Auburn University
Southern Forest Nursery
Management Cooperative

RESEARCH REPORT 15-05

THE USE OF SEED POLYMERS AND SEED COLORANTS AS SEED TREATMENTS FOR SOUTHERN PINES

by
Tom Starkey and Scott Enebak

INTRODUCTION

Seed treatments are the application of chemical ingredients and/or biological organisms to seed in order to suppress, control, or repel plant pathogens, insects, or other pests that attack seeds, seedlings or plants. Seed treatments such as inoculants, herbicide, micronutrients, plant growth regulators, seed coatings, or colorants are commonly applied to seed just before sowing. Seed that has been treated is intended for planting only and not for food or feed uses. Seed treatment begins when seeds are removed from long-term cold storage and allowed to imbibe water before placement in cold storage for a period of time. These vital steps are part of a process called "stratification" that is used to overcome embryo dormancy, coat-induced dormancy and initiation of other biochemical or physiological seed germination processes. When seeds are removed from long-term cold storage the moisture content of the seed is generally less than 6%. At this moisture level the germination process has not begun. As the seed imbibes water and the moisture content increase to 30% - 40% some of the biochemical and physiological process involved with germination begin. Many studies have shown that stratification improves the speed and uniformity of germination that are vital to producing a uniform seedling crop (Barnett 1976, Barnett and Verla 2004). Seed treatments are not the same for all southern seed or between nurseries for the same species. Both the time of soaking in water and the time in cold storage differ by species and even families within a tree species.

When seed has completed the necessary stratification time in cold storage, seeds are removed for the next phase; seed treatment. In the seedling production, there are two pesticides that are commonly added to seed just prior to sowing in the nursery. Some nurseries coat the seed with Thiram®, a registered fungicide that has both fungicidal and animal repellant properties. More commonly, seeds are coated with either Bayleton® or Proline® to provide initial fungicidal protection from fusiform rust. In order to insure that chemicals remain on the seed neither chips nor flakes off (called "dust-off") a sticker such as latex paint is commonly applied at this time. Finally, many nurseries use a seed dye to facilitate seed sowing, quality checks and establishment of history plots.

Seed Polymers: Latex paint has been used for many years as part of the seed treating operation process prior to sowing southern pine seed with the objective of adhering the pesticides to the seed. Actually the name "latex" is a misnomer since there is no latex in latex paint. True latex originally came from the Brazilian rubber tree (*Hevea brasiliensis*) but is now primarily produced in Southeast Asia. Synthetic polymers which look like natural latex are used to make latex paint but have different chemical makeup and properties than latex rubber.

For many years most nurseries used latex from Dow Chemical Company. International Forest Seed Company previously purchased this compound in large 55 gallon drums, repackaged it and sold it in smaller quantities to nurseries throughout the southeastern US. There have been many changes in the Dow product over the years. References in the literature cite the use of Dow Latex 512-R, 512-L, 630, 636 and 2028; all generally diluted for seed treatment at 1 part latex to 9 parts water when used as a seed coating.

In a survey of southern nurseries, over 80% of nursery managers use a form of latex in their sowing operations (Starkey et. al. 2015). Approximately half the nurseries still use a Dow product. The other nurseries purchase latex from a local paint or hardware store. The latter group cites the ability to purchase latex in small quantities as their primary reason. Those that are still using a Dow product all indicate difficulty in finding or purchasing latex, especially in small quantities. One nursery responded that they are still using the same product purchased more than 15 years ago. Another point of concern with the use of latex is termed "dust-off". This occurs after pesticides are applied to the seed with the latex and dried. During the handling associated with sowing, part of the latex/pesticide coating flakes off and becomes a dust irritant for workers. This concern was expressed to the Southern Forest Nursery Management Cooperative with the most recent reregistration of Bayleton® by the US EPA. Seed clumping after treating also was a concern expressed with the use of latex.

In the agricultural arena, seed polymers are being used extensively with soybean, wheat, sunflower, corn, beans and cotton. These seed polymers are designed for one function; to protect seed treatments applied to the seed by providing uniform coverage (Herbert 2003). They are compatible with fungicides, insecticides, inoculants and colorants. The use of seed polymers also reduces the "dust-off" problem. In addition they enhance seed plantability by reducing clumping and stickiness thus increasing seed drop accuracy. A quote in Farm Industry News, December 3, 2012 describes polymers by saying "Seed polymers are a bit like "force fields" *(e.g. Star Wars)*: You cannot always see them, but they are there to protect."

There are many companies that manufacture seed coating polymers and promote similar seed treatment products. Bayer CropScience and BASF are devoting significant resources in this developing field of seed coating polymers and colorants. In late 2012 BASF acquired Becker Underwood. Many nurseries in the south have reportedly used Becker Underwood as a source of seed colorant. In discussing seed treatment methodology with a Becker Underwood scientist, he recommended that CF Clear should be tested as a replacement for latex for pine seed at the rate of 0.25 fl oz/50 lbs of seed.

Seed Colorant: Seed colorants are used extensively by nurseries at the time of sowing to provide better contrast of the seed and soil. Seed colorants aid the sowing operation by increasing seed visibility during the sowing and later during sowing checks/history plots. Unfortunately, some seed colorants have been added to the seed without verifying that the dye has no effect on seed germination. In a discussion with a nursery manager, he believed that they had experienced some reduction in germination using Becker Underwood red seed colorant. The nursery manager conducted a small test at their nursery comparing Becker Underwood red seed colorant and Tempera Red paint. Tempera paint is a permanent fast drying painting medium consisting of colored pigment mixed with a water-soluble binder medium usually a glutinous material such as

egg yolk. In his small test, the nursery manager confirmed that there was a reduction in germination when the Becker Underwood red seed dye was used when compared to the Tempera red paint.

Because of the difficulties and obtaining and locating the true Dow latex, the recommendation of CF Clear, and the antidotal evidence of seed colorants and seed germination concerns, a number of studies were conducted at Auburn to determine the safety and efficacy of these products on seed germination.

METHODOLOGY

In the CF Clear studies the general methodology described below was used in all studies described below:

1. We assumed it takes 8 fl oz of liquid for 15 lbs of seed.
2. The 8 fl oz of liquid includes all chemicals and colorants.
3. 8 fl oz /15 lb seed = 27 fl oz /50 lb of seed.
4. The suggested rate for CF Clear, for pine seed was 0.25 fl oz/50 lbs of seed.
5. 0.25 fl oz CF Clear /27 fl oz for seed = a 1% solution of CF Clear.
6. All used CF Clear at 1% rate.
7. Control = water only.

CF Clear Study #1. The initial study was to compare germination of slash pine seed using 4 treatments: 2 rates of CF Clear, 1 rate of latex and a water control. The treatments of CF Clear used were a 1x rate) and a 5x rate. Latex paint was also used at the 1% rate. Following stratification, slash pine seeds were treated with the CF Clear, Latex and Water and then sown in the seedling stress boxes at Auburn University. Two rows of 50 seeds for each treatment were sown per stress box that were replicated 4 times. Once germination began, counts were made the same time each day. All data were analyzed in SAS 9.2 using analysis of variance (ANOVA). Treatment means were compared using Duncan's Mean Separation Test when treatment means were significantly different at alpha=0.05.

CF Clear Study #2. In a second experiment, three rates (1x, 4x, 8x) of CF Clear and a water control were tested on longleaf pine germination. Two rows of 25 seeds for each treatment were sown per stress box that were replicated two times. Once germination began, counts were made the same time each day. Due to the limited number of replications, the data was not analyzed statistically.

CF Clear Study #3. In a third experiment we compared CF Clear with and without seed colorant on slash pine seed. The treatments in this study were:

1. Control
2. Control + Tempra @ 2.3%
3. Control + Becker Underwood Red @2.3%
4. CF Clear @1% + Tempra @ 2.3%
5. CF Clear @1% + Becker Underwood Red@2.3%
6. CF Clear @1%

Slash pine seeds were treated and sown in the seedling stress boxes. Two rows of 50 seeds for each treatment were sown per stress box that were replicated 3 times. Once germination began, counts were made the same time each day. All data were analyzed in SAS 9.2 using analysis of variance (ANOVA). Treatment means were compared using Duncan's Mean Separation Test when treatment means were significantly different at alpha=0.05.

Seed Colorant Study. Two seedlots of loblolly (one with a confirmed high germination seedlot and a second with confirmed low germination seedlot), slash, shortleaf and longleaf pine were stratified for 40, 40, 30 and 7 days, respectively prior to the start of the study. Four plant trays (21" x 11" x 2": L xW x D) for each species filled with sand were placed on a plant heating pad on a greenhouse bench for each species. Bottom heat was used to speed germination since this study was initiated in January.

Each group of 60 seeds was slightly moistened with water and then sufficient dye placed on the seed to color. Seeds were tumbled/stirred and then the seed polymer, CF Clear (1%), was applied. Tumbling and stirring of the seeds continued until seeds were dry. Seeds were place in plastic bag in refrigerator until they were sown the following day.

Fifteen seeds of each treatment and for each species (60 seeds total) were randomly sown in each tray in a line and lightly covered with seed. Once germination began, counts were made the same time each day (Figure 5).

All data were analyzed in SAS 9.2 using analysis of variance (ANOVA). Treatment means were compared using Duncan's Mean Separation Test when treatment means were significantly different at alpha=0.05.

RESULTS AND DISCUSSION

CF Clear Study #1. The latex treatment reduced seed germination when compared to all other seed treatments examined (Figure 1). The germination of CF Clear 1x rate compared to the germination of the latex treatment is shown in Figure 2. In seven of the ten days, germination was monitored, as indicated by the red arrows, the seed treated with CF Clear 1X was greater (at alpha= 0.05) than the seed treated with latex.

CF Clear Study #2. The germination of longleaf pine seed when three rates (1x, 4x, 8x) of CF Clear and a water control were tested is shown in Figure 3. Since there were only two replications, a statistical analysis of the treatment differences was not performed. The water control germination is in the middle of the three CF Clear treatments and similar to the CF Clear 1X rate.

CF Clear Study #3. The comparison of two seed colorants and CF Clear is shown in Figure 4. An interesting observation was that the seed treated with Becker Underwood Red (CF Clear + Becker Underwood Red and Becker Underwood Red colorant) also had a reduced germination and a reduction in total germination. Results from the ANOVA and Duncan's Mean Separation for Days 1, 3, 5, 7 and 9 are presented in Table 1. For Days, 1, 3, 5 and 7 the two Becker Underwood Red treatments germination were less than the Control and CF Clear. On Day 9, the germination of

seed treated with Becker Underwood Red alone was less than the germination of both non-treated Control and CF Clear treated seed.

Seed Colorant Study. Germination for the all the pine species tested ranged from 9 to 12 days. The rate of germination for each seedlot and all treatments is shown in Figures 6-10. Accompanying each individual graph is a table that presents the treatment means separation for each day of measurement. Seed treatments followed by the same letter are not significantly different from each. Since no single dye provided the best germination at all dates measured, the treatments that were consistently in either the top three lines or bottom four lines was tabulated and is presented in Table 4.

Below are the summary points from the dye study:

- The powered dyes were harder to apply to pine seed than the liquid dyes.
- The use of Becker Underwood Red as a seed colorant reduced germination over the non-treated seed in 4 of the 5 seedlots tested. If this product is used, one should test the colorant on small seedlots and collect germination data in your nursery situations.
- The liquid dye from Chromatech, "Chromatint Red X_3353 Dispersion" dramatically reduced germination in all seedlots. When the company was informed of our results I received the following email: *"Our technical director just returned from vacation and looked at your results. After investigating, he discovered that a biocide was added to the dispersion by accident. This is what affected the germination of the seeds."* These colorants could be tested again without the biocide.
- The Prism Scarlet from Precision Lab had consistently good germination. Like all new products, small seedlot tests should be conducted in each nursery situation. At least one nursery manager currently uses this dye on all their seed

MANAGEMENT IMPLICATIONS

The seed treatment CF Clear did not negatively impact the germination of either slash or longleaf pine seed. The use of a seed polymer as a seed coat during the treatment process, such as CF Clear, offers some distinct advantages over latex. Latex is paint, not formulated for seed and thereby may contain potential ingredients that may inhibit seed germination or not protect pesticides on the seed. Seed polymers are formulated to adhere to seed and protect other seed treatments that have been applied. Seed polymers are widely used in agriculture and could find their place in forest seed nurseries over latex paint or Dow Latex. The compound CF Clear sells for approximately $60/gallon and is currently being sold at all major chemical suppliers such as Helena, Greenpoint Ag or any company selling BASF products.

The effect of the seed colorant on seed germination was unexpected. Below is a working hypothesis as to why seed colorant may impact the germination of forest nursery seed:

- When dyes are used on agricultural seeds the internal moisture of the seed is less than 10%. Agricultural seeds are not commonly stratified (soaked and stored) prior to sowing. In the nursery industry, seed, (which is also less than 10% moisture), is soaked in water from 4 to 24+ hours raising the internal moisture >30% , and placed in a refrigerator for up to 45 days during which time the internal biochemical and physiological process necessary for

germination begin. Then prior to sowing, the seed is treated with fungicides, maybe a repellant, seed dye and a coating agent.

- It is possible that the dye may be absorbed into the seed coat and thus interfere with the ongoing germination process. One seed specialist suggested acetic acid used in the manufacture of certain dyes as a possible culprit of reduced germination.
- Realize that anything added to stratified seed may potentially alter germination
- Another situation which may negatively impact germination is treated seed that remains in the cooler longer than necessary due to weather delays or equipment problems in sowing. These delays may result in seeds cracking prior to sowing or longer time for the seed treatments to be absorbed into the seed coat.

REFERENCES

Barnett, J. P. 1976. Delayed germination of southern pine seeds related to seed coat constraint. Canadian Journal of Forest Research 6(4):504-510.

Barnett, J. P.; Varela, S. 2004. A review of chemical treatments to improve germination of longleaf pine seeds Native Plants Journal. 5:18–24.

Herbert, R.M. 2003 Application of latex emulsion polymers in seed coating technology. ASTM Special Technical Publication 1449: 55-67.

Starkey, T.E.; Enebak, S.A.; South, D.B. 2015. Forest seedling nursery practices in the Southern United States: bareroot nurseries. Tree Planters' Notes. 58(1): 4–17.

Table 1. Percent germination of CF Clear and two seed colorants on slash pine in the greenhouse, Auburn, AL 2014.

Treatment	Day 1	Day 3	Day 5	Day 7	Day 9
CF Clear Polymer	30% A	64% A	74% A	81% A	85% A
Control	29% AB	64% A	75% A	82% A	84% A
CF Clear + Tempera Red	18% ABC	55% AB	72% A	80% AB	84% A
Tempera Red	17% BC	45% AB	66% AB	78% AB	82% A
CF Clear + Becker Red	12% C	38% C	57% B	71% BC	77% AB
Becker Red	11% C	35% C	53% B	65% C	71% B
lsd 0.05	*12%*	*13%*	*12%*	*9%*	*8%*

Same letters within a column indicate that the percent germination are not different.

Table 2. Seed colorant, type, form and manufactures used in the study.

Dye (TRT) Number	Name	Company	Form
1	FD&C Red #40	Organic Dyestuffs	Powder
2	Orcobrite Pigment Red Bryn 6002	Organic Dyestuffs	Liquid
3	Prism Scarlet	Precision Lab	Liquid
4	Chromatint Red X_3353 Dispersion	Chromatech	Liquid
5	FD&C Red 40 Dustmaster 2000	Chromatech	Powder
6	Becker Underwood Red	Becker Underwood	Liquid
7	**CONTROL**		

Table 3. Source of seed dyes used to treat pine seed in the study.

Dye #1 & #2
ORCO Organic Dyes and Pigments 1015 Concord Parkway, Concord, NC 28027 800-556-6784 http://www.organicdye.com/industries/seed-colorants
Dye #3
Precision Laboratories 1429 S. Shields Drive Waukegan, IL 60085 800-323-6280 http://www.precisionlab.com/agriculture/products/seed-enhancements-treatment/prism-scarlet-seed-colorant
Dye #4 & #5
Chromatech Inc. 7723 Market Drive Canton, MI 48187 800 545-5075 http://www.chromatechcolors.com/industries/seeds-dyes/
Dye #6
BeckerUnderwood New Address: BASF 801 Dayton Ave Ames, IA 50010 http://www.agproducts.basf.us/products/research-library/flo-rite-1706-tech-sheet.pdf

Table 4. Comparison of germination rates for seed colorant study for five pine seedlots.

Treatment #	Dye	Lob High	Lob Low	Slash	Shortleaf	Longleaf
1	FD&C Red #40					
2	Orcobrite Pigment Red Bryn 6002					
3	Prism Scarlet					
4	Chromatint Red X_3353 Dispersion					
5	FD&C Red 40 Dustmaster 2000					
6	Red					
7	CONTROL					

= Top three graph lines

= Bottom four graph lines

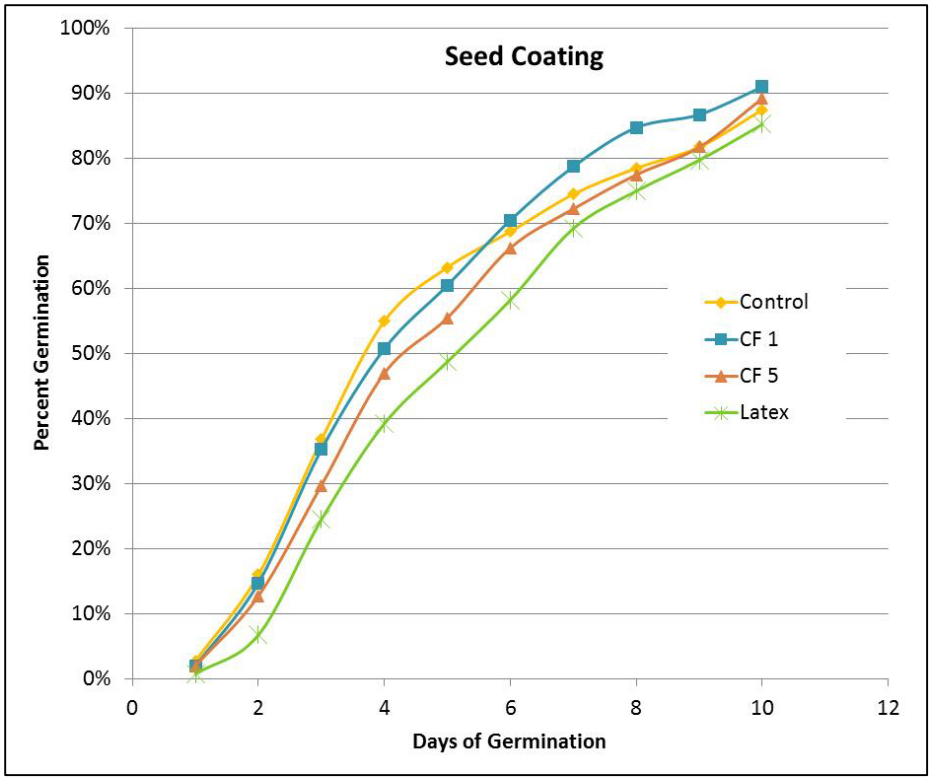


Figure 1. Germination rate of two rates of CF Clear and latex on slash pine in the seedling stress facility, Auburn AL 2014.

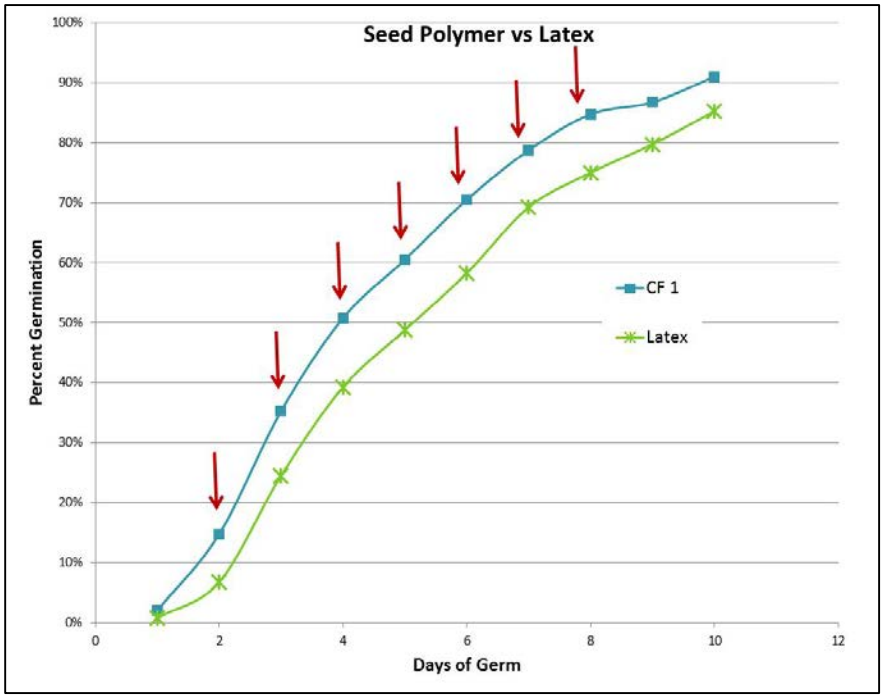


Figure 2. Comparison of the germination rate of CF Clear and latex on slash pine in the seedling stress facility, Auburn, AL 2014.

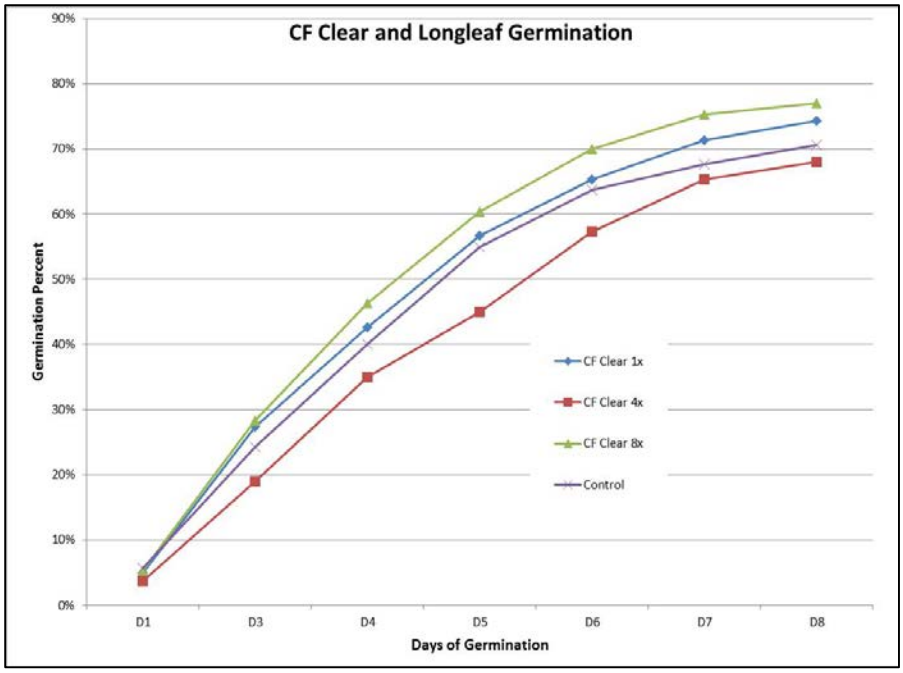


Figure 3. Germination rates of three rates of CF Clear on longleaf pine in the greenhouse, Auburn, AL, 2014.

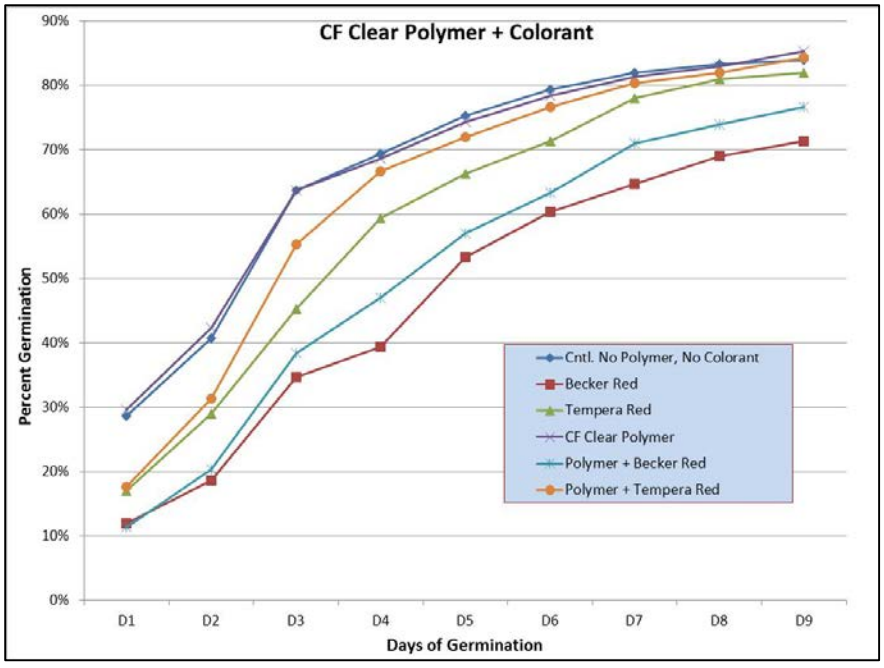


Figure 4. Comparison of the germination rates of CF Clear and two seed colorants on slash pine in the greenhouse, Auburn, AL, 2104.

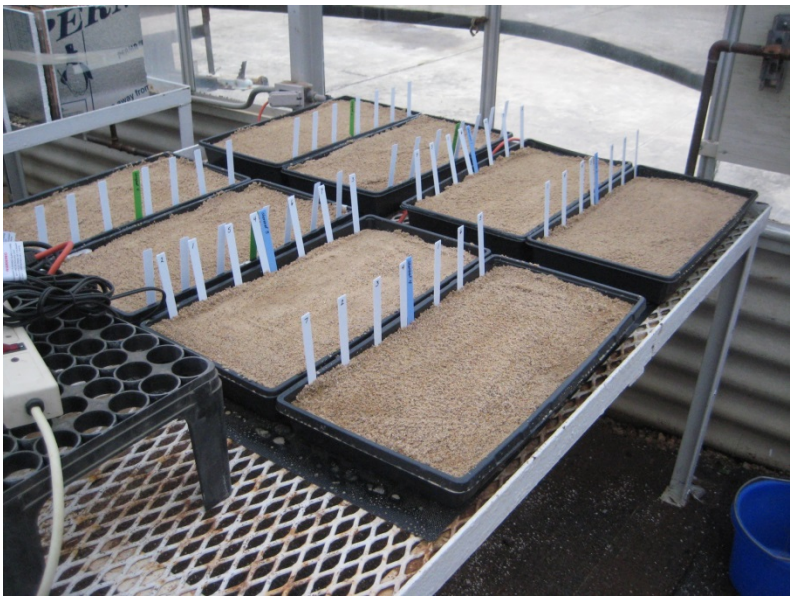
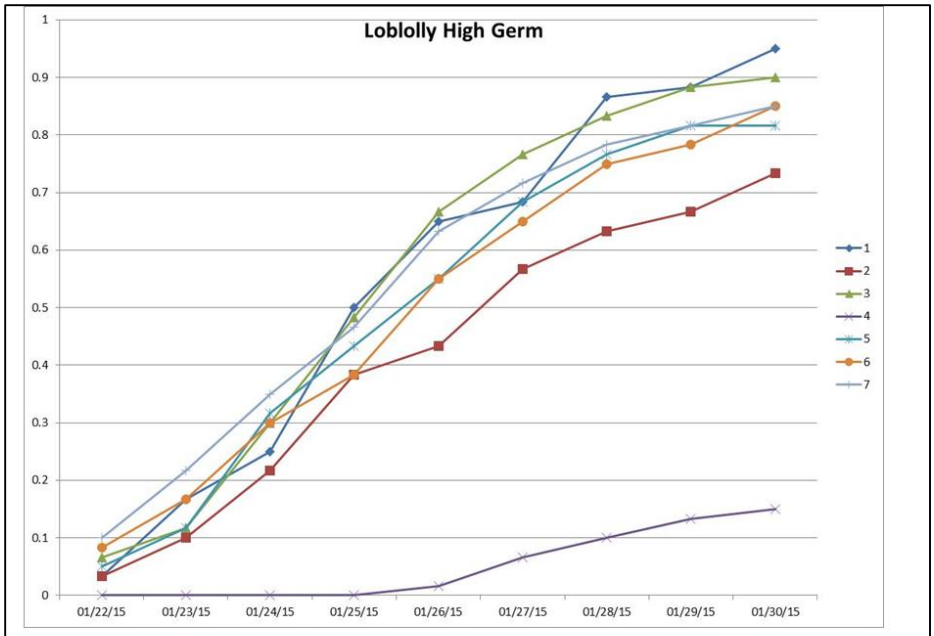


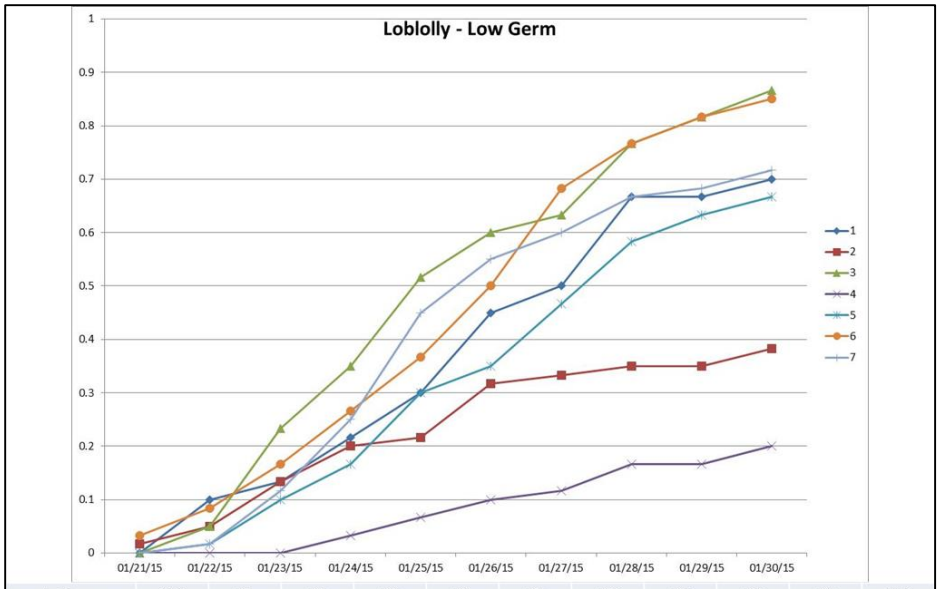
Figure 5. Greenhouse bench showing the plant trays and experimental layout for the pine seed tested for germination after treatments, Auburn, AL, 2014.



LobHi	TRT	D1	D2	D3	D4	D5	D6	D7	D8	D9
LobHi	1	A	A	A	A	A	A	A	A	A
LobHi	2	A	A	A	A	B	A	B	B	B
LobHi	3	A	A	A	A	A	A	A	A	A
LobHi	4	A	A	B	B	C	B	C	C	C
LobHi	5	A	A	A	A	AB	A	AB	AB	AB
LobHi	6	A	A	A	A	AB	A	AB	AB	AB
LobHi	7	A	A	A	A	A	A	AB	AB	AB

Figure 6. Germination rate for the seven seed colorant treatments for loblolly pine (high germ seedlot). In the accompanying table, treatments followed by the same letter within a column are not significantly different

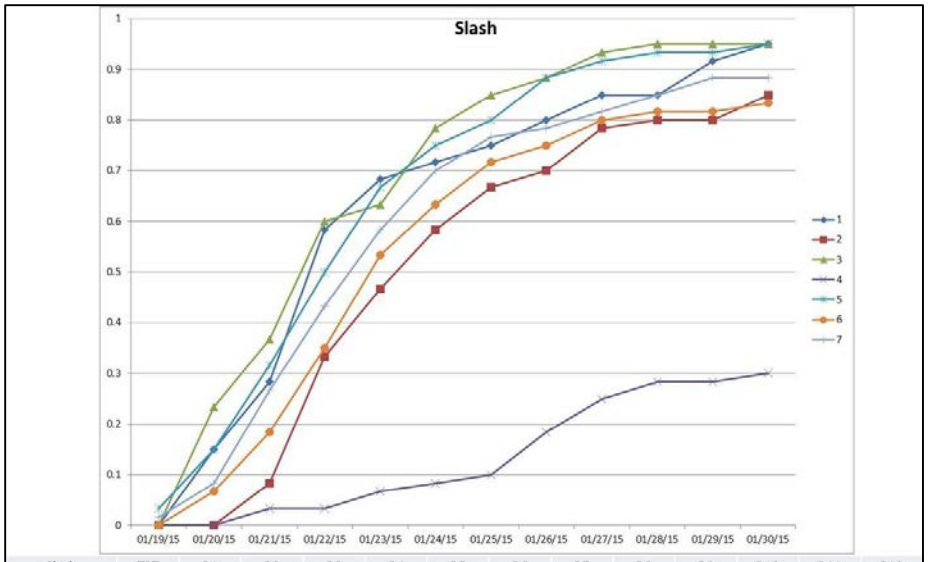
*TRT = See Table 2 for specific seed dye treatments 1-7 in Figures 6-10.



LobLow	TRT	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10
LobLow	1	A	A	AB	AB	AB	AB	AB	A	A	AB
LobLow	2	A	AB	AB	AB	BC	BC	BC	B	B	C
LobLow	3	A	AB	A	A	A	A	A	A	A	A
LobLow	4	A	B	B	B	C	C	C	B	B	D
LobLow	5	A	AB	AB	AB	AB	ABC	AB	A	A	B
LobLow	6	A	AB	A	A	AB	AB	A	A	A	AB
LobLow	7	A	AB	AB	A	A	AB	AB	A	A	AB

Figure 7. Germination rate for the seven seed colorant treatments for loblolly pine (low germ seedlot). In the accompanying table, treatments followed by the same letter within a column are not significantly different.

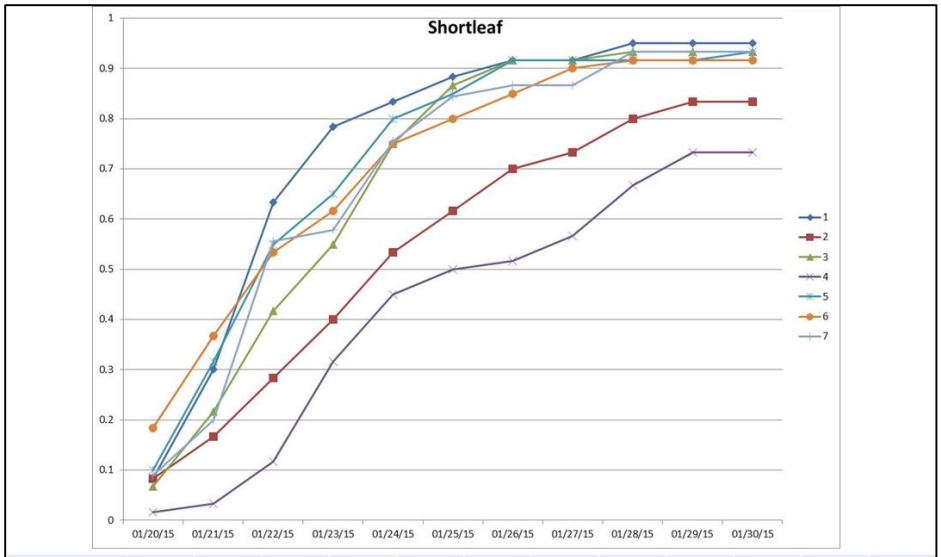
*TRT = See Table 2 for specific seed dye treatments 1-7 in Figures 6-10.



Slash	TRT	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10	D11	D12
Slash	1	A	AB	AB	A	A	AB	AB	AB	AB	AB	AB	A
Slash	2	A	C	BC	B	A	B	B	B	B	B	C	B
Slash	3	A	A	A	A	A	A	A	A	A	A	A	A
Slash	4	A	C	C	C	B	C	C	C	C	C	D	C
Slash	5	A	AB	A	AB	A	AB	AB	A	AB	AB	A	A
Slash	6	A	BC	ABC	B	A	AB	AB	AB	AB	AB	BC	B
Slash	7	A	BC	AB	AB	A	AB	AB	AB	AB	AB	ABC	AB

Figure 8. Germination rate for the seven seed colorant treatments for slash pine. In the accompanying table, treatments followed by the same letter within a column are not significantly different.

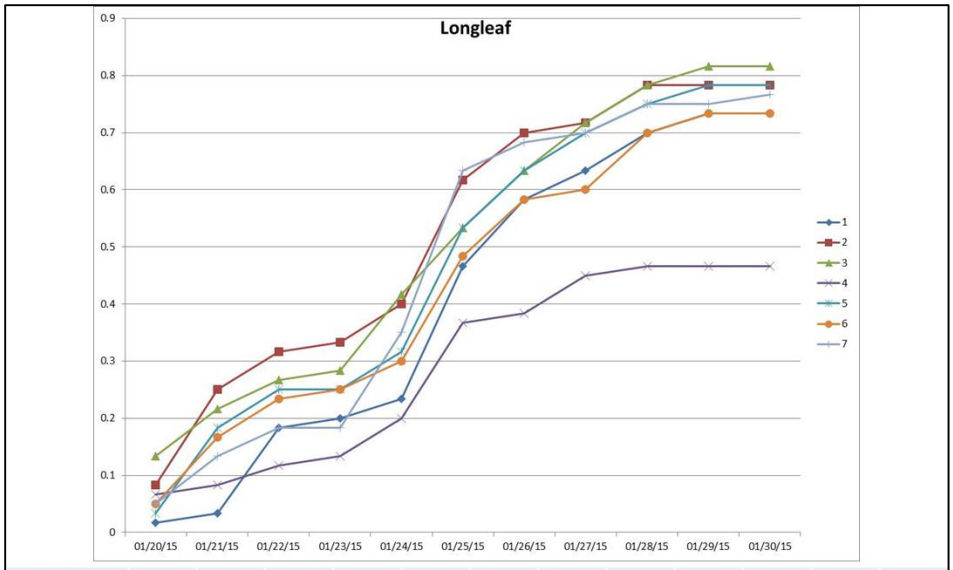
*TRT = See Table 2 for specific seed dye treatments 1-7 in Figures 6-10.



Shortleaf	TRT	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10	D11
Shortleaf	1	AB	AB	A	A	A	A	A	A	A	A	A
Shortleaf	2	AB	AB	AB	AB	B	BC	B	B	B	AB	AB
Shortleaf	3	AB	AB	AB	ABC	A	A	A	A	A	A	A
Shortleaf	4	B	C	C	C	B	C	C	C	C	B	B
Shortleaf	5	AB	AB	AB	AB	A	A	A	A	AB	A	A
Shortleaf	6	A	A	AB	AB	A	AB	A	A	AB	A	A
Shortleaf	7	AB	ABC	AB	ABC	A	A	A	A	A	A	A

Figure 9. Germination rate for the seven seed colorant treatments for shortleaf pine. In the accompanying table, treatments followed by the same letter within a column are not significantly different.

*TRT = See Table 2 for specific seed dye treatments 1-7 in Figures 6-10.



Longleaf	TRT	D1	D2	D3	D4	D5	D6	D7	D8	D9	D10	D11
Longleaf	1	B	B	A	A	A	A	AB	A	A	A	A
Longleaf	2	AB	A	A	A	A	A	A	A	A	A	A
Longleaf	3	A	AB	A	A	A	A	AB	A	A	A	A
Longleaf	4	AB	AB	A	A	A	A	B	A	A	A	A
Longleaf	5	B	AB	A	A	A	A	AB	A	A	A	A
Longleaf	6	B	AB	A	A	A	A	AB	A	A	A	A
Longleaf	7	B	AB	A	A	A	A	A	A	B	B	B

Figure 10. Germination rate for the seven seed colorant treatments for longleaf pine. In the accompanying table, treatments followed by the same letter within a column are not significantly different.

*TRT = See Table 2 for specific seed dye treatments 1-7 in Figures 6-10.