Establishing Native Vegetation and Improved Invasive Species Control on North Carolina Roadsides

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Studies were also initiated to assess the best means of control for alligatorweed and Japanese knotweed, two invasive and problematic species among North Carolina roadsides and right-of-ways. At 7 MAT, Japanese knotweed control with glyphosate, imazapyr, glyphosate plus imazamox, and glyphosate plus imazapyr was at least 93%. Glyposate plus penoxsulam controlled the plant 75%. Treatments that did not include imazapyr or glyphosate did not exceed 20% control.

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Summary

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1.0 Introduction

The North Carolina Department of Transportation (NCDOT) is responsible for operation and maintenance of the 78,500 mile North Carolina highway system. Vegetation management along these roadsides is critical for highway safety and function. Vegetation succession is the natural process of one plant community replacing another. The initial phase is one of high disturbance and few plants. A long series of interactions takes place leading to a "climatic climax", or a community of plants that will maintain itself in competition with outside species under no disturbance (Daniel et al. 1979). On roadsides, construction or renovation would be the high disturbance phase and encroachment of pine trees would be one of the final phases. Thus, management is needed to maintain roadsides at an early stage of succession to provide safety and functionality of the highway.

Currently, mowing is the predominant tool for roadside vegetation management. While this is effective in reducing plant biomass in mowed areas, most vegetation is only suppressed (especially perennial plants) and budget constraints limit the amount of mowing that can occur in a given year. In addition, tree limbs often protrude out over mowed areas forcing the routine mowers to back away and allowing the tree line to creep towards the road. Over a number of years, this slow creep results in little to no recovery zone, blocked highway signs, and the possibility of limbs hanging into the road. Rising fuel costs will further impact budget constraints as mowing contracts are renewed.

While roadsides must be maintained for highway safety under normal operating conditions, natural disasters present additional concerns for maintenance. Debris removal has been found to be twice as expensive as any other post-disaster activity and it is also the most time consuming. As a result, the Federal Emergency Management Agency is pressuring states to limit cleanup expenses.

The Clear Zone Improvement Program (C-ZIP) is an effort to incorporate highway function and safety with an improved aesthetic value. This system would maintain the mowed "recovery zone" along roadsides and establish attractive, native vegetation between the recovery zone and the forest zone. However, systems for planting and establishing native grasses and forbs in these areas are not currently available.

Desirable, native vegetation adds beauty to North Carolina's roadsides. Programs, like the NCDOT Wildflower Program, have drawn interest and compliments nationwide. An opportunity exists to expand roadside beautification with the Clear Zone Improvement Program (C-ZIP). Native grass filter strips may reduce runoff volume up to 76% (Rankins et al. 2001) and vegetative filter strips may filter suspended sediment and enhance adsorption of dissolved materials to plant and soil surfaces (Srivastava et al. 1998).

Certain herbicides are available that can assist in native plant establishment. Imazapic (Plateau) is an acetolactate synthase (ALS)-inhibiting herbicide in the imidazolinone class of chemistry. It has been used for several years in peanuts, and has recently been registered for use with native grass establishment. This compound is considered low-risk by the EPA due to a favorable toxicity profile and also has a relatively long soil residual. In previous research, imazapic plus 2,4-D was successfully used to establish native plants including little bluestem, big bluestem, indiangrass, Illinois bundleflower, purple prairieclover, and lance-leaf coreopsis (Washburn and Barnes 2000) while reducing weed cover to less than 5%. Imazapic has also been reported to control Japanese stiltgrass, an invasive species (Gover et al. 2003).

In contrast to native vegetation, non-native vegetation can invade roadsides increasing management expenses and decreasing visibility, safety, and aesthetics. It is estimated that management expenses and environmental damages and losses from invasive plants is approximately $34.7 billion per year in the United States (Pimentel et al. 2005). A single invasive plant can also overtake a complete ecosystem. An example of this can be found in California where yellow starthistle infests approximately 15 million acres of formerly productive grasslands, roadsides, and other sites (WeedRIC 2006).

Japanese knotweed (*Polygonum cuspidatum* Sieb. & Zucc) is a highly problematic perennial plant that has invaded roadsides in North Carolina and nearby states. This species was listed as one of the “100 least wanted” invasive species in the world by National Geographic magazine (McGrath 2005). Japanese knotweed forms dense, spreading monocultures on roadsides that reduces visibility, encroaches on the recovery area, and increases management expenses. Rakesh Chandran, Weed Specialist at West Virginia University, considers Japanese knotweed to be the most common and most problematic weed on West Virginia roadsides (personal communication).

Alligatorweed (*Alternanthera philoxeroides* (Mart.) Griseb.) is a perennial plant introduced from South America. This plant can completely cover drainage canals and ditches and may invade low-lying terrestrial sites. It can reduce water flow and increase flooding risks. It is relatively tolerant to glyphosate (Eberbach and Bowmer 1995), but has been controlled with imazapyr (Habitat). The alligatorweed flea beetle (*Agasicles hygrophila* Selman and Vogt) is a biological control agent for alligatorweed, but this species has not over-wintered in areas with climates colder than Wilmington, NC.

The objectives of this study were to (1) determine best planting methods, times, and seed rates for the establishment of native warm season grasses (NWSGs); (2) evaluate the effects of differing mulches in the establishment of NWSGs; and (3) Determine application timings, tank mixtures, and product formulations needed as a component of the integrated vegetation management approach for control of alligatorweed and Japanese knotweed.

2.0 Materials and Methods

2.1 NWSG Field Studies

2.1.1 Calibration, Weed Management, and General Maintenance. Seed rates and equipment were supplied by the North Carolina Department of Transportation (NCDOT). Greenscape and Tru-ax drills were calibrated using the "1/100th of an Acre Method" found in the North Carolina Agricultural Chemical Manual. Using the effective planting width of the drill and circumference of the drive wheel, calculations can then be made to determine how many revolutions of the drive wheel are needed to cover 1/100th of an acre. The planters were then filled with seed, lifted and seed were caught as the drive wheel was turned and weighed. Weights were multiplied by 100 for an acre rate, and settings were adjusted to accommodate the appropriate pure live seed. Calibration for the Finn hydro-seeder was conducted by the NCDOT using established practices. Little bluestem, big bluestem, indiangrass, Virginia wildrye, river oats, and sideoats grama required soaking prior to broadcasting to assure uniform output.

All locations and all trials had a burndown application of glyphosate at a rate of two to four quarts per acre; rates were dependant on weed profile. In the situations where weed populations were persistent and the native grasses had not emerged, glyphosate was sprayed or wicked depending on weed height or density and likelihood of presence of native species.

Plateau (imazapic) was applied at a rate of six ounces per acre (maximum rate 12 oz/acre/year) to the tolerant species (big bluestem, little bluestem, and indiangrass) in early spring and summer. Drive (quinclorac) was applied at four ounces per acre for tolerant species including tall fescue, orchardgrass, Virginia wildrye, big bluestem, little bluestem, sideoats grama, and switchgrass when control was not obtained by imazapic treatments.

For tall fescue, orchardgrass, and Virginia wildrye, wick applications of a fifty percent solution of glyphosate were applied if the weeds were taller than the NWSGs. 2,4-D was also applied to the cool season grasses at a rate of one pint per acre or aminopyralid (Milestone VM) at a rate of 5 ounces per acre to control any broadleaf weeds.

2.1.2 Planting Methods Study. The Planting Methods Study was initiated in fall 2007 and fall 2008 at three North Carolina research stations: Caswell (Kinston), Lake Wheeler (Raleigh), and Upper Piedmont (Reidsville). Each location and year had two planting dates (fall and spring), three planting methods (SBE, Tru-ax, Hydro), and nine grasses. Big bluestem (Andropogon gerardii), little bluestem (Schizachyrium scoparium), indiangrass (Sorghastrum nutans), switchgrass (Panicum virgatum), sideoats grama (Bouteloua curtipendula), river oats (Chasmanthium latifolium), Virginia wildrye (Elymus virginicus), orchardgrass (Pactylis glomerata L.), and tall fescue (Schedonorus phoenix). All grasses were planted as monocultures in this study.

The three planting methods included a conventional Greenscape S.B.E. drill, a no-till Tru-ax drill, and a Finn hydro-seeder. Studies were arranged in a split plot design with six by twenty-five foot plots and twenty-five foot buffers between the nine grass species (Figure 1). Each specie being in a three treatment, four replicate factorial arrangement. Prior to planting, hydro-seeded and conventional (SBE) plots were tilled with a six foot wide rotary tiller. Upon completion of planting, hydro-seeded plots were mulched with wheat straw; other planting methods did not receive any straw mulch. Visual ratings were taken on a 0 to 100% scale (0% being no emergence, 100% complete coverage of grass). Data was combined when possible and processed using a Mixed Procedure with season nested within replication, and means separated using Fisher's Protected LSD in SAS (version 9.1).

2.1.3 Seeding Rate Study. The Seeding Rate study was conducted to determine if increased seeding rates could provide a quicker establishment and denser cover for NWSGs. Species listed above excluding tall fescue were included. This trial was repeated twice over time at Caswell Research Station in Kinston, North Carolina; being planted in fall 2007 and fall 2008. A split plot design was used with each species designated to its own range with twenty-five foot buffers between species. Each range was setup as a four treatments, four replicate factorial and individual plot dimensions were twelve by twenty-five foot. Seeding treatments were based on the NCDOT established seeding rate as shown in Table 2, also included are recommended rates from seed producers. Treatments were 1x, ½x, 1 ½x, and 2x the NCDOT rates.

Due to strong weed pressure and severe drought the fall 2007 planting date was abandoned and a modified approach was utilized for the fall 2008. Planting method was changed from the no-till, Tru-ax drill to the conventional, Greenscape S.B.E, providing the ability to cultivate and the study area was fumigated to provide additional weed control. Ratings were taken to document establishment on a 0 to 100% scale (0% being no emergence, 100% complete coverage of grass). Using SAS (version 9.1), a simple Proc GLM was conducted per species and means separated using Fisher's Protected LSD.

2.1.4 Hydro-seeding Studies. Initiated April 20th and May 5th, 2009 at the Upper Piedmont Research Station and Cherry Farm in Goldsboro, hydro-seeding studies were conducted to observe grass establishment with different mulches. A blend of three native grasses consisting of big bluestem, little bluestem, and indiangrass were used. These grasses were selected for their tolerance to Plateau (imazapic) which was applied to one half of each plot within a week of initiation at a rate of twelve ounces per acre. Each location was tilled prior to planting to provide a good seed bed and the laid off in a Latin Square design. Plots dimensions were thirty feet by ninety feet with ten foot alleys between all plots. Mulch treatments used were wheat straw, pine straw, hydro-mulch, and no mulch.

Hydro seeder tank mix consisted of 1,000 gallons water, twenty-five pounds big bluestem, fifteen pounds little bluestem, and fifteen pounds indiangrass. All plots were seeded at an application volume of 1,400 gallons per acre. For the hydro-mulch plots, three bails of hydro-mulch were added to solution to give each plot a uniform coverage. Pine or wheat straw plots were mulched using approximately thirteen bails per plot or until adequate coverage was obtained.

Data was subjected to analysis of variance using a Mixed Procedure for interactions between mulches and imazapic application. Furthermore, Pearson's Correlation was also utilized to evaluate results with weed pressure versus NWSG establishment.

2.1.5 Month, Stratification and Mulching Study. A broad scope study was implemented April 15, 2010 to evaluate different planting regimes, mulching, and seed dormancy. Prior to initiation, a burndown application of glyphosate and imazapic was applied to the site and the location was setup with a split-split plot design and one half of every plot was broadcasted with hardwood mulch. Treatments included mulch, no mulch, seed lots with known dormancy, and seed lots with little dormancy. Between April and August, simulated hydro-seeded plantings occurred monthly, precisely on the fifteenth of each month. Species included big bluestem, little bluestem, indiangrass, sideoats grama, and purple top. Data was subjected to a basic split plot analysis utilizing SAS (version 9.1).

2.2 NWSG Greenhouse Studies.

2.2.1 Planting Depth Study. Four inch square pots containing a sandy loam medium were seeded July 5, 2010 to evaluate the effect of planting depth on big bluestem, little bluestem, Virginia wildrye, and tall fescue. Individual pots contained approximately 40 seeds per specie. Seeds were planted at depths of 0 (surface of soil), 0.25, 0.5 and 1 inch below the soil surface. Pots were then placed inside a greenhouse and approximately 0.25 inch of water was applied daily using overhead sprinklers.

2.2.2 Irrigation Studies. Two irrigation studies were initiated August, 2010 at the NCSU Weed Control Laboratories. The first study was planted in four inch square pots containing approximately 50 seeds were placed outside to receive natural rainfall and were evaluated for natural survivability. Rainfall events for the first 5 days were: 0.37, 0, 0.10, 1.20, 0.09 inch. Following these rainfall events, no additional rain occurred for the next 18 days. Following this natural drought, hand watering was started on day 24; 0.25 inch of water was applied during the week with no additional water during the weekend (five days on / two days off).

The second irrigation study was conducted in a greenhouse to ensure a controlled environment. Pots were seeded as described above and received a 0.25 inch of irrigation per day using overhead sprinklers. This watering pattern continued for five days then the water was turned off for 18 days. On day 24 the irrigation system was again turned on applying 0.25 inch of water per day. The system was allowed to run seven days per week.

2.3 NWSG Laboratory Studies

2.3.1 Tetrazolium (TZ) Study. An experiment conducted under Association Official Seed Analysts (AOSA) guidelines to determine viability of seeds. Study was conducted in winter 2009 in the NCSU

Seed Laboratory to evaluate six species for viability. Species included orchardgrass, switchgrass, indiangrass, riveroats, big bluestem, and little bluestem. In preparation of study, individual species were mixed thoroughly to obtain a homologized subsample, and 100 seeds were placed on a moistened germination paper, rolled, and labeled for separation. The rolled germination papers were then placed in a sealed container and relocated to a chamber at a constant temperature of 77 F (25 C) for 16h to imbibe seeds for cellular expansion. Once complete, seeds were removed, bisected according to standard methods , one half of each seed was placed in TZ solution buffered to a pH of 7.0, and moved to an ADW oven at 77 F (25 C) for 2 to 4h. Because of differences in switchgrass seed structure, the seed coat was pierced with a needle and placed directly in to TZ solution without cutting. Time in solution was dependent upon specific species requirements based on the Tetrazolium Testing Handbook (2009). After removal from the oven, seeds were rinsed three times with de-ionized (DI) water to remove TZ solution and nonessential seed structures. Finally, seeds were examined under a microscope to determine the viability of essential structures (Image 1).

2.3.2 Germination Table. An experiment to evaluate the optimal germination temperatures for selected species was conducted twice in 2009. Species included two NWSG species (switchgrass and indiangrass) in comparison to Virginia wildrye, a cool season standard. Prior to initiation, 20 deep Petri dishes were sterilized, filled with one cup sterilized river rock, amended with DI water, and a 3¼" AOSA certified blotter paper was placed atop gravel to contain seed. Seeds were then exposed to temperatures of 65, 74, 82, 90, or 98 degrees Fahrenheit (17, 23, 27, 32, or 36 C) utilizing a patented gradient table in the NCSU Seeds Laboratory. Species were kept segregated using a designated Petri dish per temperature and 100 seeds were placed within the each dish and covered with a watch glass to prevent evaporation. Each jar was representative of one replication. Seeds were observed for a period of two weeks, daily records of germination were taken; all germinated seeds were then removed. 500 ml flasks of DI water were placed in each temperature lane to be used in refilling experimental containers to maintain proper water level and water temperature. Light was provided by fluorescent overhead bulbs set for a 16 hour (h) light 8 h dark regime with a light intensity of 30 µmol m-2s-1. Due to the small size of the gradient table and limited availability, no other species were tested. Data were pooled and charted with SigmaPlot 11.0.

2.4 Japanese Knotweed Control.

Research was conducted in North Carolina and Virginia to determine the response of Japanese knotweed to selected aquatic herbicides. Treatments were made in September 2008 and 2009 with a CO_2 pressurized backpack sprayer equipped with handgun calibrated to deliver an application volume of 30 GPA. Chemical treatments included 0.38 lb ai/A flumioxazin, 2 lb ai/A fluridone, 1.8 lb ai/A glyphosate, 1.5 lb ai/A imazamox, 1 lb ai/A imazapyr, 0.0875 lb ai/A penoxsulam, glyphosate plus imazamox, glyphosate plus imazapyr, glyphosate plus penoxsulam, flumioxazin plus fluridone, and non-treated control for comparison. All experiments were set up in a randomized complete block design and visual control, height, and population counts were taken at 8, 12, and 21 months after treatment (MAT).

2.5 Alligatorweed Control.

Greenhouse trials. Greenhouse experiments were initially conducted to evaluate the response of alligatorweed to broad rate ranges of imazamox and penoxsulam. Alligatorweed was transplanted into 4 inch square pots and allowed to establish. Plants of uniform height and growth stage (8 to 12 inch height) were selected for each trial. The imazamox trial included rates of 0.0625, 0.125, 0.1875, 0.25, and 0.5 lb ae/A. The penoxsulam trial included rates of 0.00446, 0.0089, 0.0223, 0.0446, 0.089, and 0.178 lb ai/A. A non-treated control was included in each trial and 0.25% non-ionic surfactant (NIS) was included with each herbicide treatment. Each trial included 4 treatment replications. Alligatorweed control was visually rated at 1 month after treatment (MAT) on a 0 to 100 scale, where zero is no control and 100 is complete death. At 1 MAT, alligatorweed above-ground biomass was harvested, dried, and weighed for dry weight determination. Greenhouse data was subjected to regression analysis using the logistic equation $y = a/1+(x/x_o)^b$. The imazamox trial was conducted three times and the penoxsulam trial was conducted twice.

Field trials. The first field trial evaluated multiple rates of each herbicide in comparison to 0.3 lb ae/A imazapyr and a non-treated control. Imazamox rates were 0.094, 0.188, 0.376, and 0.752 lb ae/A, while penoxsulam was applied at 0.0876, 0.175, and 0.35 lb ai/A. Treatments were replicated three times and the trial was conducted once in 2007 and twice in 2008. Control was rated on a 0 to 100% scale at 1 and 2 MAT. Data from 1 MAT was subjected to regression analysis as described for greenhouse trials.

Another field experiment was conducted to compare 0.18 lb ae/A imazamox and 0.35 lb ai/A penoxsulam to 2 lb ae/A glyphosate, 0.3 lb ae/A imazapyr, and 4 lb ae/A triclopyr. Each treatment was replicated 3 times and a non-treated control was included for comparison. The trial was conducted three times during 2007 and 2008 and was rated at 1 and 2 MAT. Trial A in 2008 was lost prior to 2 MAT due to environmental conditions. Data was subjected to analysis of variance and Fisher's Protected LSD ($p < 0.05$) was used for mean separation.

3.0 Results

3.1 NWSG Field Studies

3.1.1 Planting Methods Study. Due to extreme variances in data, most likely caused by weather differences, locations could not be combined. Therefore, data was processed per site, per specie and presented as so with species combined when appropriate. Due to no success of NWSG establishment, the Lake Wheeler location is not presented.

The greatest success was observed at the Caswell research station in Kinston, NC. Big bluestem, little bluestem, indiangrass, switchgrass, and tall fescue all had excellent establishment after year three; however, it is important to note that no emergence was observed in year one or year two. The four NWSG species all showed the same statistical trend with a significant difference in planting season and only seven percent establishment in fall plantings compared to 68% in spring (Table 2). However, no statistical differences were observed between planting methods (Table 3). Four years after planting (YAP), these trends remained constant but with an increased average establishment of four percent (data not shown) across all NWSGs. In comparison to the cool season standard of tall fescue, data was inversely related as would be expected with 53% establishment in the fall planting compared to 6% in the spring (data not shown). However, all methods were statistically similar with a p-value of 0.3259.

In Reidsville, NC at the Upper Piedmont Research Station, data were similar to what was observed in Kinston. At four YAP, two notable differences (Tables 4 and 5) were: (1) little bluestem had a much higher success rate in the fall planting with 25% establishment compared to only five in the spring planting, and (2) there were differences in planting methods for indiangrass. This result is likely due to the low success rate of the hydro-seeded plots with only 53% establishment compared to 82 to 93% establishment with other methods. Tall fescue had no differences regardless of season or method with 95% establishment. Also, though populations were too low to collect data, the Reidsville location is the only site where two cool season grasses, orchardgrass and Virginia wildrye, persisted through the entire study period.

3.1.2 Seeding Rate Study. At 2.5 YAP, only little bluestem, indiangrass, switchgrass, and orchardgrass were present. Little bluestem and indiangrass had no differences between seeding rates and achieved an establishment range of 54 to 78%. Switchgrass and orchardgrass shared a trend of statistical difference between the 1X and 2X recommended seed rates with approximately 25% higher coverage in the 2X plots. However, the 0.5X and 1.5X seeding rates were similar to all (Figure 2). These data may indicate that the NCDOT established seeding rates may need to be reconsidered and the use of recommended producer rates be implemented to ensure economic sustainability (Table 2).

3.1.3 Hydro-seeding Studies. In the first set of hydro-seeding studies in Reidsville and Goldsboro, the best observed establishment was with pine straw mulch at 84% whereas hydro-mulched only had an average of 45% establishment. However, these values were not significantly different (Table 7).

Furthermore, after Pearson's test of correlation was utilized, there was indeed a dramatic correlation between weed pressure and NWSG establishment (data not shown) as can be seen in Image 3.

In the last hydro-seeding study (Kinston), there were no statistical differences in NWSG establishment with or without wheat straw. These results indicate that any potential allelopathic effects that wheat straw may possess did not hinder the establishment of the NWSGs. However, it was observed that erosion was a cause of concern whereas unplanted alleys beside un-mulched plots had significant emergence of NWSGs.

3.1.4 Stratification and Mulching Study. There were no differences between stratified and non-stratified seed lots. Therefore, these data were pooled and analysis of variance was used to evaluate differences between mulch and no mulch. Results of this analysis are shown in Figure 3, indicating there were significant differences in mulching and planting date. Mulching prior to planting caused an adverse effect on the total establishment with approximately a 25% decrease, most likely due to the lack of soil/seed contact. Furthermore, establishment decreased dramatically after the May 15th planting date indicating a narrow window of opportunity for successful establishment of NWSGs.

3.2 NWSG Greenhouse Studies

3.2.1 Planting Depth Study.

Emergence was greatest at the surface planting depth for all species except little bluestem and tall fescue. Emergence at depths of and greater than 0.5 inch were in most cases significantly lower than the shallower depths. The exception to this was side oats with no significant differences in planting depth. With all other species, increased planting depth resulted in lower total emergence, with no differences in rate of emergence (Figure 4).

3.2.2 Irrigation/Non-irrigation Study.

Outdoor Study 1. Big bluestem and tall fescue emerged on day 4, little bluestem on day 5 and side oats on day 7. Virginia wildrye did not emerge during this study. No additional emergence was observed after day 7. All species were dry and dead on the day 14 evaluation. Following the heavy natural rainfall a thick hard crust developed on all pots, this may have prevented additional emergence. After the supplemental irrigation started, this crust persisted. No additional plants emerged at 42 days after planting. Crusting of many soils in North Carolina can reduce emergence of many species including large seeded crops. With many small seeded species the seed may germinate, but do not have the energy to penetrate the crust and emerge as viable plants.

Indoor Study 2. Big bluestem, little bluestem, and tall fescue emerged on day 3, side oats, and Virginia wildrye emerged on day 6. After the irrigation events were discontinued, all species were dry and dead at the day 14 evaluation. The irrigation was again turned on, and seedlings started to emerge approximately 4 days later. On the 28 day evaluation all species had additional plants emerged. Most likely because of low intensity, frequent watering, no crust formed on these pots.

3.3 NWSG Laboratory Studies

3.3.1 Germination Study. Due to the limited range of temperatures on the germination table, there are no statistical differences between temperatures or species. However Virginia wildrye, indiangrass, and switchgrass had the same optimal germination range between the temperatures of 78 to 83F (Figure 5). The decreasing germination rates on either side of the parabola apex further demonstrate how proper temperatures play an important role in germination of these species. Soil temperatures should be determined before planting to ensure the best possible growing conditions.

3.3.2 Tetrazolium Study. Large differences were present in the reported and the actual germination/viability from the original seed lots used (Table 8). The range of the differences between the reported and actual germination was 21 to 64%. These differences were due to either aging of the seed lots or, in few cases, seeds may have been harvested too early and were not mature. Seed viability as determined by TZ testing is a measure to determine if a seed is biologically active and has the potential to germinate. Seed with extreme dormancy issues may eventually germinate, but not have a positive TZ test result. Immature seed or seed stored under improper conditions will also produce a negative TZ test result. In many cases, the dormancy and germination requirements may prevent a seed from producing a viable plant. The results of a TZ test should be closely related to the reported germination percent and in some cases actually are higher. The results of this study show that viability is much lower than the reported germination, and should be considered when determining planting rates. Seed lots with low viability should be seeded at higher rates. If the dormant seed eventually germinate and produce viable plants, the overall objective of quick stand establishment may not be realized.

New seed lots were purchased for all plantings after fall 2008. These seed lots were purchased from a different supplier and germination percentages were consistent for both the reported and actual germination (data not shown).

3.4 Japanese Knotweed Control.

Japanese knotweed control at 8 months after treatment (MAT) was at least 90% with all treatments containing glyphosate or imazapyr, except penoxsulam plus glyphosate at 75% (Figure 9). At 21 months after treatment, greatest control was observed with the same treatments and control ranged 72 to 93% (Figure 10). Japanese knotweed cover and number or sprouts at 12 MAT were similarly affected. Coverage was 1.3% or less with glyphosate alone, imazamox plus glyphosate, imazapyr alone, or imazamox plus imazapyr. The number of sprouts was 3.7 per m^2 with these treatments. The untreated control averaged 25% coverage and 23.7 sprouts per m^2 (Table 10).Japanese knotweed height at 21 MAT was 38 inches or less with these treatments compared to the untreated control height of 72 inches (Figure 11). Based upon these results, glyphosate alone, imazamox plus glyphosate, imazapyr alone, or imazamox plus imazapyr can provide long term control of Japanese knotweed. While not usually significantly different from the other three treatments, imazapyr plus glyphosate usually provided the numerically greatest control.

3.5 Alligatorweed Control.

Greenhouse. In the Clearcast (imazamox) trial, the regression curves for alligatorweed control and dry weight were highly significant and well correlated to the data (Figure 12). Control as indicated by the regression curve exceeded 80% at an imazamox rate of 0.088 lb ae/A. Control neared 100% with 0.5 and 1 lb ae/A imazamox. The dry weight regression curve indicated that dry weight was reduced 80% or more by imazamox rates of 0.09 or greater. Similar to control data, very little dry weight remained at 0.5 and 1 lb ae/A imazamox.

In the penoxsulam (Galleon) trial, the regression curves were also highly significant and well correlated to the data (Figure 12). Control exceeded 80% with rates above 0.065 lb ae/A as indicated by the regression curve. Near 100% control was achieved with the highest rate evaluated of 0.178 lb ai/A penoxsulam. The dry weight regression curve indicated that reductions of 80% or greater were correlated with penoxsulam rates of 0.039 lb ai/A or greater. Less than 10% of alligatorweed dry weight remained after treatment with 0.089 or 0.128 lb ai/A.

Field. In the first field experiment, alligatorweed was controlled at 1 MAT with both imazamox and penoxsulam (Figure 13). The regression curves indicated that 80% control was correlated with 0.2 lb ae/A imazamox or 0.33 lb ai/A penoxsulam. Both curves peaked at 91% control with the highest rates evaluated, 0.75 lb ae/A imazamox and 0.7 lb ai/A penoxsulam. In two of the field trials, 0.75 lb ae/A imazamox provided equivalent control at 1 MAT to 0.3 lb ae/A imazapyr, but control with 0.7 lb ai/A penoxsulam was only equivalent to imazapyr in one of the three trials.

Across the three trials, control with imazamox and imazapyr became erratic after 1 MAT (data not presented). In 2007, control at 2 MAT was 91±6% with 0.75 lb ae/A imazamox, 91±5% with 0.7 lb ai/A penoxsulam, and 99±1% with 0.3 lb ae/A imazapyr. In 2008, control at 2 MAT dropped to 65% or lower with all rates of imazamox or penoxsulam, which was significantly lower than the 99% control obtained again with imazapyr.

In the second field experiment, control with glyphosate, imazamox, imazapyr, penoxsulam, and triclopyr was 80 to 93% at 1 MAT in 2007 (Table 11). In the 2008A trial at 1 MAT, control was 80 to 98% with imazapyr, penoxsulam, and triclopyr, but control only exceeded 80% in the 2008B trial with imazapyr. At 2 MAT, only imazapyr provided acceptable control in both the 2007 and 2008B trial. Control with other products did not exceed 12% in the 2008B trial.

4.0 Conclusions

4.1 NWSG

There are many challenges to the establishment of NWSGs in North Carolina. For instance, the amount of inputs and associated costs in combatting unwanted vegetation can be expensive (Table 9). All NWSG species tested required a minimum of two herbicide applications per year including the use of multiple herbicides and modes of action (MOA). With the increasing instances of herbicide resistance, it is essential to rotate MOAs. For example, the use of imazapic (Plateau) was an integral part of the establishment of tolerant species. However, acetohydroxy acid synthase (ALS) resistant horseweed *(Conyza canadensis*) became the dominate weed species in Kinston, forcing the use of additional spray application and differing chemical classes. In comparison to the cool season standards of tall fescue and orchardgrass, very few herbicide applications were needed due to the fast and uniform establishment (Image 3).

Poor overall NWSG establishment success may be attributed to several factors. First and foremost, planting date is essential for any success and the window of opportunity is particularly small. In Figure 3, it is apparent that planting any time after the month of May can result in poor or even no establishment. Though no research was conducted as to how early NWSGs can be planted, establishment has been observed in plantings made November through May whereas little or no establishment was seen June through October.

Second, rainfall and/or irrigation are also very important as can be seen in greenhouse studies and figure 5. The Upper Piedmont Research Station had no measurable rainfall for nearly a year after the 2008 spring planting. Even though the Caswell Station in Kinston did receive more rainfall, the original seed lots did not germinate for nearly three years. Establishment in these plots may be attributed to the high seed rates established by the North Carolina Department of Transportation. Recommended seed rates in (Table 2) would have likely not been enough to counteract the actual viability rates discovered in the TZ testing (Table 8).

Third, a population shift may occur over time when multiples species are present (Figure 8). At 1 YAP, NWSG species were sporadically established but present nonetheless. However, after two years, indiangrass outcompeted both little and big bluestem, illustrating that early and uniform establishment is essential. This resulted is a monospecific stand which leads to more inquires regarding seed rates and financial sustainability seen in Table 2. Also, the USGS Plant Profile list big bluestem and indiangrass as having invasive capabilities which indicate a potential concern for further population shifts or spread outside the boundaries where they are planted. Lastly, past research from Kansas State University, has shown that indiangrass has the potential to outcompete big bluestem with any additional sources of irrigation and fertilization (Silletti and Knapp 2001).

In conclusion, large scale establishment of NWSGs on North Carolina road sides will be economically and practically challenging. Same year establishment was rarely seen in our trials, thus leaving bare and/or weedy areas creating opportunities for erosion and /or the establishment of nonnative invasive species. Furthermore, once established, NWSG stands were often patchy and sporadic which may not be considered acceptable or aesthetically pleasing (Image 3). NWSG planting should be done under optimum conditions and only during optimum planting periods in order to get NWSG establishment as quickly as possible. Thus, individual construction projects should be critically evaluated for NWSG planting depending upon whether ideal establishment conditions will be present.

4.2 Japanese Knotweed and Alligatorweed Control

At 7 MAT, Japanese knotweed control with glyphosate, imazapyr, glyphosate plus imazamox, and glyphosate plus imazapyr was at least 93%. Glyposate plus penoxsulam controlled the plant 75%. Control did not exceed 20% with other treatments. Given the relatively low cost of the post-patent products glyphosate and imazapyr, these two herbicides applied together or individually should be expected to provide cost effective Japanese knotweed control.

Alligatorweed was more sensitive to imazamox and penoxsulam in the greenhouse than in the field. This is common with foliar-applied herbicides and caution should always be observed in interpreting greenhouse results. In the field at 1 MAT, calculated alligatorweed EC_{80} was 0.2 lb ae/A for imazamox or 0.33 lb ai/A for penoxsulam. Control after 1 MAT was erratic with imazamox and penoxsulam and generally not equivalent to imazapyr. Imazapyr provided excellent control of alligatorweed in almost every trial conducted. It remains the standard alligatorweed control herbicide and since imazamox and penoxsulam have the same mode of action there is no rotational benefit with these products. Additional research should be conducted to determine why alligatorweed does not respond as consistently to imazamox and penoxsulam as it does to imazapyr.

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6.0 Appendices

Table 1. NWSG Field Studies and Initiation Dates.

Trial ID	Location	Date Initiated
METHODS-1 (FALL)	Kinston	11/30/2007
METHODS-1 (FALL)	Lake Wheeler	11/28/2007
METHODS-1 (FALL)	Reidsville	12/3/2007
METHODS-1 (SPG)	Kinston	4/15/2008
METHODS-1 (SPG)	Lake Wheeler	4/17/2008
METHODS-1 (SPG)	Reidsville	4/22/2008
METHODS-2 (FALL)	Kinston	10/27/2008
METHODS-2 (FALL)	Lake Wheeler	11/12/2008
METHODS-2 (FALL)	Reidsville	10/15/2008
METHODS-2 (SPG)	Kinston	4/16/2009
METHODS-2 (SPG)	Lake Wheeler	4/13/2009
METHODS-2 (SPG)	Reidsville	4/14/2009
SEED RATE STUDY	Kinston	10/29/2008
HYDROSEEDING-1	Reidsville	4/20/2009
HYDROSEEDING-1	Goldsboro	5/5/2009
HYDROSEEDING-2	Kinston	12/16/2009
MULCH/MONTH	Kinston	4/15/2010
PLATEAU TOLERANCE	Wallace	5/10/2011

	Big Blue	Little Blue	Indian	Switch	Side oats	River Oats	Va Wildrye	Orchard	Fescue
TRU	101	201	301	401	501	601	701	801	901
SBE	102	202	302	402	502	602	702	802	902
HY	103	203	303	403	503	603	703	803	903
SBE	104	204	304	404	504	604	704	804	904
TRU	105	205	305	405	505	605	705	805	905
HY	106	206	306	406	506	606	706	806	906
HY	107	207	307	407	507	607	707	807	907
TRU	108	208	308	408	508	608	708	808	908
SBE	109	209	309	409	509	609	709	809	909
HY	110	210	310	410	510	610	710	810	910
TRU	111	211	311	411	511	611	711	811	911
SBE	112	212	312	412	512	612	712	812	912
TRU	113	213	313	413	513	613	713	813	913
SBE	114	214	314	414	514	614	714	814	914
HY	115	215	315	415	515	615	715	815	915
SBE	116	216	316	416	516	616	716	816	916
HY	117	217	317	417	517	617	717	817	917
TRU	118	218	318	418	518	618	718	818	918
SBE	119	219	319	419	519	619	719	819	919
TRU	120	220	320	420	520	620	720	820	920
HY	121	221	321	421	521	621	721	821	921
SBE	122	222	322	422	522	622	722	822	922
HY	123	223	323	423	523	623	723	823	923
TRU	124	224	324	424	524	624	724	824	924

Figure 1. Planting methods plot map example. Grey plots represent fall planting.

Table 2. Recommended seed rates compared to North Carolina Department of Transportation established rates and cost analysis calculated in pounds of pure live seed (PLS) per acre (Ac).

Grass Specie	Cost (PLS)	Recommended Seed Rate (PLS/Ac)	DOT Established Seed Rates (PLS/Ac)	Cost Analysis		
				Min	Max	Difference
Big Bluestem	$9.00	6	7.67	$54.00	$69.03	$15.03
Little Bluestem	$10.00	3.4	8	$34.00	$80.00	$46.00
Indiangrass	$8.00	4.5	8.1	$36.00	$64.80	$28.80
Switchgrass	$14.00	2	7.6	$28.00	$106.40	$78.40

Image 1. Tetrazolium analysis illustration. Highlighted/stained embryo indicates viability of seed. Most viable illustrated on left and decreases to no viability on right.

Table 3. Three years after planting establishment data based upon planting season for Caswell Research Station, Kinston, NC. Significance determined using Fisher's Protected LSD ($P \leq 0.05$). Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

	Fall Est	Spring Est	P-value	Significantly Different
	----------------------% Coverage------------------			
Big Bluestem	13%	70%	<0.0001	YES
Little Bluestem	1%	69%	<0.0001	YES
Indiangrass	7%	74%	<0.0001	YES
Switchgrass	8%	57%	<0.0001	YES
SP*SEASON	7%	68%	0.0327	YES

Table 4. NWSG establishment bases upon planting method with pooled season data at the Caswell Research Station, Kinston, NC, three years after planting. Significance determined using Fisher's Protected LSD (P≤0.05). Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

	Hydro Est	**SBE Est**	**Tru-ax Est**	**P-value**	**Significantly Different**
	------------------------------% Coverage-----------------------------				
Big Bluestem	40%	35%	50%	0.1659	NO
Little Bluestem	35%	33%	39%	0.4615	NO
Inidangrass	38%	39%	45%	0.3732	NO
Switchgrass	29%	31%	38%	0.4611	NO
SP*METHOD	36%	35%	43%	0.9294	NO

Table 5. NWSG establishment four years after treatment based upon planting season at the Upper Piedmont Research Center, Reidsville, NC. Significance determined using Fisher's Protected LSD (P≤0.05). Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

	Fall Est	**Spring Est**	**P-value**	**Significantly Different**
	---------------% Coverage--------------			
Big Bluestem	6%	22%	0.2166	NO
Little Bluestem	25%	5%	0.0070	YES
Indiangrass	70%	81%	0.2937	NO
SP*SEASON	na	na	0.0025	YES

Table 6. NWSG establishment four years after planting based upon planting method with pooled season data at the Upper Piedmont Research Station, Reidsville, NC. Significance determined using Fisher's Protected LSD (P≤0.05). Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

	Hydro Est	**SBE Est**	**Tru-ax Est**	**P-value**	**Significantly Different**
	--------------------% Coverage----------------				
Big Bluestem	9%	10%	23%	0.2446	NO
Little Bluestem	22%	8%	15%	0.2504	NO
Indiangrass	53%	82%	93%	0.0082	YES
SP*METHOD	na	na	na	0.0074	YES

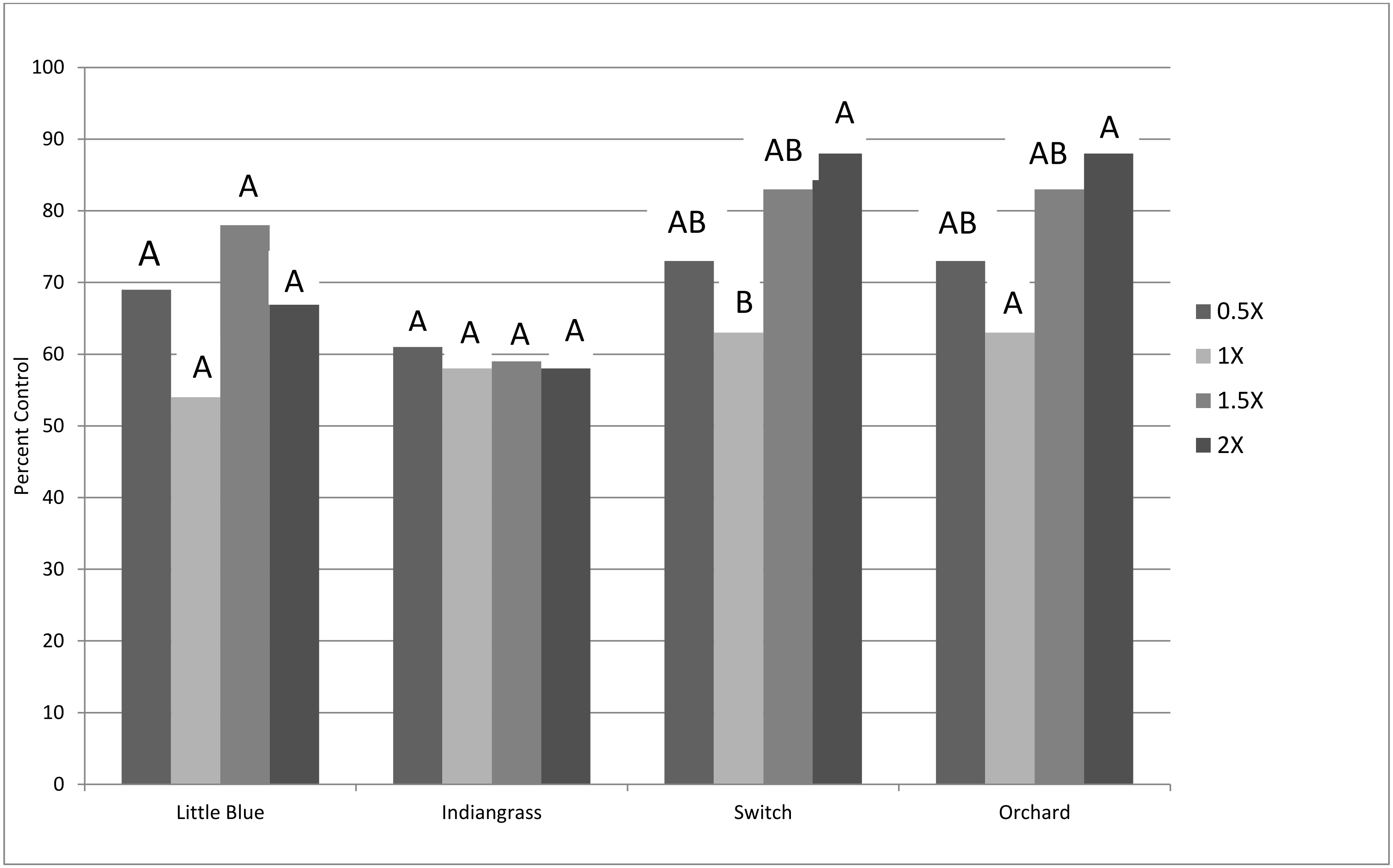


Figure 2. Percent NWSG and comparative standard (Orchardgrass) establishment (y-axis) based upon differing seed rates 2.5YAP. Significance determined using Fisher's Protected LSD (P≤0.05) and illustrated within columns; columns with like letter are not significantly different. Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

Table 7. Hydro seeding study for NWSG establishment with three different mulches and further evaluated with a split plot application of imazapic. Significance determined using Fisher's Protected LSD ($P \leq 0.05$). Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

Treatment	Establishment	Significantly Different
No-Mulch	59%	NO
Hydro-Mulch	45%	NO
Pine Straw	84%	NO
Wheat Straw	77%	NO
Imazapic	70%	NO
No-Imazapic	63%	NO

Image 2. Hydro-seeding illustration of weed pressure and NWSG establishment correlation, imazapic application applied left of dashed line.

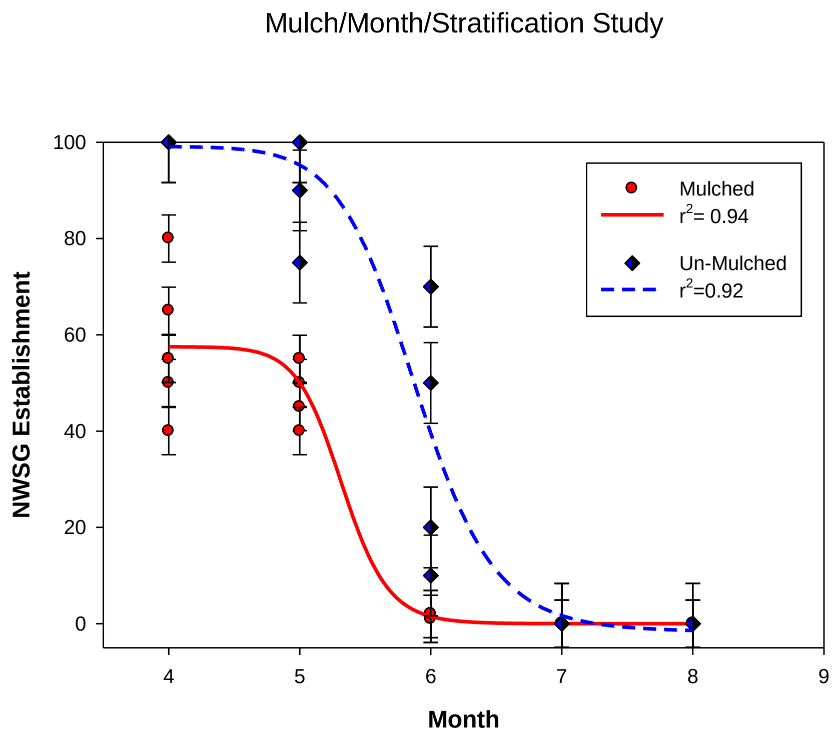


Figure 3. The effect of planting timing in correlation to mulching. Data points were calculated and a line fitted using a non-linear regression with the formula: f1 = min + (max-min)/(1 + (x/EC50)^(-Hillslope)).

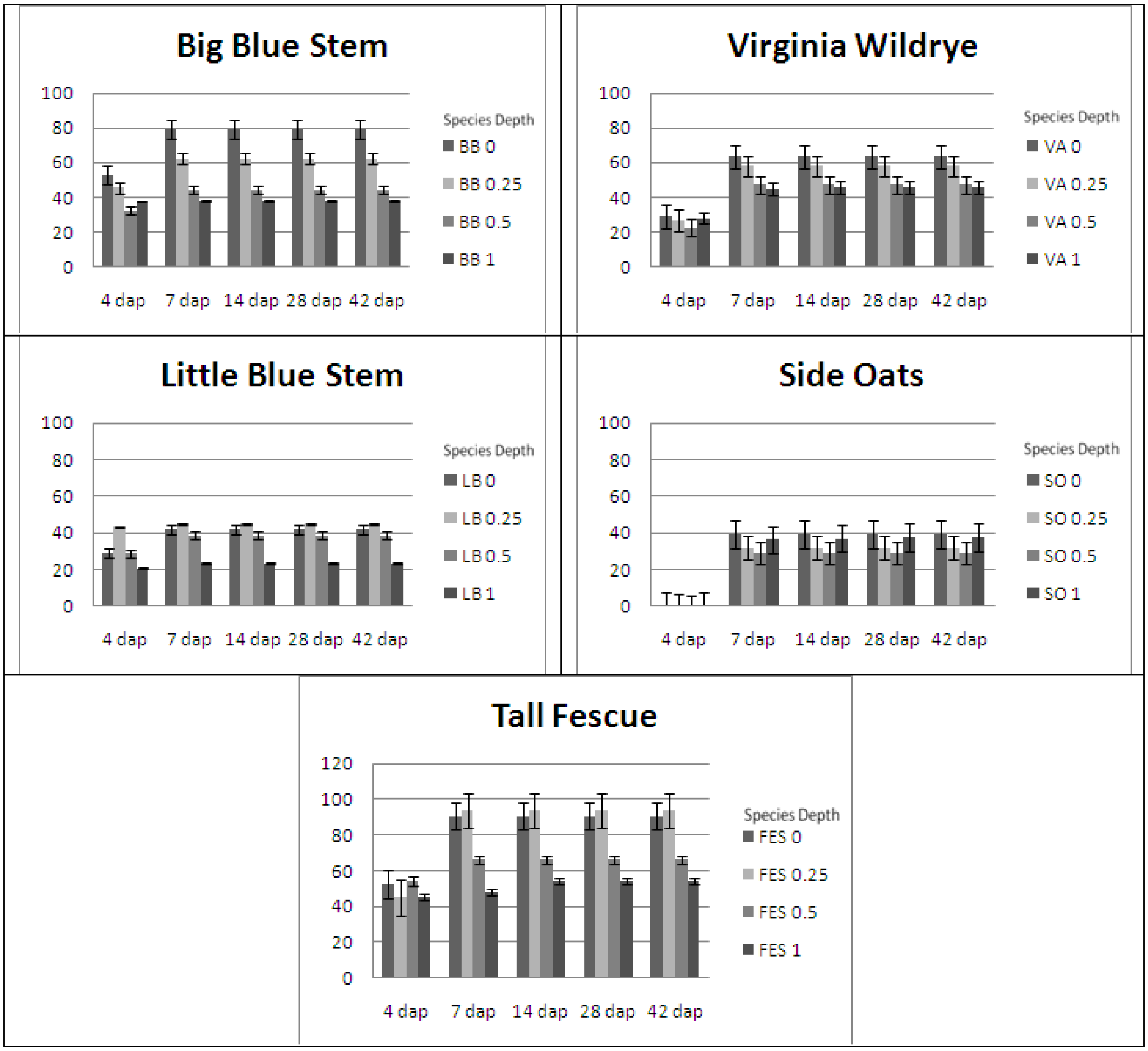


Figure 4. **Emergence of selected grasses at various depths by species.**

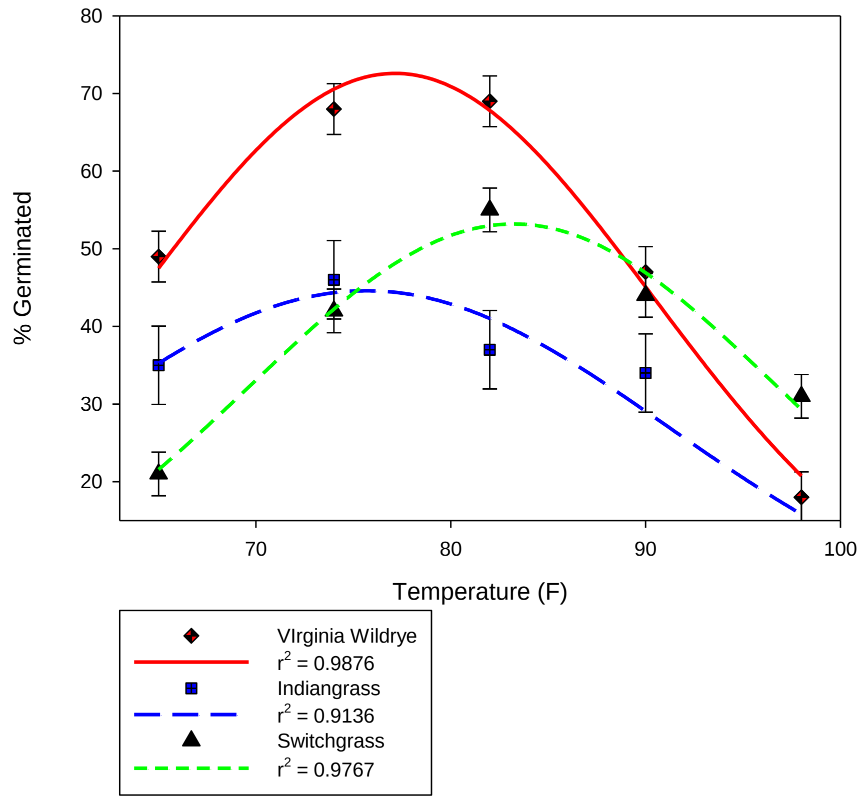
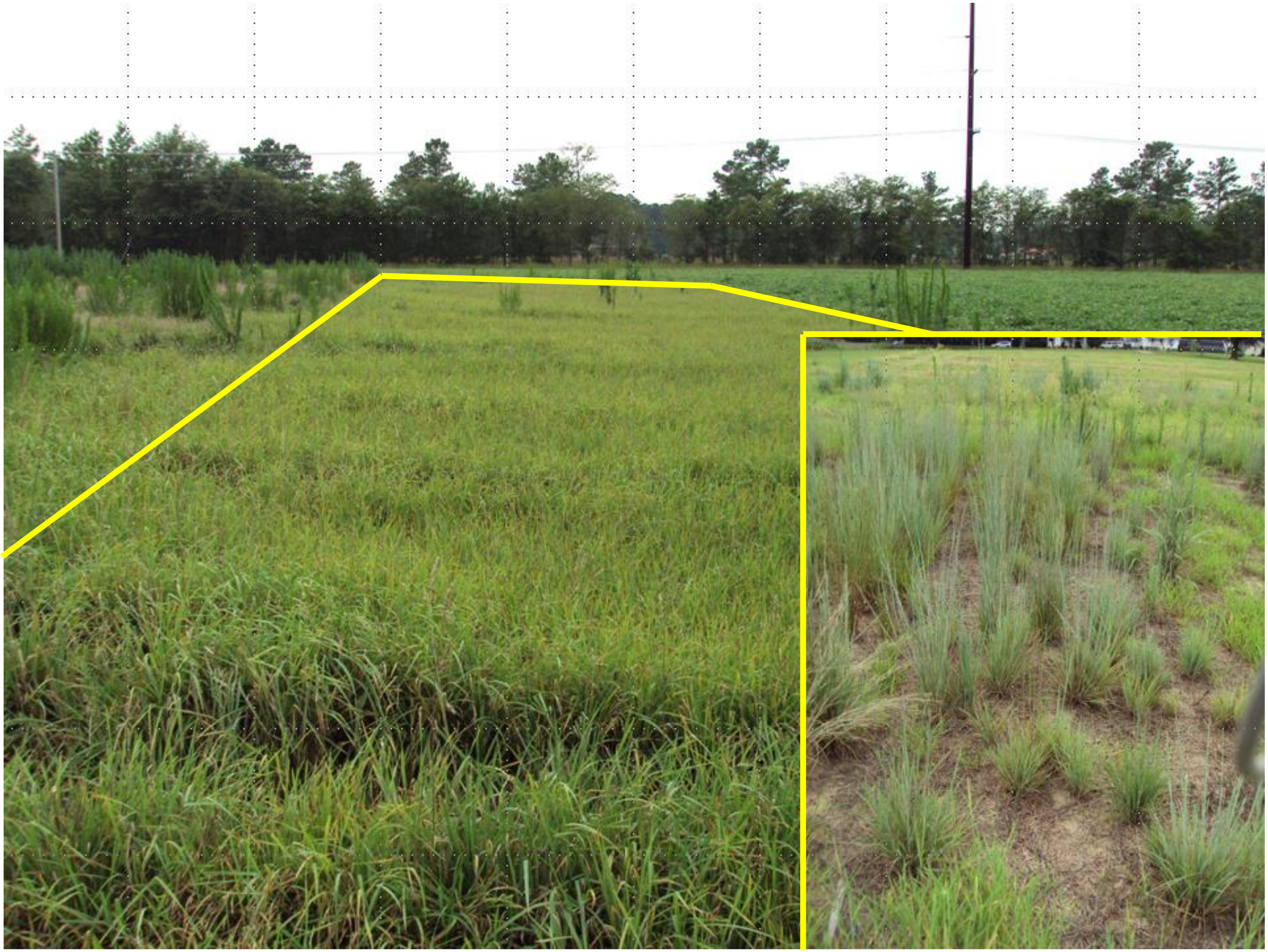


Figure 5. Germination table data for Virginia Wildrye, Indiangrass, and Switchgrass with temperatures ranging from 65 to 98 F. Data are fitted with curves using equation f=a*exp(-.5*((x-x0)/b)^2).

Table 8. Tetrazolium (TZ) test results including differences between reported seed tag germination rates and actual, laboratory viability results.

NWSG Specie	Reported Germination	Actual Viability	Difference
Orchardgrass	90%	33%	57%
Switchgrass	65%	10%	55%
Indiangrass	87%	62%	25%
Riveroats	87%	23%	64%
Big Bluestem	76%	55%	21%
Little Bluestem	95%	64%	31%

Image 2. Comparative photographs between the cool season standard (Orchardgrass) and Little Bluestem, a NWSG example (bottom right).



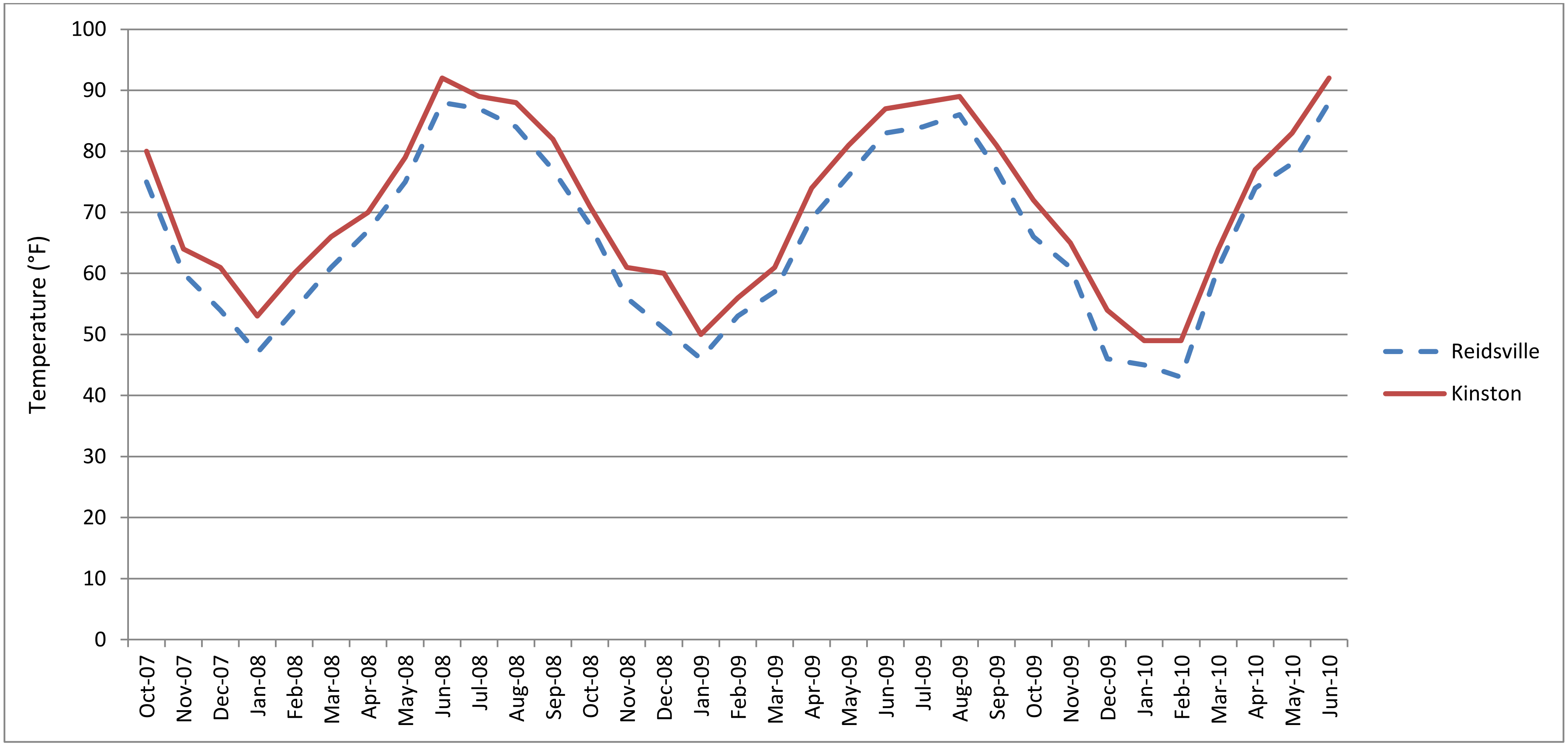


Figure 6. Maximum average monthly temperatures for the Upper Piedmont and Caswell Research Stations.

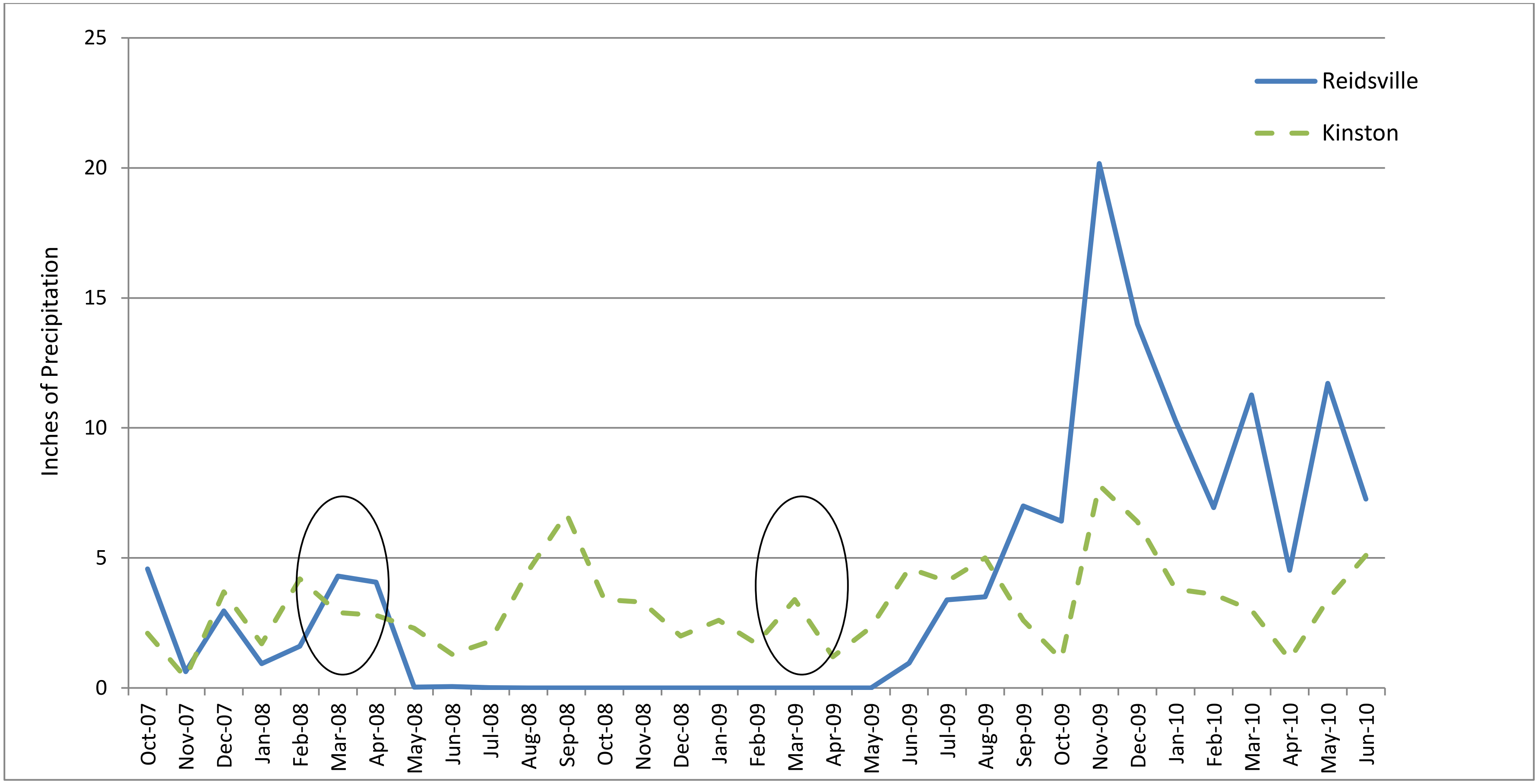


Figure 7. Averaged monthly rainfall for Upper Piedmont Research Station and Caswell Research Station from October 2007 to June 2010. 2008 and 2009 spring planting dates are illustrated within ovals express potential lack of success for first year plantings.

Table 9. Cost analysis between common herbicides utilized throughout life cycles of grass species.

Herbicides	Price	Rate	Price/Acre
Glyphosate (Round up Pro)	$19.95/gal	2-4 qt/Ac	$9.98-$19.95
Imazapic (Plateau)	$168.00/gal	6-12 oz/Ac	$7.87-$15.75
Quinclorac (Drive)	$60.35/lb	5.3 oz/Ac	$19.99
Aminopyralid (Milestone VM)	$344.00/gal	5-7 oz/Ac	$13.44-$18.81
2,4-D (Weedar 64)	$13.75/gal	0.5-4 pt/Ac	$0.86-$6.88

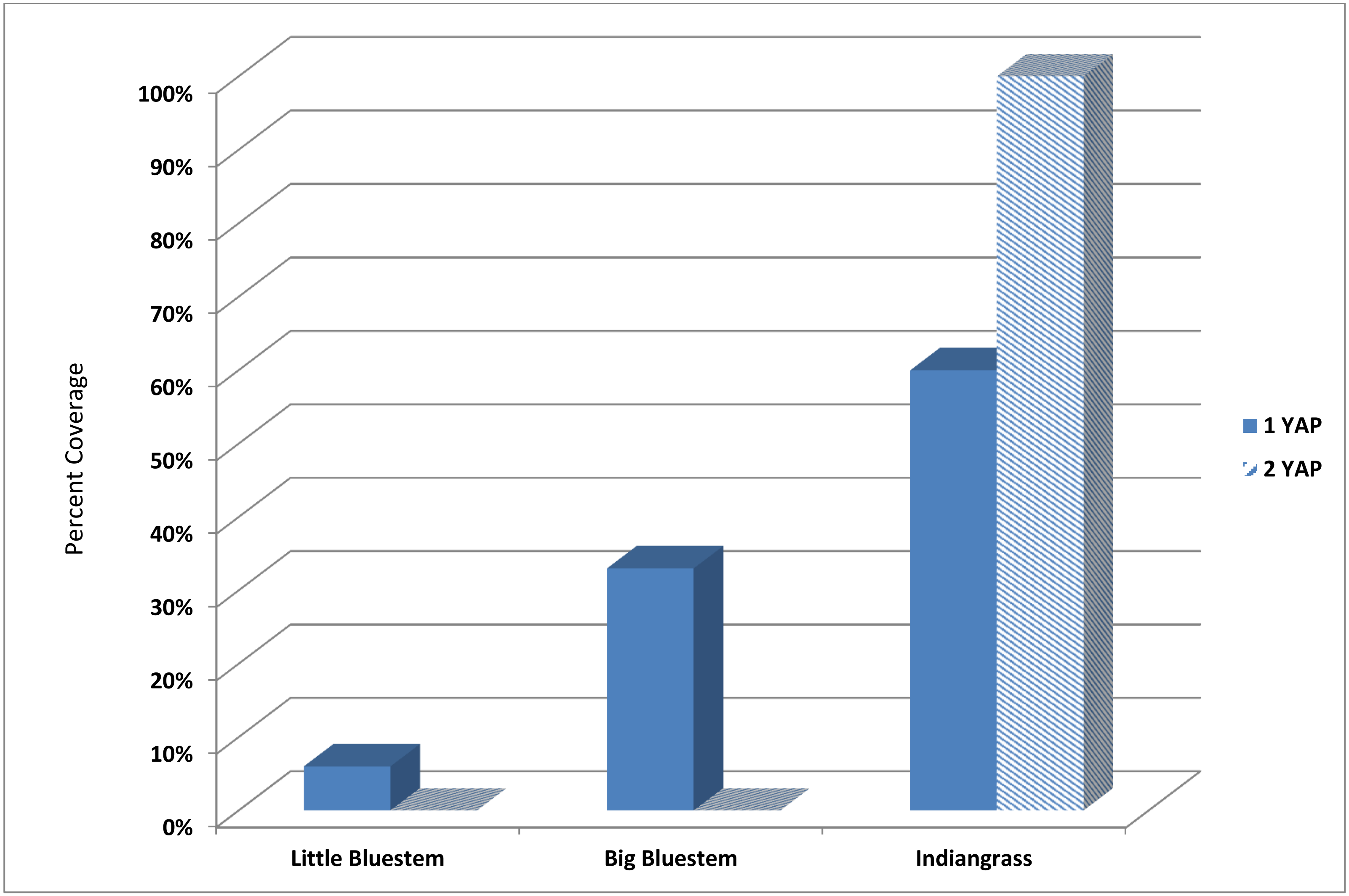


Figure 8. Population shift between two years of data from Goldsboro and Reidsville, NC. Quantitative data were taken using population counts of stem(s) per specie per meter square.

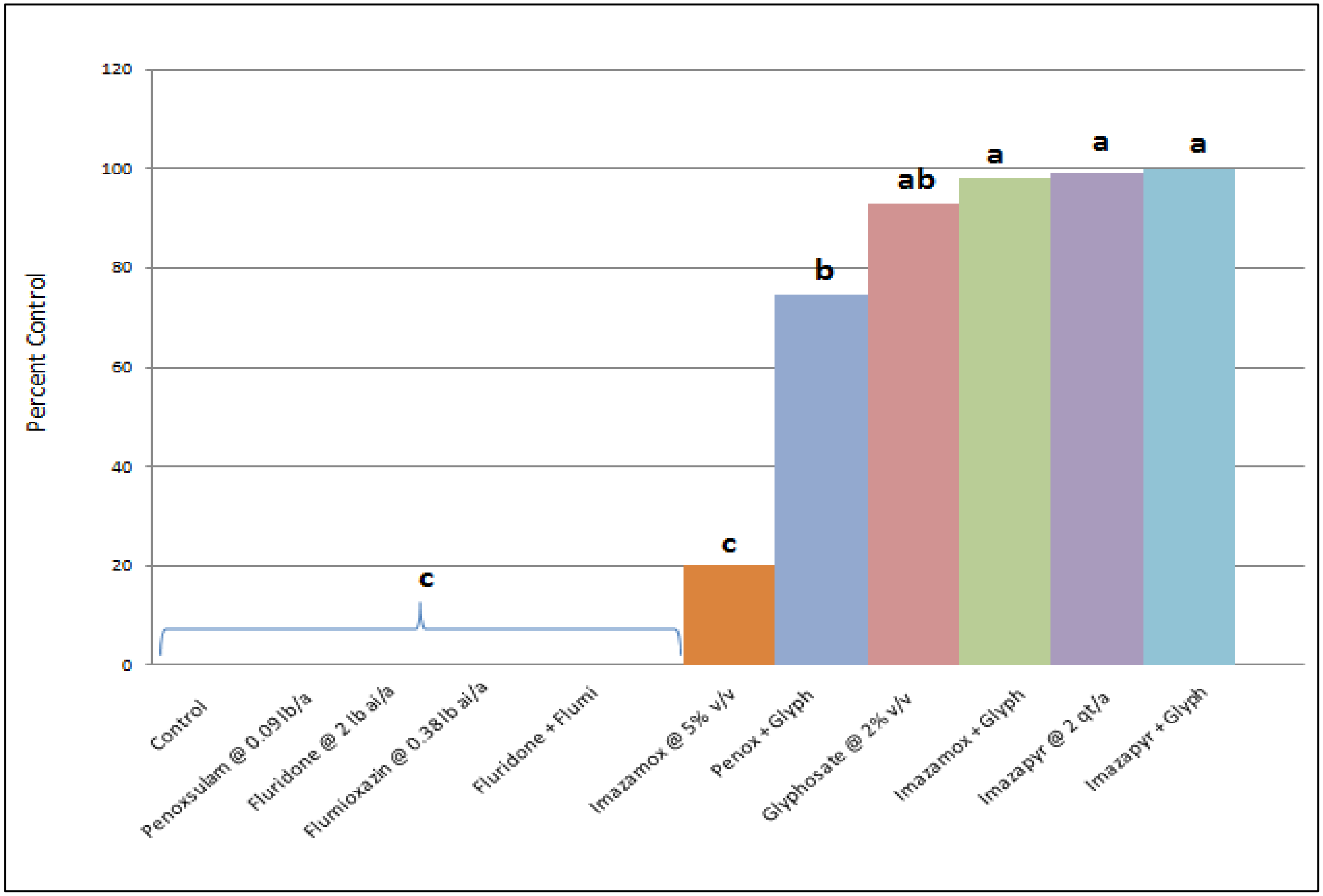


Figure 9. Japanese knotweed control 8 MAT. Significance determined using Fisher's Protected LSD (P≤0.05) and illustrated within columns; columns with like letter are not significantly different. Data was collected using visual ratings on a 0 (no control) to 100% (complete control) scale.

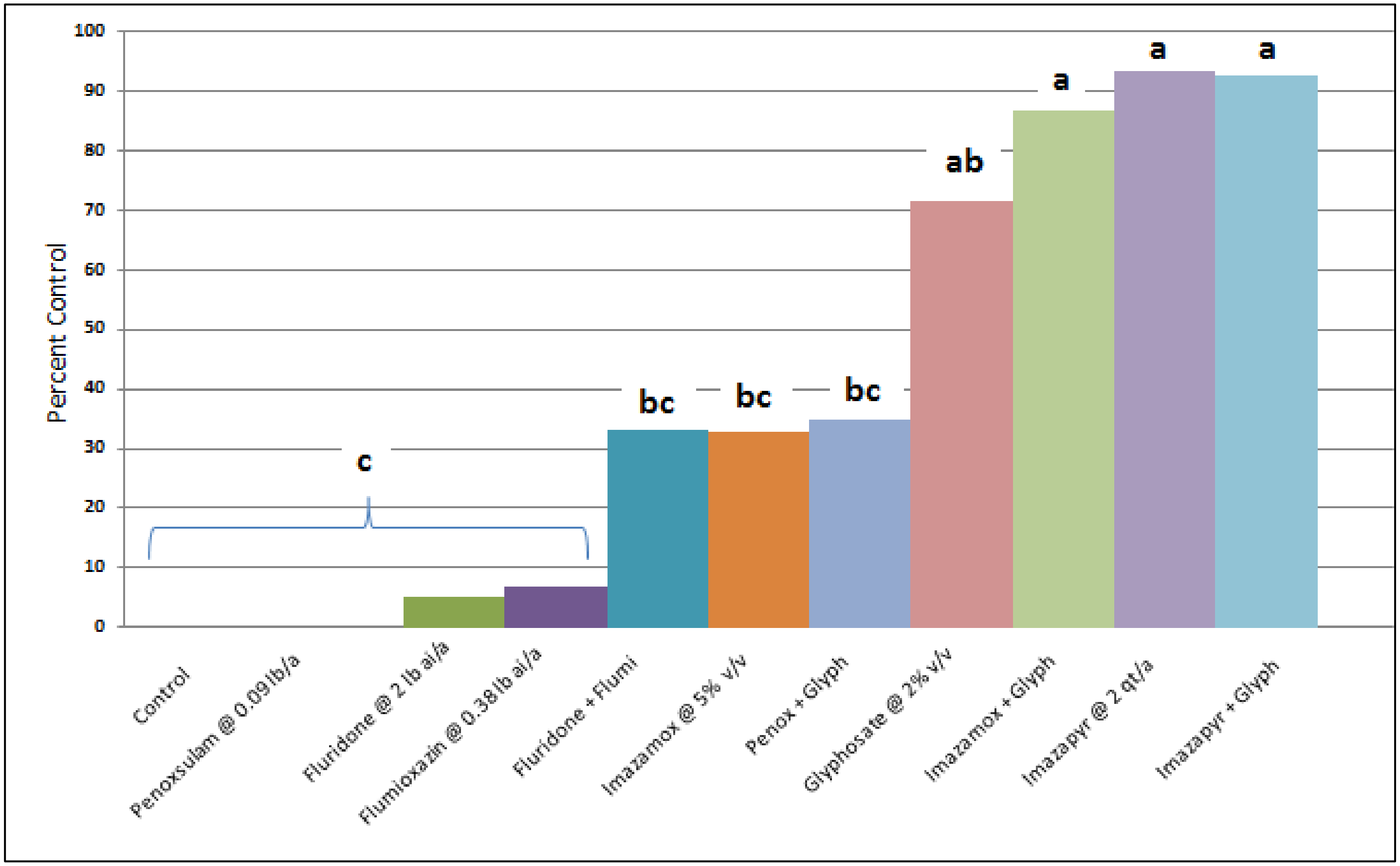


Figure 10. Japanese knotweed control 21 MAT. Significance determined using Fisher's Protected LSD (P≤0.05) and illustrated within columns; columns with like letter are not significantly different. Data was collected using visual ratings on a 0 (no control) to 100% (complete control) scale.

Table 10. Percent coverage and population counts per square meter for Japanese knotweed one year after treatment. Significance determined using Fisher's Protected LSD (P≤0.05) and illustrated within columns; columns with like letter are not significantly different. Data was collected using visual ratings on a 0 (no coverage) to 100% (complete coverage) scale.

	% Coverage	**# sprouts (m^2)**
Control	25.0 a	23.7 a
Penoxsulam @ 0.09 lb ai/a	6.3 b	8.3 bcd
Fluridone @ 2 lb ai/a	6.7 b	7.3 bcd
Flumioxazin @ .38 lb ai/a	10.0 b	17.3 ab
Fluridone + Flumi	25.0 a	25.3 a
Imazamox @ 1.5 lb ai/a	7.0 b	9.3 bcd
Penox + Glyph	9.7 b	14.3 abc
Glyphosate @ 1.8 ai/a	1.3 b	3.3 cd
Imazamox + Glyph	0.3 b	0.3 d
Imazapyr @ 1 lb ai/a	0.7 b	3.7 cd
Imazapyr + Glyph	0.3 b	0.7 d

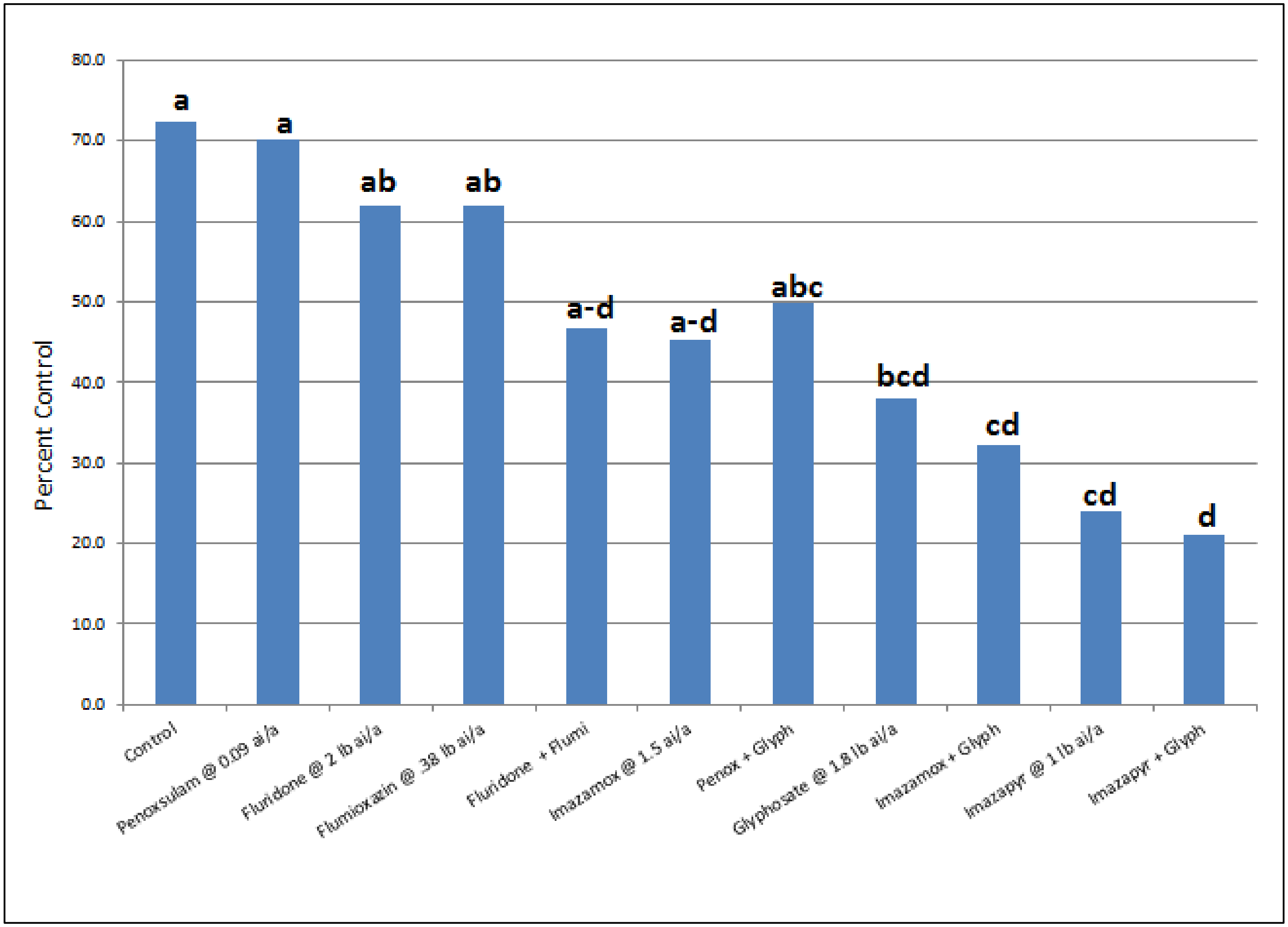


Figure 11. Japanese knotweed height (inches) 21 months after treatment. Significance determined using Fisher's Protected LSD (P≤0.05) and illustrated within columns; columns with like letter are not significantly different. Data was collected using visual ratings on a 0 (no control) to 100% (complete control) scale.

Image 4. Visual Japanese knotweed control with Habitat/imazapyr (left) and Touchdown Pro/glyphosate (right).

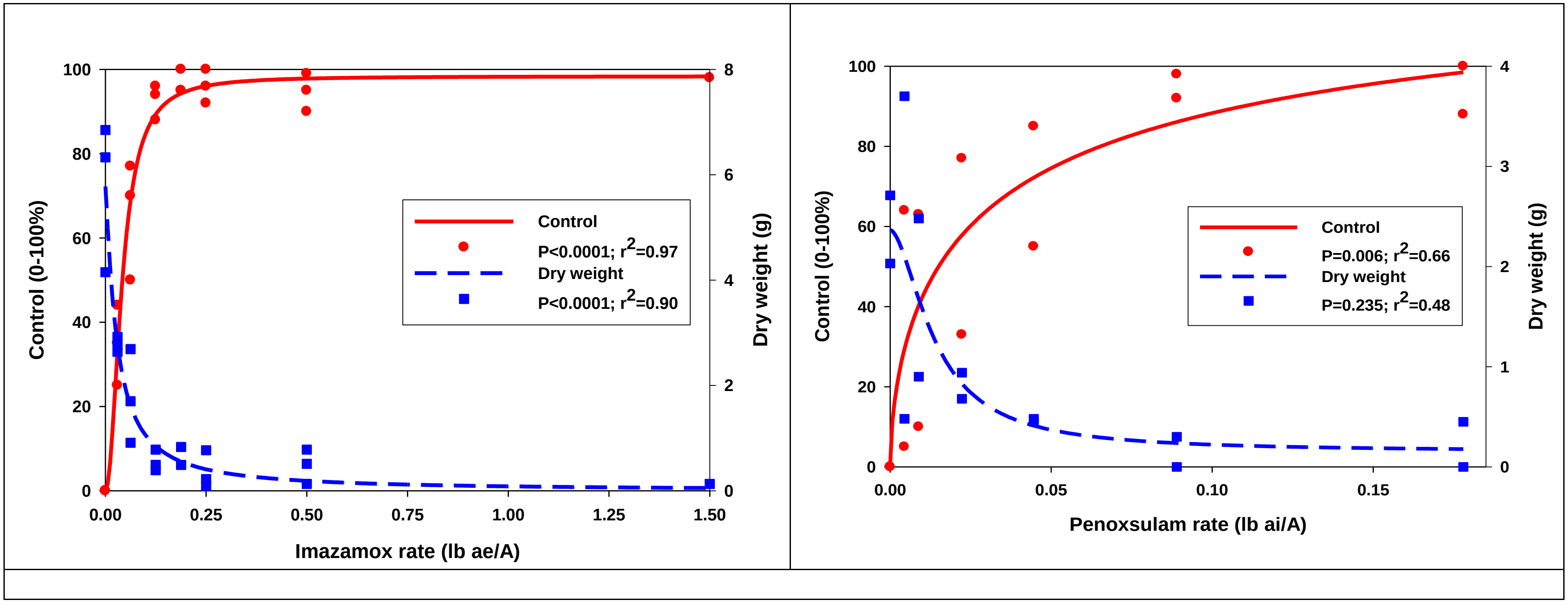


Figure 12. Alligatorweed control in the greenhouse with Clearcast/imazamox (left) and Galleon/penoxsulam (right) in pounds acid equivalent (ae) or active ingredient (ai) per acre (A).

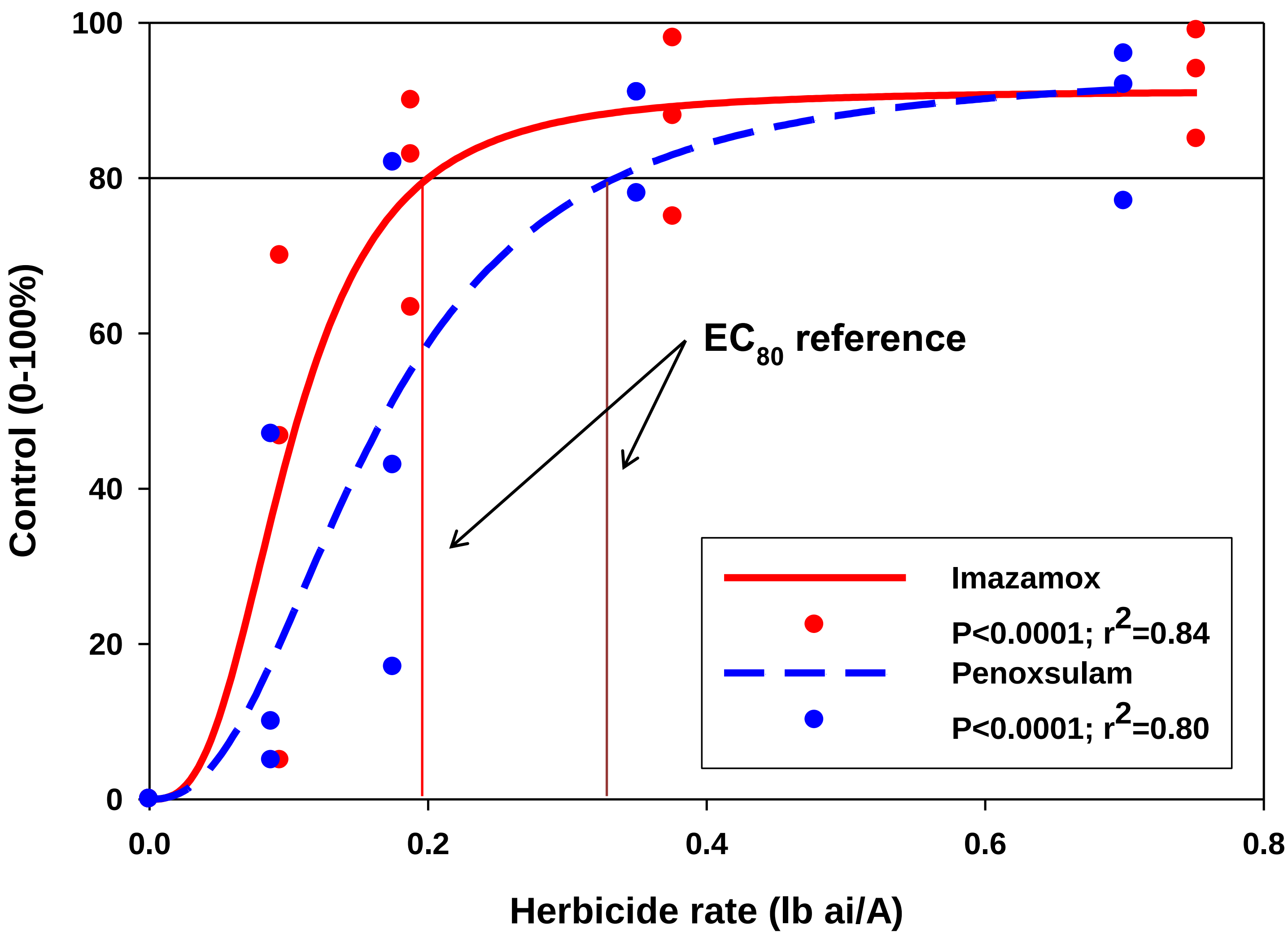


Figure 13. Alligatorweed control in the field at one month after treatment with Clearcast (imazamox) and Galleon (penoxsulam); herbicide rate given in pounds active ingredient per acre.

Table 11. Efficacy of selected herbicides on alligatorweed in the field.

Herbicide		2007		2008A	2008B	
		1 MAT	2 MAT	1 MAT	1 MAT	2 MAT
	lb ae/A	Control %				
Glyphosate	2	93	43	64	18	5
Imazamox	0.18	80	91	71	57	12
Imazapyr	0.3	91	100	93	85	91
Penoxsulam	0.35	86	47	80	50	0
Triclopyr	4	92	93	98	72	3
LSD		8	20	19	9	5