have been using a number of different species and cultivars in our improvement program (Table 2). Since 2012 we have used 40 different parental combinations and have field planted nearly 800 progeny for evaluation. We are working to combine the excellent and unique fragrance of *P. mexicanus* with branched inflorescence and increased floriferousness of cultivars like ‘Blizzard’ as well as double flowers from cultivars like ‘Miniature Snowflake’. What has been shocking is the heritability of important and often complex traits such as fragrance and inflorescence architecture. This gives me confidence that other novel traits such as glossy leaf from *P. texensis* var. *coryanus* will be heritable. We have propagated 8 selections our 2012-13 crosses that will either be used in advanced testing for potential release and/or continued use as

parents. **Our goals are to trial improved selections we have identified and continue introgressing novel traits into commercially viable forms. We hope to begin trials with participating nurseries in 2016.**

Sarcococca confusa. Fragrant sweetbox is a shrub prized for its ability to thrive in dry shade, an exposure most plants will not tolerate. It has few pest and disease problems and requires little maintenance. We have developed a number of selections in the M1 generation with altered leaf shape and more compact habit via induced mutation. We are growing a second generation (M2) from the mutated parents to select for additional clones that could have potential and have identified 4-5 promising selections. **Our goals are to trial the already identified superior forms and to select additional clones with potential from our second generation. Growth of our selections has been slow, so 2016 trials will be in-house until we can generate sufficient material.**

Lilacs. We have made more than 20,000 crosses during 2013-15 between myriad cultivars of lilacs including intra- and intersectional crosses to develop novel cultivars that are compact, disease resistant (bacterial blight), reblooming, and have “common lilac fragrance”. Our intersectional crosses have failed to and in order to effectively move the project ahead, we are refocusing our efforts on reblooming among the small-leaved lilacs and compact, disease resistant forms in common lilac. We developed a mapping population for reblooming on which we are conducting a technique called genotyping by sequencing that will generate approximately 10,000 data points across the genome. We will then use this along with other molecular and phenotypic data to develop a linkage map and hopefully discover a marker associated with reblooming. A marker for this important trait will reduce our costs and speed our breeding efforts greatly. **Our goals are to continue pursuing similar crosses to increase progeny number, evaluate hybrids we have already developed, and work toward genetic mapping of the reblooming trait to develop molecular markers for marker assisted selection (MAS).**

Cotoneaster. Cotoneasters once were an important crop for many nurseries nationally due to extreme drought resistance and overall toughness in landscapes; but production has been reduced dramatically. Contributing factors for this decline may include poor form of some that require pruning (e.g. ‘Coral Beauty’), fire blight susceptibility, weediness, and lack of new or exciting cultivars. We have been using fire blight resistant parents in our crosses and have developed a number of very promising selections (Table 3). **Our goals are to expand testing of these selections to other locations, conduct formal tests of fertility (fruit set/seed set/germination) in a glasshouse using controlled crosses, and continue developing cultivars using resistant species as parents. Three selections are being trialed at two commercial nurseries including locations around Oregon and in Alabama where disease pressure is high. I hope to identify additional interested partners in 2015.**

Styrax japonicus. Recently there have been a number of improved cultivars of Japanese snowbell released including ‘Fragrant Fountain’, ‘Evening Light’, and ‘JLWeeping’ (Marley’s Pink Parasol™). These cultivars have generated excitement around Japanese snowbell and have created an opportunity to combine distinct traits. **Our goal is to combine the weeping habit of ‘Fragrant Fountain’ and ‘JLWeeping’, respectively, with the foliage color of ‘Evening Light’ to develop new cultivars of weeping, purple foliage Japanese snowbell with either white (from ‘Fragrant Fountain’) or pink (from ‘JLWeeping’) flowers.**

Cercidiphyllum japonicum. In katsuratree there are weeping cultivars (‘Amazing Grace’ and ‘Morioka Weeping’) and there is a red foliage cultivar (‘Red Fox’ aka ‘Rotfuchs’) but there is no cultivar that combines these traits. We have made crosses in 2013 and 2014 between weeping cultivars and Red Fox that were field planted in 2015. Most plants are green and upright (there are no weepers) but there are 7 plants that continue to have red foliage as of September 15, 2015. These selections may represent an alternative to the shrubby habit of ‘Red Fox’ and be more of a shade tree that possesses red foliage, which would be a breakthrough for this species. Furthermore, we plan to go to the next generation and hopefully recover selections that possess both traits. **Our primary goal remains to combine weeping and red leaf traits but we are keeping an eye toward other novel phenotypes among our progeny.**

Methods and Timeline

Ribes sanguineum: We planted 215 hybrids between Oregon Snowflake and other cultivars in 2015 (Table 1) and hope to see these flower in 2016. We will make crosses with our 6 plants identified as disease and aphid resistant. We will grow the M2 generation and select any unique phenotypes and additional sources of resistance to powdery mildew and/or aphids.

Galtonia candicans. We have put 10 selections into tissue culture for propagation since twin scaling was largely unsuccessful in 2014. We hope to have additional M1 plants of these 10 selections for trialing by spring 2016. We have planted the M2 generation, of which many are quite precocious and are flowering after less than 1 year from seed. We will continue observing the M2 during 2016 and 2017. The 10 superior M1 selections will be dug and containerized for observation and breeding; other plants will be destroyed fall/winter 2015/2016.

Philadelphus. Evaluate superior clones in house and with participating nurseries. We stuck cuttings of selections summer 2015 and hope to have liners for distribution during winter/spring 2016. We are also collecting data on rootability. Continue expanding germplasm used as parents to introgress additional traits such as glossy leaves and more pigmented calyx.

Sarcococca confusa. Continue growing improved selections to reach appropriate size for field evaluation and distribution. Propagate these to distribute to participating nurseries for testing. Continue growing M2 population and make selections in 2016; destroy remaining plants.

Lilacs. Continue phenotyping hybrids reblooming, which will be completed by summer 2016. Conduct molecular mapping study to identify markers for reblooming that can be used in our breeding program. We should get the GBS data back by winter 2016 and will conduct bioinformatics winter-summer 2016. We hope to have our common lilac hybrids flowering in 2016 and we will continue rating these for bacterial blight resistance and try to determine heritability of this resistance from 'Old Glory', which has shown durable resistance thus far.

Cotoneasters. We will continue observations of our improved selections at more locations to determine how field resistance corresponds to our greenhouse inoculations. We have rooted between 30-110 of each selection for distribution to collaborating nurseries and researchers. We will continue making crosses with resistant parents to develop new hybrids.

Styrax japonicus. We made reciprocal crosses between both weeping cultivars and 'Evening Light' to develop four families of crosses; however, we underestimated the difficulty (at least for us) at getting sufficient seed set. We will make at least 200 pollinations of each cross in 2016.

Cercidiphyllum japonicum. Continue observations of 151 F1 progeny that were field-planted in 2015. At flowering we will backcross with 'Morioka Weeping' and 'Amazing Grace' to recover weeping trait.

Budget Summary

Salary

Faculty Research Assistant (40% FTE)	\$15,765
Other Payroll Expenses (OPE) (OSU health benefits, insurance, retirement)	\$10,720

Services and Supplies

Field and greenhouse fees	\$3,775
Genotyping by sequencing*	\$4,000

Total	\$34,260
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*approximately 50% of costs

Table 1. Results of crossing ‘Oregon Snowflake’ with commercial cultivars to develop new selections that combine habit and leaf form of ‘Oregon Snowflake’

Cross	Cutleaf	Not
Oregon Snowflake x White Icicle	22	26
Oregon Snowflake x King Edward	0	48
Oregon Snowflake x Pokey's Pink	0	15
White Icicle x Oregon Snowflake	14	17
King Edward x Oregon Snowflake	0	50
Pokey's Pink x Oregon Snowflake	38	59
Total	74	215

Table 2. *Philadelphus* crosses during 2015, including our first complex hybrids of mockorange. Germination is ongoing.

Cross	Flowers	Capsules
86.60 (Blizzard x Icelandic) x mexicanus	12	12
86.68 (Blizzard x Icelandic) x mexicanus	7	7
88.27 (Snow White x unknown) x mexicanus	11	11
Belle Etoile x mexicanus	20	18
Belle Etoile x Polar Star	8	8
mexicanus x Snow White	10	9
mexicanus x Belle Etoile	10	10
Miniature Snowflake x Belle Etoile	4	4
Miniature Snowflake x mexicanus	3	3
Polar Star x mexicanus	12	7
Snow White x Belle Etoile	9	9
Snow White x mexicanus		
12 cross combinations	126	108

Table 3. Cotoneaster accessions, % shoot necrosis in greenhouse inoculation tests, rooting percentage, and days to root (well-rooted and ready to pot).

Accession	% shoot necrosis	Rooting %	Days to root
H2011-01-002	0	100	61
H2011-01-003	17	100	61
H2011-02-001	0	87	27
H2011-02-005	1	100	27
H2013-08-012	Testing 2016	100	27
Coral Beauty	11	Not rooted in study	

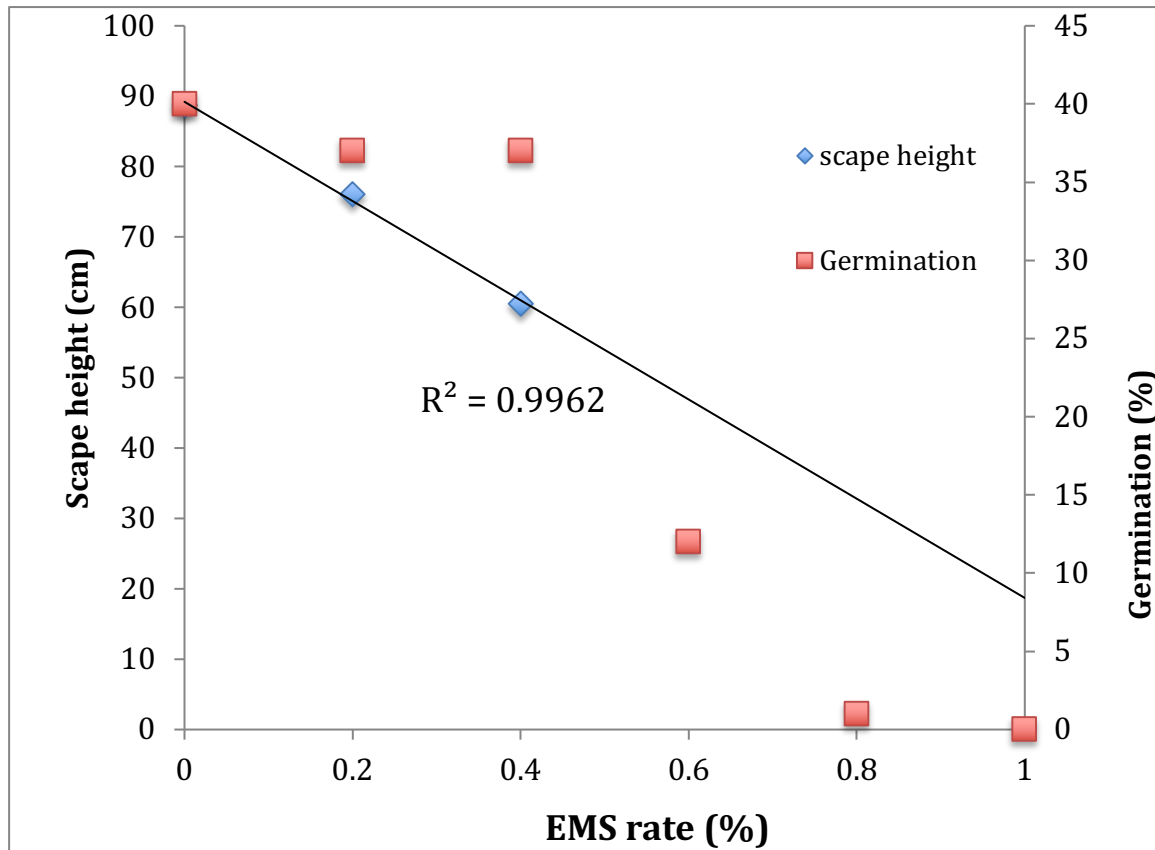


Fig. 1. Increasing rate of EMS (ethyl methane sulfonate) reduced scape height and seed germination of *Galtonia candicans*. Control (0%) plants had a mean height of 89 cm, the 0.2% treatment had a mean height of 76 cm, and the 0.4% treatment had a mean height of 61 cm. No plants survived to field planting for the 0.6% or higher treatments.

UPDATE 20 December 2016

***Ribes sanguineum*:** We grew out 285 progeny resulting from crosses between Oregon Snowflake and available cultivars to determine the heritability of ornamental traits and combine the improved leaf form and compact habit of Oregon Snowflake with superior flower colors of Pokey's Pink and King Edward. This population is segregating for our traits of interest (**Table 4, Figure 2**) and thus far we have recovered 9 plants that are pink with dissected leaves and compact habit. Unfortunately, we recovered no plants with red flowers and dissected leaves. There are several possibilities for this but based on our observations, it appears these traits are linked in repulsion. This means that the genetic recombination event that will lead to the desired trait combination will be extremely rare. In order to assess our hypothesis and also move ahead with the breeding project, we have collected F2 seed from these plants in the field to grow the next generation. The F1 plants from which we collected seed included every phenotype combination and each cross combination (including reciprocals). Seed were sown on December 19 and many were already germinating as a result of cold stratification. We should be able to phenotype the next generation for leaf morphology by early- to mid-spring although I do not expect to observe flowers until 2018. For the next round, we will discard all plants that are not cut leaf to reduce costs. Additionally, we collected from plants that had no aphid damage. I expect this will lead to an F2 generation with improved resistance as well as our traits of interest. We did not select for powdery mildew resistance, as we observed no pressure in our field during 2016.

Table 4. Flowering currant hybrids conducted to combine flower color with dissected leaf.

Cross	Cutleaf	Normal	White	Pink	Red
White Icicle x Oregon Snowflake	14	16	23	0	0
Oregon Snowflake x White Icicle	24	22	29	0	0
Pokey's Pink x Oregon Snowflake	30	63	36	37	0
Oregon Snowflake x Pokey's Pink	0	15	1	13	0
King Edward x Oregon Snowflake	0	50	1	18	26
Oregon Snowflake x King Edward	0	51	0	20	25



Fig. 2. Segregating population for dissected leaf and compact habit. These traits appear to be linked and all plants with dissected leaves are much more compact.

***Galtonia candicans*.** We propagated a number of selections from our M1 population by divisions and tissue culture. Our observations are that the form is not able to be compared to our field-grown material, as the growth is considerably more vigorous. However, the improved selections (made based on field observations) have better flowering and tighter, denser growth than controls. Therefore, it does appear that the improvements are stable, only now we need at least another year in containers to compare. Our M2 population does not show the expected improvements over the M1. This likely is due to the fact that our M1 plants that were significantly more compact were also sterile. This resulted in plants almost exclusively being pollinated by control (untreated) plants. Currently, 5 selections are in tissue culture and will be subcultured to multiply plants and then acclimated during late winter to be grown in containers for 2017.

***Philadelphus*.** Capsules collected and sown from controlled crosses made in 2015 had very poor germination. It's unclear why this happened, as in previous years there was excellent germination of nearly all crosses. We did have some success from which we made some preliminary selections based on early growth form, this is summarized in **Table 5**. We also made crosses with new germplasm in 2016 (**Table 6**).

32 selections made from our original population of approximately 500 plants are growing after transplanting in fall 2015. I have begun making selections in the population of 243 plants developed in 2014, which is very promising (**Fig. 3**). Due to lack of success with softwood or semi-hardwood cuttings, we collected cuttings on November 7, 2016, and most of these 7 selections performed better using dormant hardwood.

Overall, we have more improved selections than we have been able to manage thus far. The plan for 2017 is to conduct severe selection to reduce inferior or redundant plants and continue systematic evaluation of existing progeny before growing additional seedlings. Controlled crosses and seed collection will resume in 2018.

Table 5. Summary of *Philadelphus* progeny selected on June 3, 2016.

Hybrid accession	Female	Male	No. selected
H2015-011-01	Philadelphus Snow white	Philadelphus mexicanus	15
OP2015-01-12	Philadelphus 'Snow White x coronarius (88.22)*	Open pollinated	5
OP2015-02	Philadelphus Icelandic X Blizzard (86.34)	Open pollinated	5
OP2015-03	Philadelphus Icelandic X Blizzard (86.34)	Open pollinated	5
OP2015-04	Philadelphus SnowWhite X P. coronarius (88.27)	Open pollinated	5

*Number in parenthesis is field location.

Table 6. 2016 *Philadelphus* crosses.

Female	Male	Flowers
Belle Etoile	texensis var. coryanus	20
Miniature Snowflake	texensis var. coryanus	12
Belle Etoile	mexicanus 'Floroplana'	6
coronarius 'Aureus'	texensis var. coryanus	10
coronarius 'Aureus'	Belle Etoile	20
Snow Dwarf	texensis var. coryanus	10
Snow Dwarf	mexicanus 'Floroplana'	5
Snow Dwarf	Belle Etoile	5
Mexicanus 'Floroplana'	Belle Etoile	4
Belle Etoile	Desert Snow	15
Total		107

*These capsules have been collected and will be sown during January 2017.



Fig 3. 2014 mockorange population. Selection and removal of inferior plants will begin fall 2016.

Sarcococca confusa. Of the seven selections from the M1 population, 6 have continued to maintain their desirable habit. Brief descriptions are found in Table 7. I also have kept 8 plants from the M2 population that are promising, mainly for compact “non-wild” habit. I plan to propagate these in 2017, after another season of evaluation.

Table 7. Selections of fragrant sweetbox made in our program, traits, and status of propagation.

Accession	Propagated?	Description
0.2-48-14	No	Previously compact but has lost form – discard
0.4-24-12	No	Very compact. Maintains good habit – Cuttings failed in 2016
0.4-48-9	Yes (3 #1s)	Very compact and stable among propagules – Cuttings failed in 2016
0.4-24-40-1	No	Very narrow leaf – Cuttings failed in 2016
0.2-24-08	Yes (3 #1s)	Compact habit, narrow leaf and stable among propagules – 58 quarts from cuttings rooted in 2016 – Excellent performance thus far in production.
0.2-48-10-2	Yes (5 #1s)	Compact habit, narrow leaves with slight contorted shape. Copper and chartreuse new growth observed 7/21. Propagules stable –
0.2-24-32	Yes (20 #1s + 15 #3)	23 quarts from cuttings rooted in 2016 Chartreuse selection we have observed for a number of years. Continues to exhibit chartreuse growth in winter but has not garnered interest among growers due to lack of impressive phenotype during field day. 14 quarts from cuttings rooted in 2016..

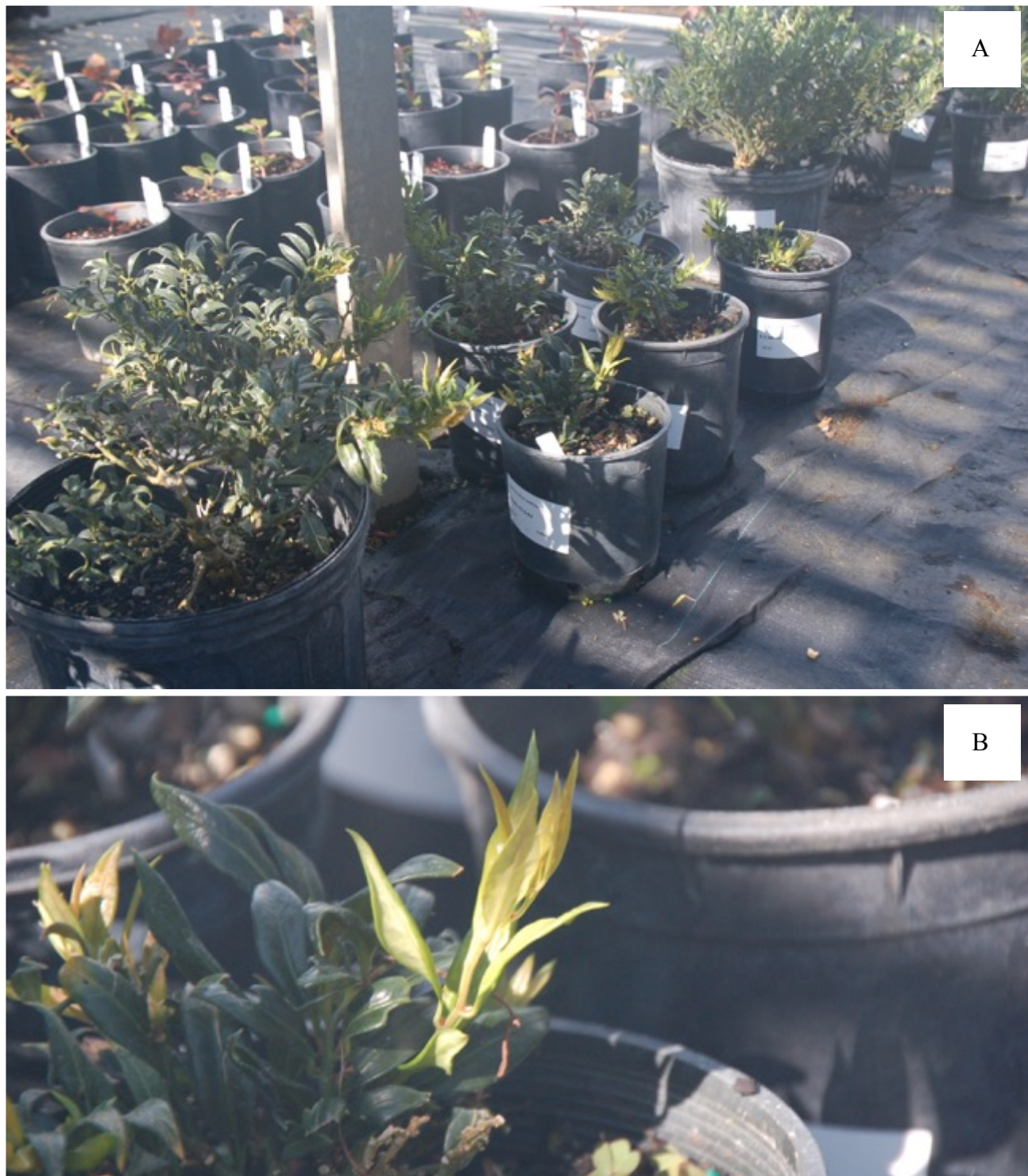


Figure 4. Mother plant (foreground) and 5 propagules of sweetbox accession 0.2-48-10-2-1. Propagules show no signs of reversion (A). A closeup of the new growth on this selection (B).



Figure 5. Sweetbox accession 0.2-24-32-01 showing chartreuse phenotype during winter 2015-2016.

Lilacs. We have confirmed that our population of ‘Palibin’ x ‘Bloomerang’ (and the reciprocal cross) are segregating for the reblooming trait. We have more than 20,000 SNP markers from GBS that are being annotated. Additionally, we conducted two other methods of NexGen sequencing (HiSeq and MiSeq) that will all be used in combination with our GBS data. We should have a linkage map and draft genome by spring 2017. With continued refining of our marker data, phenotype data, and a linkage map I hope to have useful markers for the reblooming trait by 2019. This will streamline our breeding process significantly.

There are a number of very promising selections from which we will collect cuttings either 2016 or 2017, an example of a superior selection in Figure 6. We collected a 6-8 cuttings from 8 different selections in 2016 but they have not rooted yet. We have sown 1,381 seeds collected from ‘Palabini’ x Bloomerang plants collected in 2016 and these F2 seedlings are beginning to germinate. An additional 4,688 seeds were

collected from F1 plants and are in cold stratification. I expect a high degree of variation in this population. It is possible that we could begin using our marker(s) in development on this population to expedite selection. Assuming even modest germination, marker assisted breeding will be quite useful to reduce population size.



Fig. 6. Lilac hybrid of Palibin x Bloomerang on June 30, 2016. This plant clearly shows the reblooming trait for which we are selecting and at this point is a better rebloomer than available cultivars. While it is early in evaluation, I am optimistic that we will release plants that are better growers, have better blight resistance, and better reblooming than available cultivars.

We also have many combinations of common lilac using ‘Old Glory’, that show promise for disease resistance. Progeny from crosses using ‘Old Glory’ segregate for purple leaves (**Fig. 7**). We will continue phenotyping these progeny but my hope is that disease resistance is correlated with leaf color (purple=resistant) such that leaf color can be used as an easily selectable phenotypic marker for resistance. I was not able to fully evaluate this hypothesis, as there seemed to be little bacterial blight among these progeny. To more effectively screen for resistance/susceptibility, I plan to introduce several diseased container plants to our container pad to ensure disease pressure in 2017.



Fig. 7. Segregation of progeny from ‘Old Glory’ lilac for purple foliage. In addition to being attractive, I hope pigmented foliage is related to disease resistance to allow early selection for disease resistant progeny.

Cotoneasters. Three selections of cotoneaster developed at OSU that are fire blight resistant in our greenhouse studies have continued to be distributed nationally. Currently, we have sent these selections at Kansas State University, Virginia Tech, University of Georgia, and at 7 nurseries. University collaborators will be collecting and reporting formal data on growth and disease. I hope nurseries are able to provide scheduling estimates, which will be critically important data when other growers are considering if these may be profitable cultivars for their product line.

We have continued making crosses with these improved selections to expand the palate of new cotoneasters for various landscape niches. Table 8 summarizes our 2016 crosses. Seeds were scarified and are in cold stratification to be sown on 30 January 2017. Based on previous years’ results, I expect these hybrids to include excellent growth forms and disease resistance.

Table 8. 2016 cotoneaster crosses.

Female	Male	Flowers	Seeds
dammeri	apiculatus	46	5
H2012-02-001	apiculatus	30	48
H2012-02-001	H2012-01-002	35	29
H2012-02-003	H2012-02-001	40	0
dammeri	splendens	20	0
H2012-02-001	splendens	20	12
salicifolius	splendens	27	0
salicifolius	apiculatus	87	0
Total		305	94



Fig. 8. Three cotoneaster selections from left to right are H2012-01-002, H2012-02-005, and H2012-02-001, which are being evaluated at multiple regions around the country with ‘Coral Beauty’ on the far right. In our trials our selections (unpruned) are far superior to ‘Coral Beauty’.

Styrax japonicus. 2016 crosses were successful, at least for fruit set. Data are summarized in Table 8. We have good fruit set (Table 8, Fig. 9) seed were collected and we have completed the initial warm stratification period to break morphological dormancy. To test for optimal germination, half of our seeds were cold stratified for 1 month and the other half for 2 months. The one month treatment has been moved back to the greenhouse and the 2 month treatment will be moved in 1 week. We hope to have germination by winter 2017. I expect this will be a project similar to the katsuratree – it will require growing F2 plants to recover these two traits, which I expect are both recessive.

Table 9. 2016 *Styrax* crosses to develop a purple leaf, weeping cultivar.

Cross	Flowers	Fruit
Evening Light x Marley’s Pink Parasol	105	30
Evening Light x Fragrant Fountains	142	21
Marley’s Pink Parasol x Evening Light	27	2



Fig. 9. Fruit set on 'Evening Light' japanese snowbell (July 21, 2016).

Cercidiphyllum japonicum. We have 10 'Red Fox' x 'Morioka Weeping' plants from 2013 crosses and 76 from 2014 crosses. Also, from 2014 crosses we have 16 'Red Fox' x 'Amazing Grace' plants. These are all field planted at the Lewis-Brown Hort Farm and growing well. None of these plants have flowered. Improved production methods have been implemented including improved training and irrigation. I expect these improvements will expedite this project. Also, we have rooted 2 genotypes that we will try to force to flower in containers. While I was unsuccessful in the past, we are also considering attempting to encourage flowering using GA treatment of field plants.