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# Zoysiagrass Expansion on Roadsides



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**RESEARCH &  
DEVELOPMENT**

# Zoysiagrass Expansion on Roadsides

## FINAL REPORT

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Research and Development Unit  
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16. Abstract In the past, NCDOT has used a combination of chemical and mechanical weed control management strategies that have proven time consuming and expensive to implement under and around over 1000 miles of median rail. NCDOT spent about \$40 million in fiscal year 2019 for mowing. Zoysiagrasses are known to be a thick sod-producing turfgrass that once adequately established have minimal weed invasion. Zoysiagrass germplasm that can be established quicker using sprigging methods may provide a solution to their establishment and long-term maintenance. Thus, this project was conducted to evaluate different varieties, methods, timings, and companion crops for establishment of zoysiagrass on roadside plots. These evaluations were done on roadsides in different climatic regions of NC. For one objective, cultivars were seeded or sprigged at different timings (fall or spring) in "operational-sized" roadside plots. Zoysiagrass planted via seed was faster to establish with greater coverage for both locations and all timings compared to sprigging. Although establishment and coverage from sprigging material was not rapid, it should be noted that 'El Toro' zoysiagrass sprigs were able to show comparatively similar coverage to seed plantings after about 22 months. A secondary objective was to evaluate fall and spring zoysiagrass establishment using seed or sprigs with companion crops (wildlife mix, brown top millet, or none). Companion crops showed promise in sandy soils but did not increase zoysiagrass establishment in loamy soils. Overall, results from this research suggest that zoysiagrass seed or sprigs can be established on NC roadsides with minimal inputs but additional research is needed to refine the methods to increase establishment success and zoysiagrass cover. Additionally, progress was made in developing new vegetative zoysiagrasses with improved expansion rates and persistence under no inputs as wells as new populations with increased seed yields. Further evaluation of these newly bred materials is needed to confirm their suitability for roadside applications.			
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## SUMMARY

This research project was conducted to assess different methods and timings for establishment of zoysiagrass on roadsides, and to evaluate establishment speed when planted with companion crops. Results from this field research provide evidence that both seed and sprigs can be used to establish zoysiagrass (*Z. japonica*) with minimal inputs across varying environmental conditions present on NC roadsides. Using commercial cultivars, greater zoysiagrass coverage, overall, was observed in Yadkin County compared to Harnett County, NC, likely because of major soil differences between locations. Broadcasting and slit seeding zoysiagrass seed showed accelerated establishment and greater percent coverage for all monthly plantings throughout data collection, with just a few exceptions. In Yadkin County, sprigging 'El Toro' zoysiagrass planted in the spring had similar (>80%) cover as seeded entries by 22 months after planting.

These results agree with previous NC-DOT research (Milla-Lewis & Miller, 2020) in which zoysiagrass seeding or sprigging was recommended in low-input situations on NC roadsides. However, seeding and sprigging are not without the potential for failure. In this research, results ranged from <3% to > 80% coverage in 22 months from the same plant timings. Fall planting typically assured greater success in this study. Although establishment and coverage from sprigging material were not rapid, 'El Toro' zoysiagrass sprigs were able to show comparatively similar coverage (51%) to seed plantings after about 22 months in Harnett County. Under roadside conditions, zoysiagrass was generally able to outcompete weeds and produce a near weed-free stand in  $\leq 2$  years. Implications from the presented research indicate that broadcast or drill seeding may be the most effective route for zoysiagrass establishment on NC roadsides.

Additionally, zoysiagrass establishment on roadsides and under guardrails has potential value that may not be quantified, such as continuous spread and growth and minimal maintenance during dormancy, as well as increased safety due to lower maintenance. Sprigging and seeding warm-season turfgrasses can be a great cost-saving compared to sodding. Different companion crops were tested to reduce zoysiagrass and soil erosion and increase zoysiagrass establishment. Zoysiagrass (seed or sprigs) and companion crops applied in Yadkin County, NC achieved greater zoysiagrass coverage compared to Moore County, NC. Increased establishment in Yadkin County was most likely due to the loamy soil and increased water holding capacity. The wildlife mix companion crop increased zoysiagrass cover in Moore County when planted in the spring. However, zoysiagrass planted with and without companion crops in Yadkin County had similar ground cover at 22 months after planting.

The presented research suggested that the single greatest impact to zoysiagrass establishment may be limited water availability during the early phase of establishment. While there were differences in performance between locations and among years, in general seeded 'Compadre' performed the best among commercial cultivars and was the most stable across locations and years. For the breeding component of this project, promising vegetative and seeded zoysiagrasses were developed. Evaluation of over 350 new vegetative hybrids identified 12 lines with aggressive lateral spread and superior ability to persist under minimal inputs. These materials were advanced to replicated nurseries in eastern (Goldsboro) and western (Yadkinville) North Carolina to evaluate their suitability for roadside applications. For seeded zoysiagrasses, lines with high inflorescence abundance were crossed in different combinations to evaluate their potential as parents in development of cultivars with improved yields. Large differences were observed among lines for their ability to perform as male versus female parents: Some were prolific both, some as only one, and some as neither. Furthermore, germination tests found that high variability exists for seedling recovery, but a few lines with high recovery rates were identified. A replicated trial was established at the Lake Wheeler Turfgrass Field Lab (Raleigh) to evaluate yield as well as to generate enough seed for future evaluations in replicated roadside trials.

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## **INTRODUCTION**

The Roadside Environmental Unit of NCDOT has found zoysiagrasses to be a promising roadside vegetation, especially in harder-to-manage areas around median rails. The use of zoysiagrass may provide reduced long-term maintenance and improve associated aesthetics. Reduced maintenance can translate into decreased worker presence and increased safety.

Zoysiagrasses possess considerable variation in many traits. These include winter-hardiness and high temperature tolerance (Beard, 1973), evapotranspiration rates (Green et al., 1991), response to drought (White et al., 1993), rooting (Marcum et al., 1995), salinity response (Quian et al., 1998), and insect responses (Reinert and Engelke, 1992). Because of favorable responses to many of these traits, several zoysiagrass cultivars are commercially available throughout the transition zone and the southeast US. The adoption of a zoysiagrass cultivar in the transition zone is usually determined by its ability to persist through the coldest of winters. From an aesthetic perspective, a finer leaf blade and darker green genetic color are more desirable for landscape uses of turfgrasses, including zoysiagrasses. Common complaints with some of the older zoysiagrass cultivars were their slow establishment and recuperative rates.

Research to evaluate zoysiagrass sod establishment along guardrails was previously conducted for NCDOT by Gannon et al. (2017) with findings in (Report No. FHWANC/2013-17). In their study, the commercial cultivars 'El Toro', 'Meyer', and 'Zeon' were evaluated from sod strips. They found El Toro to be good at spreading from the vegetation piece in year 2 of their study; whereas Meyer and Zeon spread at least 11% lower. There was significant variability in establishment between the two years evaluated in this study. Their results suggested that watering and other management inputs, in addition to environmental conditions and germplasm, could substantially influence success in establishing zoysiagrass. Specifically, Gannon et al. (2016) attributed the primary failure of zoysiagrass seeding failure on roadsides to inadequate water.

Contrary to the results of Gannon et al. (2017), Milla-Lewis and Miller (2020) reported that both seed and sprigs can be used to establish zoysiagrass with minimal inputs across varying environmental conditions present on North Carolina roadsides. Using commercial cultivars, greater zoysiagrass coverage, overall, was observed in heavier, loamy soils compared to sandy soils. Broadcasting seeding zoysiagrass showed accelerated establishment and greater percent coverage for all monthly plantings throughout data collection, with just a few exceptions. However, seeding still had the potential for failure. Results ranged from seed failure to > 70% coverage in less than 5 months for the same plant timings. Planting in late spring typically assured greater success (Milla-Lewis & Miller, 2020). Although establishment and coverage from sprigging material were not rapid, sprigs were able to show comparatively similar coverage to seed plantings after 12-15 months from planting. Zoysiagrass, even under roadside conditions, was generally able to outcompete weeds and produced a near weed-free sward of zoysiagrass in  $\leq 2$  years (Milla-Lewis & Miller, 2020).

As previously mentioned, finer leaf texture and darker green genetic color are aesthetically more desirable for landscape uses of turfgrasses, including zoysiagrasses. However, turfgrasses must combine a rapid establishment rate with strong aesthetics to be accepted for landscape uses. A number of zoysiagrass cultivars have been released on the market possessing finer leaf texture and darker green color, including 'Lobo', a recent release from the NC State University's Turfgrass Breeding and Genetics Program. In addition, research has addressed the freezing tolerance of zoysiagrass. The cultivar Lobo and

germplasm XZ14070 exhibited rapid establishment (spread) rate on NC roadsides in Lenoir and Yadkin Counties (Milla-Lewis & Miller, 2020).

To date, little research has been performed using “operational-sized” roadside plots regarding use and management of zoysiagrass in North Carolina. Turfgrass quality considerations are very different for roadsides compared to landscape uses. For roadside use, the most desirable traits are rapid establishment and recuperative rates, dense canopy to suppress weed encroachment, tolerance to a variety of soils and nutritive situations, and freezing tolerance. A wide leaf blade (relative to other turf-type grasses) is acceptable, and a longer internode length and larger node diameter may offer advantages for roadside use compared to landscape uses. The primary goal of this project is to find the best zoysiagrass cover for NC roadsides. To accomplish this goal, it must first be determined if sprigging new zoysiagrass germplasm will allow quicker establishment compared to currently available varieties. A preliminary objective for this project is to determine which zoysiagrass varieties are currently commercially available for consideration in this research. To accomplish this goal, the following tasks will be pursued:

1. Evaluate potential establishment methods of large, operational-sized plots that require lower inputs than sodding during periods of low-water requirements compared to more traditional establishment periods.
2. Evaluate the use of companion crops to optimize zoysiagrass establishment and minimize soil and zoysiagrass seed erosion.

Additionally, the turfgrass breeding and genetics program at NC State works on the development and selection of zoysiagrass breeding lines for improved speed of establishment, drought tolerance, persistence, and cold hardiness, among other traits. New hybrids need to be tested for their aggressiveness and persistence under minimal inputs to determine if they have promise for roadside use. Furthermore, evaluation of new zoysiagrass breeding lines for their seed yield potential and ability to establish from seed would be desirable. Therefore, additional tasks for this project were the development and selection of new vegetative and seeded zoysiagrass materials.

Outcomes from this project will provide NCDOT with guidelines for the establishment of zoysiagrass on roadsides. Additionally, this research might identify zoysiagrass cultivars and germplasm that are better adapted to roadside conditions. Results generated from this research will provide NCDOT with the information needed to select the best planting practices as well as the best germplasm for reduced long-term maintenance and improved associated aesthetics. Reduced maintenance can translate into decreased worker presence and increased safety.

**CHAPTER 1: ESTABLISHMENT TIMINGS OF OPERATIONAL-SIZED ROADSIDE PLOTS**

Materials and Methods

Field research was initiated in the summer (July) and fall 2021 and spring 2022 (April; year 1) and repeated the following fall 2022 (October) and spring 2023 (May; year 2) on North Carolina (NC) roadsides. Two locations were selected to represent the varying climatic and edaphic conditions present in NC. Harnett County [at a border with Lee County] (35°26'20"N 79°02'18"W) in the coastal plain region (USDA zone 8a) was chosen as an eastern site. Yadkin County (36°07'52"N 80°50'29"W) in the piedmont region (USDA zone 7b) was chosen as a western site. Zoysiagrass (*Z. japonica*) planting material evaluated at both locations included ‘El Toro,’ ‘Lobo,’ and XZ14070 sprigs and ‘Compadre’ seed (broadcast or drilled). Planting materials were arranged in a randomized complete block design with three replications, and plots measured 12.2 m × 3.1 m. In year 1 of the study, one summer, fall, and spring plantings were evaluated. In year 2 of the study, one fall (October) and one spring (April or May) were evaluated (Table 1). No irrigation was applied throughout this research.

Table 1. Zoysiagrass planting dates in Harnett and Yadkin County, NC, during all years.

Location (County)	-----Year 1-----		
	Summer	Fall	Spring
Harnett	23 July 2021	22 October 2021	22 April 2022
Yadkin	-	20 October 2021	29 April 2022
Location (County)	-----Year 2-----		
	Summer	Fall	Spring
Harnett	-	5 October 2022	5 May 2023
Yadkin	-	19 October 2022	4 May 2023

Zoysiagrass material for sprig treatments was selected for their favorable, aggressive lateral spread (Milla-Lewis & Miller, 2020; Patton et al., 2007). Compadre seed was chosen because it is commercially available and is less likely to desiccate compared to sprigs. Broadcast seeding Compadre established rapidly and performed well in previous research (Milla-Lewis & Miller, 2020). The drill seeding treatment was included because it required no tillage before planting, yet still provided good seed-to-soil contact. For the selection of planting seasons, spring and summer are typically suggested for planting warm-season turfgrass. Fall plantings were selected for their decreased air temperatures and lower water demands during establishment.

Four weeks before each planting, glyphosate (3.8 kg ai ha<sup>-1</sup>) was applied to control any existing vegetation. Zero to fourteen days before planting, planting areas were rototilled to a 15-cm depth to control pre-existing vegetation at each site. Plots to be drill-seeded were not rototilled before planting. Sprigs were made from sod removed from established plots at North Carolina State University’s Lake Wheeler Turf Field laboratory in Raleigh, NC. Approximately 24 to 48 hours before planting, sod measuring 15.2 m × 0.46 m × 0.02 m was harvested. During planting (Table 1), sod was continuously fed by hand into the sprigging unit (Strickland Bros. Enterprises Inc., Spring Hope, NC), was mechanically shredded, and was deposited on top of the soil surface of the planting areas. Pre-weighed amounts of seed were

broadcast by hand at 98 kg pure live seed (PLS) ha<sup>-1</sup>. Drilled seeding treatments were applied with a tractor-mounted drill seeder (Model: Tye 2007 Pasture Pleaser, Tye Co. Lockney, TX) at a rate of 49 kg ha<sup>-1</sup> (1 pound per 1000 sq ft). After planting, a rolling cultipacker (Model: KP-48-ATV, King Kutter Inc., Winfield, AL) was pulled across the entire planted area behind a utility vehicle (Model: X900, Kubota Tractor Corporation, Grapevine, TX) to ensure soil contact with seeds and sprigs. Experimental units received no supplemental irrigation beyond natural rainfall.

Beginning one month after July planting, sites were clipped with a rotary mower (Scag V-Ride II, Scag Power Equipment, Mayville, WI 30284) to a 7.62 cm (3 in) height of cut on a one to two month basis during the growing season. Fertilization was withheld throughout the study. Meteorological data was acquired from the closest weather station provided by the North Carolina State Climate Office Cardinal Data System (Cardinal, 2024). East weather data was gathered from Sanford-Lee County Airport (35°26'20"N 79°02'20"W), approximately 15 km from the Broadway plots. West data was collected from Wilkes County Airport (36°07'52"N 80°50'30"W), approximately 25 km from the Cycle site in Yadkin County. Beginning 1 September through 31 August, during both years, weekly averages of maximum and minimum air temperature (°C), and cumulative weekly precipitation (cm) were collected from both stations.

The percent zoysiagrass cover was visually estimated every one to three months during the year. Percent cover ratings were on a 0 to 100% scale with 0 = no zoysiagrass cover and 100 = full zoysiagrass coverage. The visual representation of 0% cover could still have weedy vegetation within the respective plot area. Shortly after each monthly planting, volunteer vegetation would occupy the majority of the plots throughout this study. Data collection began in September 2021 for year 1 and February 2023 for year 2. Data of all monthly plantings within each year were recorded during all rating events.

This study analyzed planting season and material across two locations in NC over two years. Harnett and Yadkin Counties were selectively chosen to represent general climatic and edaphic conditions present in the coastal plains and piedmont regions of NC, respectively. Therefore, location was a fixed effect. Cover data were subjected to analysis of variance using the PROC GLIMMIX procedure in the Statistical Analysis System software (version 9.4; SAS Inst. Inc., Cary, NC) to determine treatment effects and interactions. Significant year × location × season × treatment × rating date interaction occurred, therefore, data were sorted by year, location, and rating date and were presented separately. The identified significant main effects and interactions were sorted and analyzed accordingly using least significant difference with a probability level of 0.05. Progress in turfgrass establishment during multiple years, is presented as monthly means of percent zoysiagrass cover.

## Findings and Conclusions

### ***Environmental Influences***

Significant year and location interactions occurred throughout data collection, as research locations were selected to represent the varying climatic and edaphic conditions present across NC. With no supplemental irrigation being applied, differences in precipitation patterns and amounts between years were the most likely cause for yearly interactions. During year 1, Harnett County received a total of 57.9 .3 cm of rainfall (22 Oct 2021 to 21 Oct 2022) while 65.9 cm of rain fell during year 2 (5 October 2022 to 6 October 2023). During year 1 in Yadkin County, 93.4 cm of precipitation fell during year 1 (22 October 2021 to 21 October 2022) while a total of 100 cm of rain fell during year 2 (19 October 2022 to 20 October 2023). Like the majority of eastern NC soils, the Harnett County site is dominated by sand-sized particles (≥ 70%). Sand particles have a low specific surface area, low water-holding capacity, and contribute very little to plant nutrition. In contrast, the heavier soils at the Yadkin County site contained at least 15% clay-sized particles. Clay particles have a large specific surface area, adsorb a great deal of water, and are a

more effective buffer between rainfall events. Data suggest this led to greater overall zoysiagrass coverage in Yadkin compared to Harnett County during both years.

### ***Yadkin County (Cycle, NC)***

Analysis of variance determined significant season × treatment × planting material × rating date interaction on zoysiagrass cover in both years and both locations (Yadkin and Harnett County, NC). In Yadkin County, at 36 weeks after the fall planting (WAFP) (1 July 2022), the fall broadcast treatment had the greatest zoysiagrass cover (58%) of all treatments and 35 to 39% more zoysiagrass cover compared to the fall drill and spring broadcast seeding treatments (Table 2). The fall broadcast treatment had ≥51% greater zoysiagrass cover compared to all sprigged treatments. The fall drill seeding treatment had ≥12% greater zoysiagrass cover compared to fall and spring sprigged treatments. The fall broadcast treatment had the greatest zoysiagrass cover of any treatment from 36 through 97 WAFP. From 36 through 97 WAFP, the fall broadcast seed treatment had 11 to 49% greater zoysiagrass cover compared to the fall drill and spring broadcast seed treatments. Broadcast treatments were rototilled before planting, thus had decreased weed pressure and increased seed to soil contact compared to non-rototilled, drill seeding treatments. However, by 97 WAFP, fall- and spring-broadcast and drill-seeded treatments had similar ≥87% zoysiagrass cover. Although drill seeding was slower to establish, it had good zoysiagrass cover (85%) at 97 WAFP, required minimal seedbed preparation, and had decreased potential of soil erosion. These benefits may justify drilling compared to broadcast seeding in loamy soils.

Of the sprigged treatments, El Toro planted in the fall had the greatest zoysiagrass cover from 67 WAFP through 97 WAFP (Table 2). By 97 WAFP, El Toro had similar zoysiagrass cover as the spring broadcast, as well as fall and spring drilled treatments. Milla-Lewis and Miller (2020) observed acceptable (albeit slower) establishment with Crowne and Compadre sprigs compared to broadcast seeding Compadre or Zenith zoysiagrasses. Results of this current study demonstrate that sprigging and seeding of zoysiagrass is a viable establishment method for NC roadsides and agree with Milla-Lewis and Miller (2020). El Toro had superior establishment compared to Lobo and XZ 14070. This result was unexpected. In previous research, Lobo and XZ 14070 sprigs had a similar to superior establishment rate compared to commercially available varieties (Milla-Lewis & Miller, 2020). However, in their study, Lobo and XZ 14070 received irrigation immediately after planting (Milla-Lewis & Miller, 2020). The age of the turfgrass stand the sod was harvested may have affected sprigging success. El Toro was harvested from a mature (>10 year old) plot. Lobo and XZ 14070 stands were less mature (<3 years old) and may have had fewer viable rhizomes and stolons (sprigs).

In year two at Yadkin County, no differences in zoysiagrass cover were observed until 46 WAFP when the fall and spring broadcast treatments had the greatest zoysiagrass cover (28 to 47%). The spring sprigging and fall drill seeding treatments had 7 to 13% cover at 46 WAFP (Table 3). Increased zoysiagrass sprig establishment in the spring treatment was attributed to increased soil moisture at planting and timely rainfall after planting. The site received >12.7 cm of rainfall ≤ 7 days before planting and received 0.2 cm < 5 days after planting in the spring. The fall planting received 6.8 cm ≤ 7 days before planting, but the site received no rainfall < 5 days after planting, which likely resulted in increased sprig mortality. Although the spring sprig treatments had similar cover as fall sprigged treatments (0%). Differences between planting timings and treatments are anticipated to decrease after another full growing season.

### ***Harnett County (near Broadway, NC)***

At the Harnett County site in year 1, establishment in the summer planting was superior to fall and spring plantings. In the summer planting, the broadcast and drill seeding treatments consistently had the greatest zoysiagrass cover during all ratings. El Toro sprigs had similar zoysiagrass cover (≥ 95%) at 104 weeks after the summer planting. Lobo and XZ 14070 had 27 to 60% less zoysiagrass cover, respectively,

compared to seeded and El Toro treatments at the final rating date. However, Lobo and XZ 14070 had 35 and 68% cover, respectively, at the final rating date, and the cover of both treatments is anticipated to increase in future ratings.

At the Harnett County site in year 1, no differences in zoysiagrass cover were observed until 67 WAFP. However, all treatments had  $\leq 1\%$  zoysiagrass cover (Table 4). Broadcast seeding and sprigged El Toro in the fall had similar cover ( $\sim 50\%$ ) by 97 WAFP. Milla-Lewis and Miller (2020) observed slow but continuous seed and sprig establishment in the NC coastal plain when planted in the spring or fall.

In the first year, establishment in the spring planting was poor, and all treatments had  $< 2\%$  zoysiagrass cover by 97 WAFP (Table 4). The lack of timely rainfall and high crabgrass pressure resulted in increased sprig mortality and poor seed establishment.

In year two at the Harnett County site, no differences were observed between treatments and the timing of planting. All treatments had  $\leq 6\%$  zoysiagrass cover by 46 WAFP (Table 5). Milla-Lewis and Miller (2020) observed slower zoysiagrass establishment in the NC Coastal plain compared to the NC Piedmont because of previously mentioned differences in soil texture and precipitation.

Table 2. Cover estimates of zoysiagrass planting material planted in fall and spring months in Yadkin County (Cycle), NC during year 1 (2021-22). Cell color represents a similar statistical group within the column, usually the top performers.

Season/Planting Material	Planting Season	5/1/2022	7/1/2022	10/1/2022	2/1/2023	5/1/2023	9/1/2023
----- % cover -----							
Fall							
XZ 14070		0.4 a	0.2 d	1.3 d	1.3 d	0.7 d	7.3 cd
El Toro		1.3 a	6.7 cd	18.3 cd	26.7 c	18.3 cd	80.0 b
Lobo		0.2 a	0.5 d	3.0 d	3.3 d	0.8 d	21.7 c
Compadre Broadcast		7.7 a	58.3 a	75.7 a	78.3 a	80.7 a	98.3 a
Compadre Drilled		5.7 a	19.3 bc	30.0 bc	40.0 bc	35.0 bc	87.3 ab
Spring							
XZ 14070		0.0 a	1.0 d	0.7 d	1.7 d	1.0 d	2.3 d
El Toro		0.7 a	1.2 d	3.3 d	6.3 d	5.0 d	25.0 c
Lobo		0.1 a	0.0 d	0.7 d	0.5 d	0.0 d	2.3 d
Compadre Broadcast		0.7 a	23.3 b	43.3 b	55.0 b	53.3 b	87.3 ab
Compadre Drilled		0.3 a	6.3 cd	13.3 cd	26.7 c	18.3 cd	85.0 ab

Table 3. Cover estimates of zoysiagrass planting material planted in fall and spring months in Yadkin County (Cycle), NC, during year 2 (2022-23). Cell color represents a similar statistical group within the column, usually the top performers.

Planting Material	Planting Season	----- % cover -----		
		2/1/2023	5/1/2023	9/1/2023
	Fall			
XZ 14070		0.0 a	0.0a	0.0c
El Toro		0.0 a	0.0a	0.0c
Lobo		0.0 a	0.0a	0.0c
Compadre Broadcast		0.0 a	0.0a	46.7 a
Compadre Drilled		0.0 a	0.0a	12.0bc
	Spring			
XZ 14070		-	-	12.7bc
El Toro		-	-	11.7bc
Lobo		-	-	7.3bc
Compadre Broadcast		-	-	28.3 ab
Compadre Drilled		-	-	1.0c

Table 4. Cover estimates of zoysiagrass planting material planted in fall and spring in Harnett County (Broadway), NC, during year 1 (2021-22). Cell color represents a similar statistical group within the column, usually the top performers.

Planting Material	Planting Season	5/1/2022	7/1/2022	10/1/2022	2/1/2023	5/1/2023	9/1/2023
----- % cover -----							
	Fall						
XZ 14070		0.2 ab	0.1 b	0.0 a	0.0 c	0.0b	3.0b
El Toro		0.5 a	0.3 b	0.3 a	0.8 a	2.0ab	50.7a
Lobo		0.5 a	0.2 a	0.1 a	0.0 c	0.3a	2.4b
Compadre Broadcast		0.3 ab	1.5 a	0.3 a	0.7 ab	2.7a	53.3a
Compadre Drilled		0.2 ab	0.9 ab	0.3 a	0.3 bc	1.8ab	29.0ab
	Spring						
XZ 14070		0.0 b	0.0 b	0.0 a	0.0 c	0.0b	1.2b
El Toro		0.0 b	0.0 b	0.0 a	0.0 c	0.0b	1.6b
Lobo		0.0 b	0.0 b	0.0 a	0.0 c	0.0b	0.0b
Compadre Broadcast		0.0 b	0.0 b	0.0 a	0.0 c	0.0b	0.5b
Compadre Drilled		0.0 b	0.0 b	0.0 a	0.0 c	0.0b	1.6b



Table 6. Cover estimates of zoysiagrass planting material planted in fall and spring in Harnett County (near Broadway), NC during year 2 (2022-23).

Planting Material	Planting Season	% cover		
		2/1/2023	5/1/2023	9/1/2023
	Fall			
XZ 14070		0.0 a	0.0a	0.6 b
El Toro		0.0 a	0.0a	3.5 ab
Lobo		0.0 a	0.0a	0.3 b
Compadre Broadcast		0.0 a	0.0a	4.2 ab
Compadre Drilled		0.0 a	0.0a	6.1 a
	Spring			
XZ 14070		-	-	0.5 b
El Toro		-	-	1.5 ab
Lobo		-	-	0.2 b
Compadre Broadcast		-	-	0.4 b
Compadre Drilled		-	-	0.2 b



Fall 2021 broadcast seeding Compadre zoysiagrass in Yadkin County on 15 February 2023 – 16 months (64 weeks) after planting.



Poor zoysiagrass establishment of Spring 2022 planting in Moore County on September 28, 2023.



Excellent weed (crabgrass) suppression of zoysiagrass established in Summer 2021 planting in Moore County on September 28, 2023.

## CHAPTER 2: COMPANION CROPS' IMPACT ON ZOYSIAGRASS ESTABLISHMENT

### Materials and Methods

Field research was initiated in July 2021, October 2021, and May 2022 (year 1) and repeated the following October 2022 and May 2023 (year 2), in Moore and Yadkin Counties, NC. Moore County (35°11'14"N 79°40'36"W) in the coastal plain region (USDA zone 8a) was chosen as an eastern site. The Cycle location (35°41'45.5" N, 80°37'43.3" W) in Yadkin County, NC (USDA zone 7b) was chosen as a western site. A month prior to study initiation, North Carolina State University (NCSU) Sandhills Research Station staff applied glyphosate (3.8 L a.i. ha<sup>-1</sup>) as a burn down, followed by tillage to 15 cm to control pre-existing vegetation at each site. For all plantings, Compadre zoysiagrass (*Z. japonica*) seed or El Toro sprigs were planted. Sprigs were harvested from an area measuring 1.5 m × 0.61 m × 0.02 m from the Lake Wheeler Turfgrass Field Lab in Raleigh, NC, approximately 24 hours before use as the sprig source. For planting, sprigs were broadcast by hand at a rate of 13 m<sup>3</sup> ha<sup>-1</sup> and were pressed into the soil with vertical coulter blades. For the seeded treatment, pre-weighed amounts of Compadre seed were broadcast by hand at 98 kg pure live seed (PLS) ha<sup>-1</sup>. Companion crops Fall Wildlife Mix at 98, 147 kg ha<sup>-1</sup> (Southern States Cooperative, Richmond, VA) or brown top millet at 29.5 kg ha<sup>-1</sup> (Southern Seeds Inc., Middlesex, NC) were broadcast by hand. After broadcasting sprig and seed treatments, a rolling cultipacker (Model: KP-48-ATV, King Kutter Inc., Winfield, AL) was pulled across the entire planted area behind a utility vehicle (Model: X900, Kubota Tractor Corporation, Grapevine, TX) to ensure soil contact with seeds and sprigs. Experimental units received no supplemental irrigation beyond natural rainfall. Beginning one month after July plantings, sites were clipped with a rotary mower (Models: VII, Scag Power Equipment, Mayville, WI and 30284, The Toro Company, Bloomington, MN) to 7.62 cm (3 in) height of cut on a one to two month basis during the growing season. Fertilization was withheld throughout the study. Meteorological data for each study location was acquired from the closest weather station provided by the North Carolina State Climate Office Cardinal Data System (Cardinal, 2024).



Fall 2021 planting of companion crop (wildlife mix and brown top millet) treatments in Yadkin County taken six months after planting (29 April 2022).

Zoysiagrass seed, sprigs, and companion crops were applied in the fall (20-22 October 2021 and 5-19 October 2022) and spring (22-29 April 2022 and 4-5 May 2023) of each year. The Moore County site

received an additional summer (July) planting in 2021 that was not repeated in year two. This study was arranged in a randomized complete block design with whole plots measuring 6.1 m × 1.5 m with three replications. The study area received no supplemental irrigation beyond natural rainfall.



Fall 2021 companion crop planting in Yadkin county on 15 February 2023 – 16 months (64 weeks) after planting.

Beginning one month after spring plantings, sites were clipped with a rotary mower (Models: 74201 and 30284, The Toro Company, Bloomington, MN) at a 6.35-7.62-cm height of cut on a monthly basis during the growing season. Both sites received no fertilization or irrigation throughout the study.

Percent zoysiagrass cover was visually estimated on a 0 (no cover) to 100% (complete zoysiagrass cover) scale 41, 49, 80 and 97 weeks after fall plantings (WAFP) in year 1. Year 2 estimates were recorded 41 and 46 WAFP. Meteorological data for each study location was acquired from the closest weather station provided by the North Carolina State Climate Office Cardinal Data System (Cardinal, 2024). East weather data was gathered from Sandhills Research Station (35°11'14"N 79°40'35"W), where trials were being conducted. West data was collected from Wilkes Co Airport (36°13'26"N 81°06'07"W) 25 km from the Cycle plots. This study analyzed planting material, cover crops, and season of planting in a combined analysis of location and year, with multiple zoysiagrass cover rating dates analyzed. Zoysiagrass cover data were subjected to analysis of variance using the PROC GLIMMIX procedure in the Statistical Analysis System software (version 9.4; SAS Inst. Inc., Cary, NC) to determine treatment effects and interactions. Significant year ( $P = 0.0082$ ) and location × treatment × season × rating date ( $P \leq 0.0001$ ) interaction occurred; therefore, data were sorted by year, location, rating date and were presented by season of planting × treatment. The identified significant main effects and interactions were sorted and analyzed accordingly using mean separation at a probability level of 0.05. Progress in turfgrass establishment for both years is presented as means of percent zoysiagrass cover every one to three months.

### Findings and Conclusions

#### ***Environmental Influence***

Significant interactions with year and other factors occurred due to varying climatic conditions between years. The most likely cause of interactions with year was due to inconsistent precipitation between years as no supplemental irrigation was applied beyond natural rainfall.

### ***Yadkin County (Cycle, NC) Plantings***

Analysis of variance determined significant location × treatment × planting season interactions on zoysiagrass cover estimates evaluated 36, 49, 67, 80, and 97 weeks after fall planting (WAFP) in year 1; therefore, zoysiagrass cover was presented by treatment and season at each location. Yadkin County treatments planted in the fall of each year had minimal time to establish prior to winter dormancy; however, they generally benefited from early spring weather and had greater zoysiagrass coverage compared to spring treatments. All fall-seeded treatments had similar cover from 49 WAFP. Sprigged fall and spring treatments had <12% cover at 97 WAFP, and 65 to 70% less cover compared to all seeded treatments by 97 WAFP (Table 6). Companion crops in fall plantings did not increase zoysiagrass cover compared to the no companion crop treatment. The wildlife mix and brown top millet companion crops in spring treatments increased zoysiagrass cover compared to the no companion crop treatment at 36, 49, 67, and 80 WAFP. However, by 97 WAFP the spring no-companion crop treatment had similar zoysiagrass cover as all companion crop treatments. All fall seeded, and spring high wildlife mix, brown top millet, and no companion crop treatments had similar ≥81% zoysiagrass cover by 97 WAFP. Although Milla-Lewis and Miller (2020) observed increased zoysiagrass cover when sprigging and seeding zoysiagrass in the spring compared to the fall, differences between planting seasons were minimal in this study. Similar cover of fall and spring, no companion crop treatments at 97 WAFP suggest that zoysiagrass may be seeded in the fall or spring if rapid establishment (<2 years) is not necessary.

In year 2 at 46 WAFP, the no companion crop fall and spring treatments had 52 and 23% zoysiagrass cover, respectively (Table 7). The no companion crop treatments had similar to superior zoysiagrass cover as all other treatments, including all companion crop treatments. Therefore, companion crops did not increase zoysiagrass cover during the second year of plantings.

Table 6. Zoysiagrass cover estimates of companion crop treatments planted in fall and spring in Yadkin County, NC, during year 1. Cell color represents a similar statistical group within the column, usually the top performers.

Planting Material	Planting Season	6/1/2022	7/1/2022	10/1/2022	2/1/2023	5/1/2023	9/1/2023
----- % cover -----							
Fall							
Drill	--	35.0 bcde	71.7 ab	73.3 abc	84.3 ab	99.0 a	
High Wild	--	61.7 ab	92.3 a	91.7 ab	93.0 a	100.0 a	
Low Wild	--	73.3 a	95.0 a	95.3 a	90.3 a	100.0 a	
Millet	--	56.7 abc	91.7 a	86.3 ab	95.3 a	100.0 a	
Seed	--	50.0 abcd	61.7 ab	83.0 ab	78.0 abc	99.0 a	
Sprig	--	0.2 e	0.7 c	0.3 e	1.7 d	5.0 c	
Spring							
Drill	1.5 a	26.7 bcde	38.3 bc	31.7 de	45.0 bcd	90.0 ab	
High Wild	0.8 a	35.7 bcde	59.7 ab	63.3 abcd	69.3 abc	96.7 ab	
Low Wild	1.0 a	36.0 abcde	45.0 bc	56.7 abcd	55.0 abc	76.7 b	
Millet	0.8 a	15.3 de	50.7 ab	51.7 bcd	63.3 abc	91.7 ab	
Seed	1.3 a	20.0 cde	31.8 bc	38.3 cde	39.0 cd	81.7 ab	
Sprig	0.0 a	0.2 e	0.7 c	0.7 e	1.3 d	11.7 c	

Table 7. Zoysiagrass cover estimates of companion crop treatments planted in the fall and spring in Yadkin County, NC, during year 2. Cell color represents a similar statistical group within the column, usually the top performers.

Planting Material	Planting Season	5/1/2023	8/1/2023	9/1/2023
----- % cover -----				
Fall				
Drill		0.0 a	13.7 ab	23.3 abc
High Wild		0.0 a	4.3 ab	10.0 bc
Low Wild		0.0 a	17.3 a	38.3 ab
Millet		0.0 a	6.7 ab	9.3 bc
Seed		0.0 a	15.0 ab	51.7 a
Sprig		0.0 a	0.2 b	0.7 c
Spring				
Drill		-	-	38.3 ab
High Wild		-	-	1.3 c
Low Wild		-	-	12.0 bc
Millet		-	-	3.3 c
Seed		-	-	22.7 abc
Sprig		-	-	0.3 c

### ***Moore County (Jackson Springs, NC) Plantings***

Treatments planted in Moore County during year 1 had poor ( $\leq 15\%$ ) zoysiagrass cover through 80 WAFP (Table 8). At 97 WAFP, the spring-planted low wildlife mix treatment had 52% zoysiagrass cover, which was  $\geq 29\%$  zoysiagrass cover compared to all other fall and spring treatments. All other fall and spring-planted treatments had  $\leq 13\%$  zoysiagrass coverage through 97 WAFP. The spring-planted low wildlife mix may have increased the soil's organic matter content and water holding capacity. The wildlife mix contained multiple legumes that may have increased soil nitrogen content. These increases in edaphic properties with minimal competition likely increased zoysiagrass seed establishment.

All treatments planted in Moore County during year 2 had similar ( $\leq 17\%$ ) zoysiagrass cover at 46 WAFP (Table 9). All companion crop treatments had similar cover and were not different from the no companion crop treatment. However, differences between treatments are anticipated during the second year of evaluation.

The Moore County site received 113 and 105 cm of precipitation in year 1 and 2, respectively. The Yadkin County site received 95 and 99 cm in year 1 and 2, respectively. Despite receiving 6 to 19% less precipitation than the Moore County site, the Yadkin County site had greater zoysiagrass cover compared to the Moore County site. In Yadkin County, the soil has a greater clay content than Moore County; therefore, more available water at or near the soil surface was available and yielded greater zoysiagrass cover. There were no differences among companion crop treatments in heavier soils by 97 WAFP. Therefore, companion crops in loamy soils may not be justified because of the soil's inherent higher water holding capacity and cation exchange capacity. However, the low wildlife companion crop demonstrated some promise in zoysiagrass establishment in sandy soils. The observed faster establishment in heavier loamy soils compared to sandy soils agrees with Milla-Lewis and Miller's (2020) results.

Table 8. Zoysiagrass cover estimates of companion crop treatments planted in the fall and spring in Moore County, NC, during year 1. Cell color represents a similar statistical group within the column, usually the top performers.

Planting Material	Planting Season	6/1/2022	7/1/2022	10/1/2022	2/1/2023	5/1/2023	9/1/2023
----- % cover -----							
Fall							
Drill		0.8 a	3.0 a	1.3 a	1.3 b	3.3a	1.7b
High Wild		0.7 a	0.5 a	2.3 a	8.0 ab	10.7a	7.3 b
Low Wild		1.3 a	2.0 a	4.5 a	7.3 ab	11.7a	7.0b
Millet		0.5 a	1.0 a	1.8 a	5.0 ab	8.0a	2.7b
Seed		0.3 a	0.7 a	1.0 a	2.7 ab	5.3a	2.7b
Sprig		0.5 a	0.5 a	1.3 a	0.3 b	2.8a	8.0b
Spring							
Drill		0.5 a	0.4 a	0.7 a	1.7 ab	5.2a	3.7b
High Wild		0.8 a	0.7 a	1.3 a	3.3 ab	9.3a	2.7b
Low Wild		0.8 a	1.7 a	5.8 a	11.7 a	15.0a	52.0a
Millet		0.5 a	0.4 a	1.0 a	2.0 ab	3.7a	1.7b
Seed		1.7 a	1.8 a	3.7 a	7.0 ab	15.3a	13.0b
Sprig		0.4 a	1.0 a	2.8 a	1.7 ab	13.3a	11.3b

Table 9. Zoysiagrass cover estimates of companion crop seeding in the fall and spring in Moore County, NC, during year 2.

Planting Material	Planting Season	9/1/2023 ---- % cover ----
	Fall	
Drill		12.0 a
High Wild		6.7 a
Low Wild		6.7 a
Millet		3.3 a
Seed		1.7 a
Sprig		2.7 a
	Spring	
Drill		9.0 a
High Wild		10.3 a
Low Wild		6.7 a
Millet		1.7 a
Seed		13.0 a
Sprig		16.7 a

## **CHAPTER 3: DEVELOPMENT AND SELECTION OF NEW ZOYSIAGRASS GERMPLASM**

### **3a. Development and evaluation of vegetative breeding lines**

#### Materials and Methods

Ten coarse-textured zoysiagrasses were selected based on their aggressive growth habit. These lines were crossed with lines with superior freeze tolerance. A total of 362 new hybrids were recovered from these crosses and planted in the spring of 2021 in unreplicated nurseries at the Upper Mountain Research Station (Laurel Springs, NC), The Sandhills Research Station (Jackson Springs, NC), and the Turfgrass Field Lab (Raleigh, NC). Plots were evaluated 2021-2023. During establishment year, plots were evaluated on a monthly basis for establishment speed (percent green cover) on a scale of 1 to 5, where 1 = no lateral spread and 5 = complete plot coverage. A last establishment rating was collected in the spring of year 2. For winter survival, entries were evaluated on a scale of 1 to 9, where 1 = 100% injury and 9 = 0%injury at all locations. Drought tolerance was evaluated at Jackson Springs only, where irrigation was turned off in the spring of each year after the plots had completely come back from dormancy. Plots were then rated for turf quality after two weeks of no rain on a scale of 1 to 9, where 1= dead and 9 = completely green. At least one drought period occurred under each year of evaluation. Additionally, plots were rated for turf quality at all location on a scale of 1 to 9, where 1 = poor and 9 = excellent turf. Large patch and leaf spot incidences were observed at Laurel Springs in 2021 and 2022, respectively. Plots were rated for resistance on a scale of 1 to 9, where 1 = severe damage and 9 = unaffected. At the end of 2023, the full data set was evaluated to select lines with superior aggressiveness and stress tolerance.

#### Findings and Conclusions

Establishment rates were fastest at the Raleigh nursery and slowest at Laurel Springs (LS) with Jackson Springs (JS) falling in between. These differences can be attributed to the lower temperatures at LS and the drought stress at JS. However, lines with superior lateral spread as compared to the checks were identified at all locations. When looking at performance across years and locations, several lines outperformed all checks, including Lobo, which is known for its aggressive lateral growth. The images below show averages across locations by year for all checks and the top ten performing lines.

Entry	Raleigh		Jackson Springs		Laurel Springs		OVERALL
	2021	2022	2021	2022	2021	2022	
Empire	3.5	5	1.67	3.67	2	5	3.47
Innovation	3	4.5	3.00	4.00	2.5	5	3.67
Jamur	4	5	1	4	1.5	2	2.92
Meyer	2	4.5	2	4	2.5	5	3.33
LOBO	3	4.5	3.33	4.00	2.5	5	3.72
XZ 14070	4	5	1.5	4.5	2.5	3.5	3.50
Zeon	2	3	1	1	1	1	1.50
XZ21038	4	5	4	5	3	5	4.33
XZ21047	4	5	4	5	2	5	4.17
XZ21071	4	5	4	5	2	5	4.17
XZ21139	4	5	4	4	3	5	4.17
XZ19100	3	5	3	5	3	5	4.00
XZ21028	4	5	3	4	3	5	4.00
XZ21033	4	5	3	5	2	5	4.00
XZ21036	4	5	3	4	3	5	4.00
XZ21037	4	5	3	5	2	5	4.00
XZ21046	3	5	4	4	3	5	4.00
XZ21048	4	4	4	5	3	4	4.00
XZ21070	4	5	4	3	3	5	4.00
XZ21073	4	5	3	4	3	5	4.00
XZ21142	4	5	4	4	2	5	4.00

Means for establishment rates for 2021 and 2022 at all three locations for commercial checks and the top ten performing lines. Plots were evaluated on a scale of 1 to 5, where 1 = no lateral spread and 5 = complete plot coverage. Cells highlighted in pink represent values one standard deviation above the mean.



Nursery plots at the Turf Field Lab (Raleigh, NC) showing distinct differences in establishment rates among entries.

In terms of drought tolerance, several lines outperformed the checks including Jamur, which was the drought control. Line XZ 18014 appears especially promising as it not only obtained high values but also it had the most stable performance across years.

Entry	DROUGHT TOLERANCE			OVERALL
	2021	2022	2023	
Empire	4.00	6.00	4.00	4.67
Innovation	3.00	3.00	3.00	3.00
Jamur	6	6	3.5	5.17
Meyer	6	4	3.5	4.50
LOBO	4.00	4.33	3.67	4.00
XZ 14070	4.5	4	2.5	3.67
Zeon	1	1	1	1.00
XZ18014	7	7	5	6.33
XZ21071	7	8	2	5.67
XZ21033	5	7	5	5.67
XZ18010	7	5	5	5.67
XZ19026	8	5	4	5.67
XZ19028	6	7	4	5.67
XZ18008	6	6	4	5.33
XZ18009	7	5	4	5.33
XZ19065	5	7	4	5.33

Drought tolerance scores for 2021-2023 at the Sandhills Research Station for commercial checks and the top nine performing lines. Plots were rated for turf quality after two weeks of no rain on a scale of 1 to 9 where 1= dead and 9 = completely green. Cells highlighted in pink represent values one standard deviation above the mean.

When considering performance across traits including establishment rate, drought tolerance, winter survival, overall turf quality, and disease response, 12 lines were selected for advancement based on performance across traits, locations, and years. These lines were propagated in the greenhouse. In late spring 2024, these will be transplanted in two roadside locations selected based on the diverse climatic regions of NC. These sites will need to be prepared for planting by NCDOT personnel by spraying out existing vegetation and tilled for seedbed preparation. Breeding lines will be sodded (or plugged, depending on availability of planting material) at these sites. Commercial checks will be included for comparison. Plots will be evaluated monthly for coverage. In fall 2024, these lines along commercial cultivars Compadre, El Toro and Lobo were plugged into 4x4 ft plots with six replications at two roadside locations: along US-70 in Goldsboro and US-421 I Yadkinville. These sites were selected based on the diverse climatic regions of NC. The first Spring visit to the western and eastern sites was on April 15th and April 22<sup>nd</sup>, respectively. All plugs survived post planting despite the zero-input management, indicating these materials show great promise for roadside applications. Evaluation of these nurseries will continue beyond the life of this project.

**3b. Development and evaluation of seeded populations**

Materials and Methods

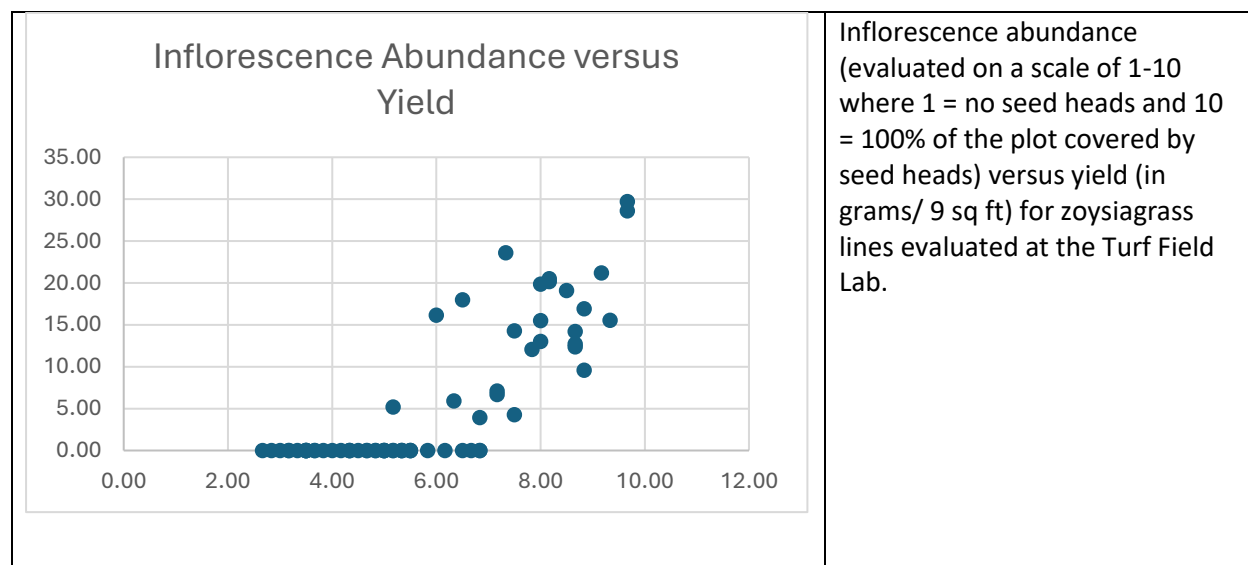
Advanced lines selected for their superior aggressiveness and abiotic stress tolerance were planted in a replicated field trial in 2019. The most prolific inflorescence producers were selected and harvested in fall 2020. Seed was threshed, cleaned, and processed for planting at the Turf Field Lab (Raleigh, NC) for seeding in late August. Plots were to be evaluated for germination ability, speed of establishment, and uniformity over the summer of 2021.



Additionally, a crossing program was initiated during late summer 2021 to develop populations for evaluation of yield and establishment rates and to identify the best combination of parents. For this purpose, the top-yielding lines from the 2019 Zoysia Advanced Trial were selected as parents, and a few commercial cultivars were also included. All entries were crossed against each other as both males and females. Seed heads were harvested from each individual female, and each female x male crossing was considered a family. Seeds were processed, cleaned, and stored. Initially, one hundred seeds per family were germinated according to Qian et al (2013). In a second round of germination tests, all seeds from each individual family were germinated. Individuals were tagged individually and grown in individual pots in the greenhouse. Families will be planted at the Lake Wheeler Turfgrass Field Lab in a replicated trial.

Findings and Conclusions

Significant ( $p = 0.05$ ) variation among entries was observed for both inflorescence abundance and seed yield. Only lines with abundance above a score of 5 out of 10 were harvested. Only two lines with high abundance and yield were identified. Seed across reps was bulked and processed for planting in 2021. A seeded trial was established in August 2021 at the Turf Field Lab. However, the trial was lost due to heavy rains post-planting. Soil was washed on top of the plots, and no germination was observed. Saved seed was used to conduct germination tests to check seed quality. Results indicated that the correlation between yield and seedling recovery was low.



The crossing program for seeded materials was completed in December 2022. A total of 769 crosses were made among 14 parents in all possible combinations. Seed was harvested and processed in late December. The table below summarizes the amount of seed recovered from each cross. Some parents were prolific as both females and males, some only as one of those, and some as neither. Regardless of family structure, a large number of progenies were produced.

	XZ 150060	XZ 150163	XZ 150290	XZ 150329	XZ 150351	XZ 150416	XZ 150653	XZ 150675	XZ 150783	XZ 150793	XZ 151116	XZ 151150	XZ 151191	JAMUR	TOTAL
XZ 150060		298	479		22	76		78	362	28		1299	322		2964
XZ 150163			358		70	173		49	335	17	37	1094	76		2209
XZ 150290		182			61	132		56	113	15	40	725	73		1397
XZ 150329		233	60			33		32	122	12		266		69	827
XZ 150351		240	559			231		49	301	69	32	484	28	67	2060
XZ 150416		291	368		93			50	199	34	60	381	60		1536
XZ 150653		532	495		137	235		68	389	36	69	741	64		2766
XZ 150675		226	429		82	185			163	30	46	761	46		1968
XZ 150783		47	115		57	100		25		24		393	134		895
XZ 150793		118	408		14	53			139			475	58		1265
XZ 151116		114	120		24	94		22	183	29		154	22		762
XZ 151150		321	170		54	150		104	173	29	20		54		1075
XZ 151191		80	127			12			233	19	20	576			1067
JAMUR						100			99			44			243
TOTAL	0	2682	3688	0	614	1574	0	533	2811	342	324	7393	937	136	21034

Number of seeds produced from each female (leftmost column) by male (top row) combination. Black cells indicate no seeds resulted from those crossings.

Results after the first round of germination (Table 10) indicated that high variability exists for seedling recovery among the limited materials evaluated. We obtained 641 seedlings from 8600 seeds (100 for each of 86 crosses) included for germination, which represents an average 7.5% recovery rate. Line XZ 151150 (red boxes on Table 10) was found to have much higher recovery rates, however.

**Table 10.** Results from diallel mating among zoysiagrass lines with the highest inflorescence production. Only families with 50-100 seeds were included for germination. Numbers before and after the slash in each cell indicate the number of seeds produced and the percentage of seedlings recovered, respectively. Totals for number of seeds obtained, number of seedlings recovered, and final recovery rate across families are highlighted in yellow. Families with a >25% recovery rate are highlighted in green.

	XZ150060	XZ150163	XZ150290	XZ150329	XZ150351	XZ150416	XZ150653	XZ150675	XZ150783	XZ150793	XZ151116	XZ151150	XZ151191	JAMUR	TSP	TslingR	ARR
XZ150060		298/2	479/2	0	22	172/9	0	78/0	362/12	28	0	1299/46	322/13	0	2964	84	12.0
XZ150163	0		358/0	0	70/0	173/1	0	49	335/1	17	37	1094/21	76/0	0	2209	23	3.8
XZ150290	0	182/0		0	61/0	132/1	0	56/0	113/2	15	40	725/13	73/0	0	1397	16	2.3
XZ150329	0	23	165/0		0	33	0	32	122/1	12	0	266/3	0	69/0	827	4	1.0
XZ150351	0	240/0	559/0	0		231/0	0	49	301/0	69/0	32	484/0	28	67/0	2060	0	0.0
XZ150416	0	291/0	368/37	0	93/2		0	50/16	199/24	50/12	60/0	381/45	60/7	0	1536	148	16.4
XZ150653	0	532/0	495/1	0	137/0	235/6		68/0	389/0	36	69/0	741/12	64/0	0	2766	19	2.1
XZ150675	0	226/0	429/1	0	82/0	185/0	0		163/0	30	46	761/24	46	0	1968	25	4.2
XZ150783	0	47	115/0	0	57/2	100/0	0	25		24	0	393/27	134/4	0	895	33	6.6
XZ150793	0	118/2	408/6	0	14	53/4	0	0	139/3		0	475/29	58/3	0	1265	47	7.8
XZ151116	0	114/0	120/1	0	24	94/0	0	22	183/4	29		154/24	22	0	762	29	5.8
XZ151150	0	321/33	170/59	0	54/11	150/37	0	104/7	173/33	50/8	20		54/13	0	1075	201	25.1
XZ151191	0	80/0	127/0	0	0	12	0	0	233/1	19	18	576/11		0	1067	12	3.0
JAMUR	0	0	0	0	0	100/11	0	0	99/17	0	0	50/14	0		243	43	14.3
TSP	0	2682	3688	0	614	1574	0	533	2811	342	324	7393	937	136	21034	641	7.5
TslingR	-	37	107	-	15	58	-	23	81	20	0	255	45	0			
ARR	-	3.7	8.9	-	2.1	5.3	-	4.6	6.2	6.7	0	19.6	5.6	0			

\* TSP= total seeds produced, TslingR = total number of seedlings recovered, ARR = average recovery rate

Due to low recovery rates in the first round of germination experiments, all extra seeds were germinated in a second round of experiments. A total of 1,008 individuals were recovered from 116 individual families.



Germination process for families recovered from the seeded crossing program. Seeds harvested from each female x male pairing were considered a family and were germinated in petri plates. Once seedlings emerged, they were transferred to float trays where families were planted in the same row. Once seedlings were at the 6-8 leaf stage, they were transplanted into individual pots.

Ultimately, 20 families were chosen based on recovery rates. These families [20 families with 24 genotypes per family, making a total of 480 genotypes], 11 parents, and two commercial cultivar checks (Compadre and Zenith) were planted in the field in a randomized complete block design with four replicates in the summer of 2024. For families, parents, and checks, each plot consisted of six individual genotypes. All 4 x 4 ft plots were established using 5 x 5 cm plugs. Phenotypic evaluations will be conducted per plant/individual for three years, where in year 1 the primary focus will be to evaluate the rate of establishment, and in years 2-3, inflorescences will be harvested and dried to estimate seed yield. Families with superior yield, germination, and establishment rates will be advanced. These populations will lay the foundation for the development of seeded zoysiagrass cultivars that are not only adapted to North Carolina, but that have the ability to establish and persist under roadside conditions.

## **RECOMMENDATIONS**

These recommendations are based on previous experience that indicates zoysiagrass is an excellent turfgrass species for use on NC roadsides, with its greatest limitation perhaps being the ability to effectively (timely and economically) establish.

1. Research results from the establishment timing study suggested that seeding may be the most effective way to establish zoysiagrass on NC roadsides, although our experiences also demonstrated the limitations in zoysiagrass seed availability and reminded us that selection of seeded cultivars limits NC-DOT to only two cultivars (limits diversity for varied environments).
2. Vegetative establishment from sprigs is a viable option for zoysiagrass establishment, although it has many challenges.
  - a. The primary challenge is soil water availability to sustain young sprigs. This research demonstrated that this is a more significant challenge when establishing zoysiagrass on sandy soil with low water holding capacity. Our recommendation is to concentrate most of the spring-planting in areas with “heavier soils” and perhaps seeding in areas with “sandier soils”. More liberal use of post-planting watering should also be a consideration.
  - b. The second challenge is achieving a quicker establishment. This may be offset with improved germplasm as well as by varied post-establishment cultural practices (using post-plant watering and/or fertilization, etc.). Our recommendation would be to continue to refine the sprigging and seeding equipment that was utilized in this project, along with evaluation of several zoysiagrass seeding rates.
3. Spring and fall planting resulted in similar establishment in heavier soil.
4. Previous research identified zoysiagrass breeding lines with excellent lateral spread. While two lines (including XZ 14069, which was commercially released as LOBO™) were selected for continued roadside work, the lack of stringent drought tolerance evaluations in their original selection resulted in their persistence not being high enough for roadside applications. In this project, we increased the stress level in drought evaluations and selected materials based solely on that trait in combination with lateral spread. We have identified 12 lines that should possess the persistence needed for establishment under minimal irrigation. We will continue evaluation of these lines on roadside nurseries to confirm those results.
5. While seeded zoysiagrasses are highly desirable for the overall turf industry, limited information exists on the genetic and physiological components underlying important seed traits like yield, dormancy, and germination. This has hindered the development and commercialization of high yielding seeded cultivars. Important information was gathered through this project on yield components and the relationship between inflorescence abundance, yield, and seedling recovery. Furthermore, we were able to identify good parents for those traits and develop 20 families with improved seedling recovery. These newly developed materials will be used for phenotypic recurrent selection, which ultimately will result in the development of experimental seeded varieties for roadside applications.

## **IMPLEMENTATION AND TECHNOLOGY TRANSFER PLAN**

Findings from this research indicate that zoysiagrass is a viable turfgrass for use on NC roadsides, but that rate of establishment (includes planting method, germplasm, and environmental conditions factors) is the greatest detriment to its more widespread adoption. This research investigated seeding and sprigging with some level of success in operational-sized plots. The absence of water/rainfall at the time of establishment, especially with sandy soils, still contributes to the limited success of sprigging and to a lesser extent, seeding.

Hundreds of experimental vegetative zoysiagrass materials were evaluated in nurseries, and promising lines with excellent persistence and drought tolerance were identified. Continued evaluation of these 12 lines on operational-sized plots will confirm their suitability for roadside applications. Furthermore, this project established the foundation for the development of seeded zoysiagrass cultivars by generating populations with superior yield and germination rates.

Another establishment method that offers some promise is gap sodding, which is a method that space-plants sod pieces or strips at intervals and relies on its ability to spread over non-planted areas. Sodding offers some advantages over sprigging since some roots are intact with the soil that is part of the sod. This helps to extend the period of time from planting until additional moisture is required for the newly planted turfgrass. All other inputs being equal, the more aggressiveness of newer germplasm will allow quicker establishment, what has been previously measured with older cultivars.

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Table A2. Cover estimates of zoysiagrass planted in summer, fall, or spring in Harnett County (Broadway), NC during year 1 (July 2021-April 2022) pooled over five planting materials.

Planting						
Season	5/1/2022	7/1/2022	10/1/2022	2/1/2023	5/1/2023	9/1/2023
	----- % cover -----					
Summer	11.8 a	21.6 a	17.28 a	19.93 a	44.87 a	78.93 a
Fall	1.69 b	8.81 b	12.94 a	15.15 a	14.23 b	43.31 b
Spring	0.18 b	3.19 b	6.13 a	9.02 a	7.77 b	20.68 c

Brown top millet 50 lb VNS.

Fall Wildlife Mix 50 lb mix: 52% rye (VNS); 19% Austrian winter pea, 7.5 oat (VNS), 4.95% red clover (VNS), 3.75% Dixie crimson clover, 2.25% Trophy Buckbuster rape seed, 2% triticale (VNS).

