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Establishment of mixed native warm-season grass forage utilizing two nurse crops

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Abstract

Establishing native warm-season grasses (NWSGs) can be challenging due to their small seed size and slow growth, which necessitate comprehensive weed control and minimal defoliation during the seedling year. Use of nurse crops may partially alleviate both of these issues. Two nurse crops, browntop millet (*Urochloa ramosa* (L.) Nguyen; BTM) and Ray's Crazy Mix (a warm-season diversity blend of cereal, legume, broadleaf, and brassica species; diversity mix [DM]), were evaluated for their potential to provide forage while reducing weed competition and enhancing NWSG recruitment. Six treatments, four non-grazed (NG) nurse crops (BTM, DM, no nurse crop/no imazapic herbicide [NNC-NI]), and no nurse crop with imazapic (NNC-I), and two grazed (G) treatments (BTM-G and DM-G) were replicated four times in a randomized block design. A blend of three NWSGs was drilled 2 weeks before the nurse crop planting. Overall, BTM and DM reduced ($p < 0.05$) NWSG seedling density compared to NNC-I, but not NNC-NI. Weed cover rating was greater in NNC-NI (80%), followed by both nurse crops (40%), and lastly by NNC-I (20%). Grazing between 7 and 10 weeks after planting (WAP) reduced ($p < 0.04$) leaf area index by 70% just after grazing and 40% by 15 WAP. Grazing did not affect NWSG populations but did have an impact on weed cover rating at 12 WAP, where G treatments had an 8% lower weed cover than NG treatment (42% vs. 50%). With well-timed grazing, nurse crops can produce desirable forage and contribute to reduced weed pressure but also may impose substantial competition on NWSG seedlings.

Plain Language Summary

Establishing native warm-season grasses (NWSGs) can be challenging due to their small seed size and slow growth, which necessitate comprehensive weed control and minimal defoliation during the seedling year. Use of nurse crops may partially alleviate both these issues. Two nurse crops, browntop millet and a warm-season diversity

Abbreviations: BBS, big bluestem; BTM, browntop millet; DM, diversity mix; IG, Indiangrass; LBS, little bluestem; NG, non-grazed; NI, no imazapic; NNC, no nurse crop; NWSG, native warm-season grass; WAP, weeks after planting.

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blend of cereal and broadleaf species, were evaluated against the herbicide imazapic to provide forage while reducing weed competition. Overall, NWSG populations were greatest in the imazapic treatment but were similar between nurse crops and the control. Weed canopy rating was lowest in the imazapic treatment and highest in the control with both nurse crops being intermediate between the two. Grazing 7–10 weeks after planting did not affect NWSG populations but did reduce weed canopy cover. With well-timed grazing, nurse crops can produce desirable forage and contribute to reduced weed pressure, but otherwise, they impose substantial competition on NWSG seedlings.

1 | INTRODUCTION

Native warm-season grass (NWSG) prairies were a major ecosystem located throughout much of the North American continent from Canada to Mexico and from the US Midwest to the East Coast of the United States (Mitchell & Britton, 2000). At present, nearly all of the land that was once native grassland has been converted to other land use with the majority relating to crop and forage production (Barnes, 2004). These changes have led to a loss of 98% of native grasslands in the United States and high levels of fragmentation of those that remain (Barnes, 2004; Miller, 1998). This loss, along with a growing shift towards more ecologically sustainable agricultural practices, has increased interest in the re-establishment of native grasslands over the last 50 years. In general, re-establishment has gained the most traction in the context of biofuel production (Backus et al., 2017; Hedtke et al., 2014; Zhang et al., 2015), wildlife habitat (Temu et al., 2015; West et al., 2016; Yeiser et al., 2015), cattle forage (Anderson, 2000; Brazil et al., 2020; Keyser, Holcomb, et al., 2016), or in combination under a working lands conservation model (Dertien & Baldwin, 2022; Iglay et al., 2019; Keyser et al., 2020).

The NWSG's big bluestem (*Andropogon gerardii* Vitman; BBS), little bluestem (*Schizachyrium scoparium* (Michx.) Nash; LBS), and Indiangrass (*Sorghastrum nutans* (L.) Nash; IG) have proven to be productive forage, producing 8900–11,200 kg ha⁻¹ and average daily gains in cattle of 0.89–1.33 kg day⁻¹ (Brazil et al., 2020; Burns & Fisher, 2013; Mitchell et al., 2005). However, they are slow to establish and typically do not produce forage during the seedling year (Doll et al., 2011; Török et al., 2012; Washburn & Barnes, 2000). This establishment gap creates a substantial obstacle for producers who cannot afford to lose forage production for an entire year (Meyer & Gaynor, 2002; Török et al., 2012). In the eastern United States, the typical establishment period for NWSGs is June–August. In these areas cool-season forages are often the dominant component of the forage system, so utilization of nurse crops that are available during that time-frame could help alleviate the forage gap. Another challenge

to successfully establishing NWSG is that their small seed size requires a high-quality seedbed consisting of fine-textured, firm soil that promotes seed soil contact and minimal weed pressure (Dewald et al., 1996; Keyser et al., 2019). The most common form of modern competition control is through the use of herbicides (Harper et al., 2002; Keyser et al., 2019; Miesel, 2012), although tillage (Barnes & Washburn, 2000; Doll et al., 2011), cover cropping (Hedtke et al., 2014; Keyser, Holcomb, et al., 2016; Richwine et al., 2021; Sadeghpour, Hashemi, DaCosta, Jahanzad, et al., 2014), mowing (Curran et al., 2012), and prescribed fire (Hall et al., 2012) have also been used in establishing NWSGs. Another option for competition control in more northern areas of the United States is to use cool-season crops typically used for pre-season weed suppression as nurse crops to provide early-season weed control for the NWSG seedlings (Haramoto & Gallandt, 2004; Sadeghpour, Hashemi, DaCosta, Gorlitsky, et al., 2014; Singh et al., 2003). Several Midwest US studies have successfully used barley (*Hordeum vulgare* L.) and oats (*Avena sativa* L.), as well as warm-season species such as corn (*Zea mays* L.) and sorghum (*Sorghum bicolor* L.) as nurse crops to achieve acceptable stands of NWSGs (Hintz et al., 1998; Jungers et al., 2015). In the eastern United States, relatively few studies have been conducted using nurse crops during NWSG establishment (Cossar & Baldwin, 2002; Richwine et al., 2021), and the majority have focused on establishing switchgrass [*Panicum virgatum* L. (SG)] (Cossar & Baldwin, 2002; Wolf & Fiske, 2009).

To that end, an experiment was conducted from 2019 to 2021 to explore NWSG establishment using two warm-season annual nurse crops that could reduce lost forage production during the seedling year. Our hypothesis was that both nurse crop treatments would reduce weed pressure but, in turn, reduce NWSG seedling density as seen by Richwine et al. (2021). As a monoculture, browntop millet's (BTM) canopy structure was hypothesized to become denser before grazing and therefore reduce density of both weeds and the NWSG seedlings more than the warm-season diversity mix (DM), which has a more heterogeneous canopy structure.

We also hypothesized that the grazed treatments would have greater NWSG seedling density than the NG plots by the end of the season due to the reduction in canopy density during a critical time in their development (Lawrence et al., 1995; Richwine et al., 2021). As the current best practice, the imazapic treatment was hypothesized to outperform the other treatments.

2 | MATERIALS AND METHODS

2.1 | Study area description

An NWSG field experiment was conducted twice in Brown County, Ohio (38.85° N, -84.03° W), on property owned and operated by the Greenacres Foundation. The first trial was conducted on a 4-ha field from March 2019 to August 2020. The experiment was repeated (Trial 2) on a second, 4-ha field at the same property from March 2020 to August 2021. The soils for both fields consisted of a poorly drained, Jonesboro-Rossmoyne silt loam (fine-silty, mixed, superactive, mesic Glossaquic Hapludalfs) with a pH of 6.4. Phosphorus (4.5 kg ha⁻¹ Morgan extraction method) and K (118 kg ha⁻¹ Morgan extraction method) were both in the adequate range based on soil test results conducted at Cornell Nutrient Analysis Laboratory, Cornell University. Both sites previously grew soybeans (*Glycine max* (L.) Merr.) for at least 5 years preceding the project.

On April 16, 2019, the field used for Trial 1 received an application of glyphosate (N-[phosphonomethyl] glycine, isopropylamine salt; 41%) at 2.3 L ha⁻¹ for initial competition control. A second round of glyphosate was applied at the same rate on June 10, 2019 followed by an application of CimarronPlus (metsulfuron-methyl methyl 2-[[[(4-methoxy-6-methyl-1,3,5-triazin-2-yl)amino]carbonyl]amino]sulfonyl]benzoate 48% and chlorsulfuron 2-chloro-N-[(4-methoxy-6-methyl-1,3,5-triazin-2-yl)aminocarbonyl]benzenesulfonamide 15%) at 35 g ha⁻¹ on July 1, 2019. The field used for Trial 2 received an application of glyphosate at 2.3 L ha⁻¹ on May 7, 2020 for initial competition control.

2.2 | Experimental design

Nurse crops used in this study were BTM variety not stated (VNS) and DM (Ray's Crazy Mix; Southeast AgriSeeds): 61% cowpea [*Vigna unguiculata* (L.) Walp], 11% NutriKing BMR6 sorghum × sudangrass [*Sorghum × drummondii* (Nees ex Steud.) de Wet & Harlan], 7% Daikon radish [*Raphanus sativus* (L.)], 7% hybrid pearl millet [*Pennisetum glaucum* (L.) R. Br.], 5% Sweet Six BMR6 sorghum × sudangrass, 4% Peredovik sunflower [*Helianthus annuus* L.], and 3% Win-

Core Ideas

- Native warm-season grass populations were greatest in the imazapic treatment but were similar between nurse crops and the control.
- Weed cover rating was lowest in the imazapic and highest in the control with both nurse crops being intermediate between the two.
- Grazing (from 7 to 10 weeks after planting) did not affect native warm-season grass populations but did reduce weed canopy cover.

fred hybrid brassica [*Brassica oleracea* var. sabellica × napus L.]. Imazapic was employed as a chemical weed suppression treatment for comparison. The study was comprised of six treatments: four NG treatments: BTM-NG, DM-NG, a no nurse crop/no imazapic (NNC-NI-NG) control, and no nurse crop with imazapic (NNC-I-NG), and two grazed treatments: BTM-G and DM-G. DM treatments were planted at 33 kg ha⁻¹ and the BTM at 11 kg ha⁻¹. Both nurse crop planting rates were reduced (60% and 40%, respectively) from the minimum recommended forage rate to limit canopy cover while still providing adequate grazing (King's Agriseeds, n.d.). The size of each experimental unit was 7.6 m × 61 m (NG) and 61 m × 61 m (G). The experiment was set up as a randomized block design with four blocks and repeated measures (trial week). Plant counts were conducted at 6, 9, and 12 weeks after planting (WAP). Each season, the six treatments were replicated four times for a total of 24 experimental units.

2.3 | Management and measurements

2.3.1 | Trial 1, 2019

On July 2, 2019, a blend of Prairie View BBS (6.7 pure live seed [PLS] kg ha⁻¹), Camper LBS (1.1 PLS kg ha⁻¹), and Prairie View IG (3.4 PLS kg ha⁻¹) purchased from Ernst Conservation Seed was no-till drilled into the entire trial area using a 3-m wide drill with 19-cm row spacing (Great Plains Min-Till Native Grass Series II Drill) at a rate of 11.2 kg ha⁻¹. On July 12, 2019, an application of imazapic (±)-2-[4,5-dihydro-4-methyl-4-(1-methylethyl)-5-oxo-1Himidazol-2-yl]-5-methyl-3-pyridinecarboxylic acid (Plateau) was applied at 0.29 L ha⁻¹ for pre-emergence weed control on the assigned treatments. On July 16, 2019, DM and BTM (Walnut Creek Seeds) were drilled using the same equipment as used to plant the NWSG. Grazing was accomplished using ten 340-kg lowline Angus (*Bos taurus*) feeder steers. Due to the limited stock available, the cattle were

rotated sequentially through both nurse crops within each block before being moved to the next block.

The paddocks were rotationally grazed based on plant maturity to a target residual canopy height of 36 cm beginning August 19, 2019. Once selected, cattle remained within a block until all treatments were grazed before moving to the next block. The cattle grazed through paddocks, on average, every 35 h until September 10, 2019. The grazing was originally scheduled to be completed between 6 and 8 WAP but had to be extended into week 10 to complete the grazing of all four blocks. Temporary paddocks were installed around each assigned grazing unit (61 m × 61 m) using single-strand step-in electric fencing. However, due to the explosive growth of both nurse crops and weeds early in the 2019 season, the paddocks of the third and fourth blocks were further partitioned into 30 m × 61 m grazing units to mitigate grazing selection and achieve even grazing.

2.3.2 | Trial 2, 2020

On June 9, 2020, the same NWSG blend was planted using the same protocols and equipment as in 2019. A 0.29 L ha⁻¹ application of imazapic was applied on June 24, 2020 for pre-emergence weed control on the assigned treatments. Both nurse crops (DM and BTM) were planted 17 days after the NWSGs, on June 26, 2020. In 2020, in order to achieve more timely grazing of all four blocks, a total of 20, 410-kg Angus cows were utilized. As was the case in 2019, cattle were rotationally grazed based on plant maturity with a target residual canopy height of 36 cm. The first round of grazing began on August 6, 2020 and ended on August 13, 2020. Plant growth during 2020 was both more gradual and sparser than in the 2019 season due to prolonged drought. These differences facilitated uniform rotation times as well as allowed for a second round of grazing, which began on August 16, 2020 and ended on August 23, 2020. The cattle rotated, on average, every 24 h. Both rounds of grazing took place between 8 and 10 WAP.

2.4 | Data collection

Data collection started at 3 WAP on July 23, 2019 and June 26, 2020 and was repeated every 3 weeks through 12 WAP (September 24, 2019 and August 31, 2020). Although we collected data beginning at 3 WAP, the small size of seedlings in 2019, which made identification to species uncertain, and a complete lack of seedlings in 2020 due to severe drought, we dropped this sample period from the analysis. Native grass seedling densities were counted per species. A visual weed canopy cover rating (WR) from 0% to 100% to the nearest 10% was also assessed by estimating how much of the

plant canopy consisted of weedy species leaf area. Weed ratings included any species not planted as part of the respective treatment and were taken starting at 6 WAP. We used weed canopy cover rather than ground cover because it represented the direct competition for light between the nurse crops, weeds, and NWSGs. Seedling counts and WR were conducted using a 0.50 m × 0.33 m quadrat, oriented with 0.50 m length perpendicular to the seed drill rows. This orientation allowed for three 0.33 m lengths totaling 1.0 m of drill row to be assessed per sample. Prior to the initial data collection each summer, eight randomly located sampling locations were established and used for subsequent sampling to monitor seasonal changes in density. Additionally, in 2020, canopy (leaf area index [LAI]) readings accounting for all plants within each sample area were collected on three different dates, using a LI-COR LAI-2200C Plant Canopy Analyzer (LI-COR). The readings were taken on August 3, 2020 (7 WAP, pre-grazing), August 28, 2020 (10 WAP, post-grazing), and September 18, 2020 (15 WAP, 6 weeks post-grazing). For each sample date, above- and below-canopy readings were taken to establish a ratio between the available sunlight to sunlight reaching ground level (i.e., m⁻² leaf cover per m⁻² of ground). Three readings were randomly taken within each plot. Although the LI-COR sensor was not available during the 2019 trial, grazing in 2020 was initiated and managed employing comparable plant stage (early-boot) and canopy closure (visually, >75%) targets to those utilized during the 2019 trial. Therefore, while direct comparisons cannot be made, the grazing management resulted in similar canopy removal and post-grazing conditions of nurse crops for both trials.

Air temperature and precipitation data were obtained from the National Oceanic and Atmospheric Administration (NOAA, 2024) weather station located in Maysville, KY (KZZ49, located 8 km southeast of the experiment site, 38.78° N 84.02° W, elev. 168 m) and compared to the 30-year means for that location (Figure 1).

2.5 | Statistical analysis

Plant density (combined NWSGs, BBS, LBS, and IG) and WR in the NG plots were analyzed using a mixed model analysis of variance (ANOVA) (PROC GLIMMIX) in SAS 9.4 (SAS Institute, 2023) to determine differences ($\alpha = 0.05$) among main effects and their interactions. Fixed effects were the NG treatments, weeks post-planting, trial year, and their interactions. An additional model was run on the 12 WAP (post-grazing) data to evaluate the results of grazing. This model included the same factors as the first model but dropped WAP and included BTM-G and DM-G as additional treatments for a total of six. The 2019 paddocks that required subdivision to achieve even grazing were initially

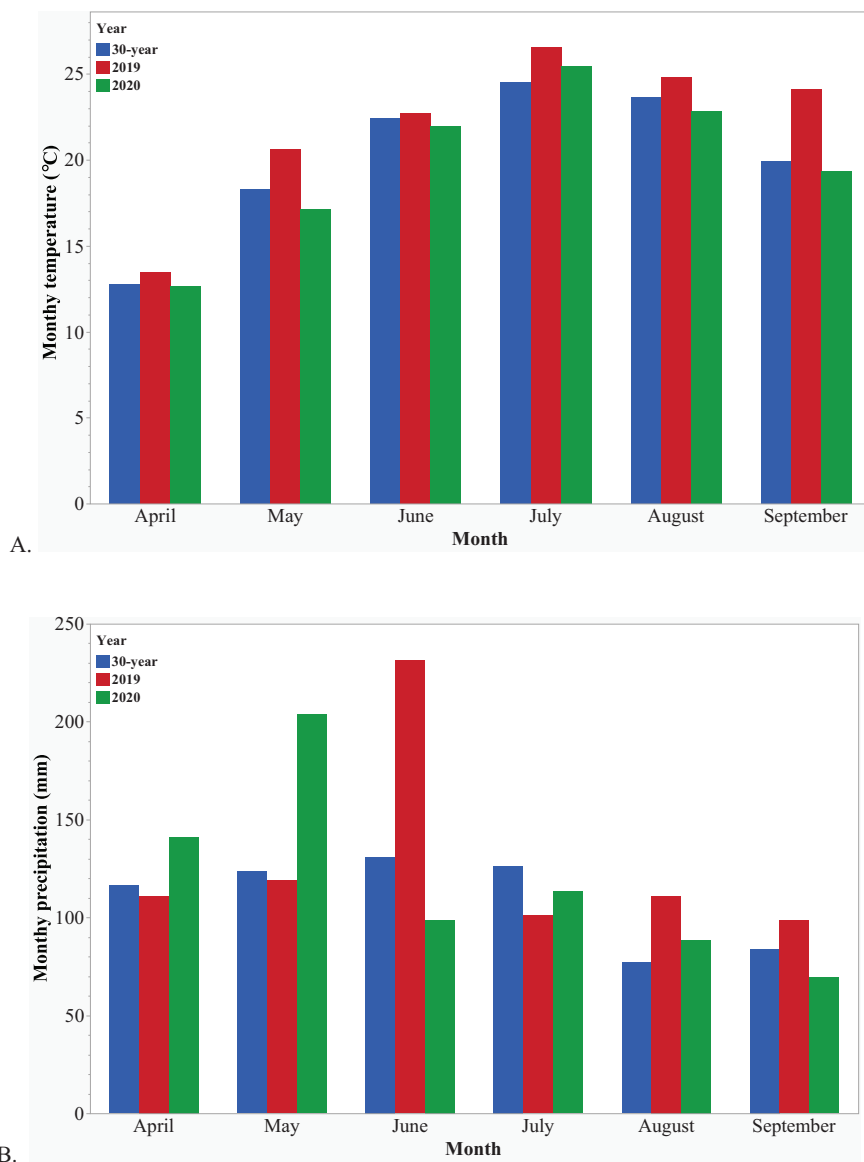


FIGURE 1 (A) Mean monthly air temperature (°C) and 30-year mean and (B) total monthly precipitation (mm) and 30-year mean for August, Kentucky (8 km from the field site), 1991–2020. Some months' data are missing in the overall 30-year period from 1991 to 2020.

analyzed separately using mixed model ANOVA, but when no differences were observed, they were reconstituted as one treatment for the final model. Year \times block, year \times block \times treatment, and year \times block \times treatment \times sample were random effects in both models. Data for LAI were analyzed independently using mixed model ANOVA (Proc MIXED). Fixed effects were treatment and date [pre-grazing and post-grazing (1 and 6 weeks)] with block as a random effect in each model. All models were examined for normality using the Shapiro–Wilk test and for equality of variances using the Levene test and conformed to these assumptions. Fisher's least significant difference was used for mean separations.

3 | RESULTS

3.1 | Environmental conditions

During the 2019 growing season (July through September), monthly air temperatures were above the 30-year mean (Figure 1). Monthly precipitation during the growing season remained within 30 mm of the 30-year mean with the exception of June, which had 230 mm compared to the 30-year mean of 130 mm (Figure 1). During the 2020 growing season (June through September), mean monthly air temperatures remained near the 30-year mean (Figure 1). Monthly precipitation was 20% below average in June and slightly below

TABLE 1 Analysis of variance (ANOVA) results for weeks 6–12 post-planting, total native warm-season grasses (Total), big bluestem (BBS), Indiangrass (IG), and little bluestem (LBS) densities, as well as a visual rating of weed canopy cover (WR) in non-grazed (NG) treatments. Treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Weed rating based on visual percent weed canopy cover per m^{-2} plot to the nearest 10%. Lewis Township native warm-season grass (NWSG) establishment trials, Brown County, Ohio, 2019 and 2020. * < 0.05, ** < 0.01, and *** < 0.001.

Effect	Total	BBS	IG	LBS	WR
Year	0.01*	0.00*	0.02*	0.22	<0.01**
Week	<0.01**	0.02*	<0.01**	<0.01**	<0.01**
Year × week	0.01*	<0.01**	<0.01**	0.23	<0.01**
Treatment	0.05*	0.01**	0.36	0.04*	<0.01**
Year × treatment	0.13	0.02*	0.64	0.00*	0.04*
Week × treatment	<0.01**	<0.0001***	0.24	0.17	<0.01**
Year × week × treatment	0.99	0.03*	0.20	0.63	<0.01**

average in July (Figure 1). In addition to the weather station data, staff at the experiment site noted a lack of rainfall on the property from the last week of May through the first week of July.

3.2 | Native grass density

3.2.1 | Nurse crops

Native grass densities varied for the combined (total) NWSGs as well as by individual species (Table 1).

Total NWSG species plant density remained stable through 9 WAP in 2019 but began to drop after 6 WAP in 2020 (Table 2).

Across both years, total NWSG populations interacted between nurse crop treatment and week with all treatments being the same at 6 WAP but by 9 and 12 WAP, densities were greater in NNC-I compared to the other treatments (Figure 2).

Individually, BBS and LBS were also affected by the nurse crop treatments (Table 1). A three-way interaction (year × week × treatment) was observed in BBS wherein plant density was greater in 2019 NNC-NI than in the DM and NNC-I at 6 WAP but not in 2020 (Table 2). By 9 WAP, however, populations in NNC-I plots became greater than the other treatments and remained so through 12 WAP. At 9 WAP, NNC-NI density was greater than DM but not that of BTM (Table 2). In 2020, no differences were observed within weeks among nurse crops for seedling density for BBS. IG populations differed between the first and second trial, but in both cases were greatest at 6 WAP and decreased over the course of each season (Table 2). LBS density interacted between treatment and year wherein NNC-I facilitated the highest populations, followed by DM and then BTM in 2019, while no differences were observed in 2020 (Table 2). The NNC-NI populations did not differ from either nurse crop in 2019 (Table 2).

3.2.2 | Weed rating

Weed canopy rating was affected by the three-way interaction of year, week, and treatment (Table 1). Weed canopy cover tended to be higher by the end of the season during both trials, but an exception was observed for DM both years wherein WR remained similar from 6 to 12 WAP. The WR of NNC-I also remained constant during the 2019 trial season. In 2019 at 6 WAP every treatment shared a similar WR (Figure 3).

The trend continued through 9 WAP. However, by 12 WAP, NNC-I had a lower WR than either BTM or NNC-NI but was still similar to DM (Figure 3). In 2019 both nurse crops were similarly effective at reducing WR and, at 12 WAP, had a WR between NNC-I and NNC-NI (Figure 3). In 2020, NNC-I had a lower WR at 6 WAP than NNC-NI but not the two nurse crops (Figure 3). By 9 WAP the highest WR was observed in the NNC-NI, a trend that continued through 12 WAP.

3.2.3 | Post-grazing

An interaction of treatment and date was observed for LAI (Table 3).

Prior to grazing, there was no difference observed between the BTM and DM treatments designated for grazing and those to remain NG (Figure 4).

Note that 2 weeks after grazing (August 28) there was a 60% numerical reduction of LAI in grazed BTM and an 80% reduction in the grazed DM plots compared to their NG counterparts (Figure 4). Note that 6 weeks after grazing (September 18), the grazed BTM paddocks still had a 40% numerically lower LAI than the NG paddocks, while the grazed DM paddocks maintained a 70% reduction (Figure 4). The week preceding grazing, NNC-I had a lower LAI compared to NNC-NI and both DM treatments but was similar to both BTM treatments. At 2 weeks post-grazing, NNC-I was similar to all treatments except ungrazed-diversity mix (UG-

TABLE 2 Mean density (m^{-2}), total native warm-season grasses (Total), big bluestem (BBS), Indiangrass (IG), and little bluestem (LBS) in non-grazed (NG) treatments. Time factors were year (2019 or 2020) and weeks after planting (6, 9, and 12). Treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). For clarity, letter groupings for three-way interactions are presented as combinations of week and treatment within year. Plant populations sharing the same letter within each interaction do not differ ($p < 0.05$). Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2019 and 2020.

NWSG	Year	Week	Treatment	Density	NWSG	Year	Week	Treatment	Density
BBS	2019	6	BTM-NI-NG	4.4 ± 1.5CE	IG	2019	6	–	14.2 ± 1.7A
BBS	2019	6	NNC-NI-NG	8.6 ± 1.5BC	IG	2019	9	–	9.6 ± 1.7B
BBS	2019	6	DM-NI-NG	3.0 ± 1.5E	IG	2019	12	–	5.5 ± 1.7CD
BBS	2019	6	NNC-I-NG	4.3 ± 1.5DE	IG	2020	6	–	3.4 ± 1.7C
BBS	2019	9	BTM-NI-NG	7.2 ± 1.5BD	IG	2020	9	–	1.9 ± 1.7CD
BBS	2019	9	NNC-NI-NG	8.9 ± 1.5B	IG	2020	12	–	1.4 ± 1.7D
BBS	2019	9	DM-NI-NG	4.1 ± 1.5DE	LBS	2019	–	BTM-NI-NG	0.5 ± 0.7C
BBS	2019	9	NNC-I-NG	16.4 ± 1.5A	LBS	2019	–	NNC-NI-NG	0.8 ± 0.7BC
BBS	2019	12	BTM-NI-NG	3.8 ± 1.5E	LBS	2019	–	DM-NI-NG	2.2 ± 0.7B
BBS	2019	12	NNC-NI-NG	6.0 ± 1.5CE	LBS	2019	–	NNC-I-NG	4.0 ± 0.7A
BBS	2019	12	DM-NI-NG	3.1 ± 1.5DE	LBS	2020	–	BTM-NI-NG	0.9 ± 0.7BC
BBS	2019	12	NNC-I-NG	14.5 ± 1.5A	LBS	2020	–	NNC-NI-NG	1.0 ± 0.7BC
BBS	2020	6	BTM-NI-NG	1.8 ± 1.5AB	LBS	2020	–	DM-NI-NG	0.8 ± 0.7BC
BBS	2020	6	NNC-NI-NG	3.5 ± 1.5AB	LBS	2020	–	NNC-I-NG	0.5 ± 0.7BC
BBS	2020	6	DM-NI-NG	4.8 ± 1.5A	Total	2019	6	–	21.7 ± 2.7A
BBS	2020	6	NNC-I-NG	1.3 ± 1.5AB	Total	2019	9	–	20.4 ± 2.7A
BBS	2020	9	BTM-NI-NG	1.3 ± 1.5AB	Total	2019	12	–	14.0 ± 2.7B
BBS	2020	9	NNC-NI-NG	0.2 ± 1.5B	Total	2020	6	–	8.4 ± 2.7B
BBS	2020	9	DM-NI-NG	0.3 ± 1.5B	Total	2020	9	–	3.1 ± 2.7C
BBS	2020	9	NNC-I-NG	2.1 ± 1.5AB	Total	2020	12	–	2.3 ± 2.7C
BBS	2020	12	BTM-NI-NG	0.0 ± 1.5B					
BBS	2020	12	NNC-NI-NG	0.0 ± 1.5B					
BBS	2020	12	DM-NI-NG	0.2 ± 1.5B					
BBS	2020	12	NNC-I-NG	2.8 ± 1.5AB					

TABLE 3 Analysis of variance (ANOVA) results for pre- and post-grazing leaf area index (m^{-2} of leaf cover per m^{-2} of ground). The time factor was date (August 3, 2020 [7 week after planting [WAP], pre-grazing], August 28, 2020 [10 WAP, post-grazing], and September 18, 2020 [15 WAP, 6 weeks post-grazing]). Non-grazed (NG) treatments were imazapic (I-NG), no-imazapic (NI-NG), browntop millet (BTM-NG), and warm-season diversity mix (DM-NG). Grazed (G) treatments were BTM-G and DM-G. Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2020. * < 0.05, ** < 0.01, and *** < 0.001.

Effect	Number of degrees of freedom (NDF)	F value	Pr > F
Date	2	6.9	<0.01**
Treatment	5	13.9	<0.01**
Date × treatment	10	13.9	0.04*

DM). At 6 weeks post-planting, NNC-I had an LAI similar to both grazed nurse crop treatments and a lower LAI than the other NG treatments (Figure 4). The LAI of NNC-NI was similar to both nurse crop treatments throughout the trial except at 6 weeks post-grazing wherein UG-DM had a greater LAI (Figure 4).

For the total as well as each individual NWSG species, 12-WAP densities in 2019 always exceeded those for 2020 (Table 4).

Post-grazing (12 WAP), density for Total NWSGs, as well as for all three species, differed by treatment (Table 4). The greatest native grass populations were observed in the NNC-

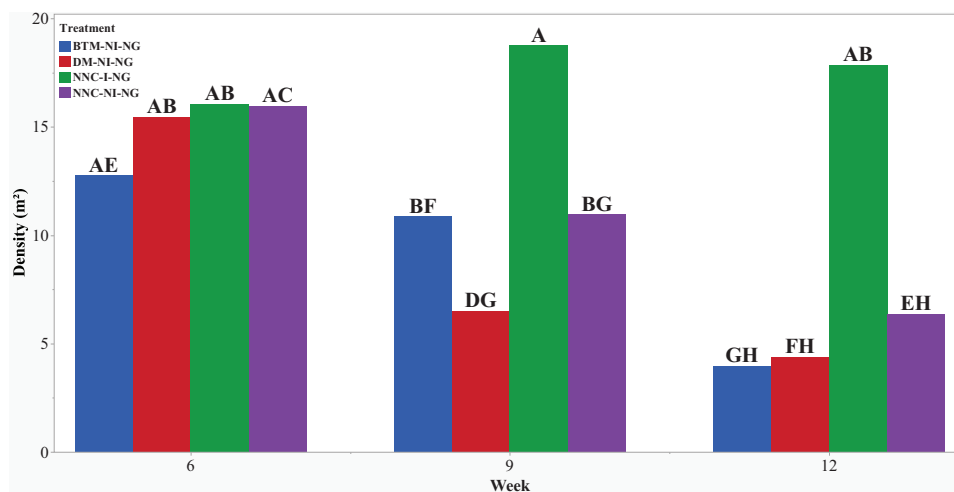


FIGURE 2 Mean density (m^{-2}) of total native warm-season grasses by weeks after planting (6, 9, and 12). Treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Plant populations sharing the same letter do not differ ($p < 0.05$). Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2019 and 2020.

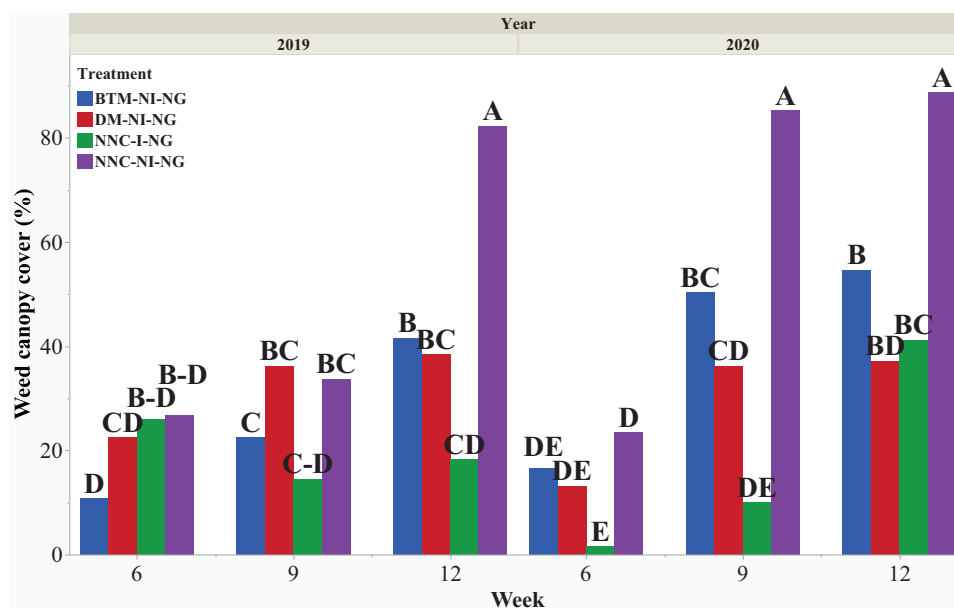


FIGURE 3 Mean percent weed canopy cover. Time factors were year (2019 or 2020) and weeks after planting (6, 9, and 12). Treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Means sharing the same letter within each year does not differ ($p < 0.05$). Weed rating based on visual percent weed canopy cover per plot to the nearest 10%. Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2019 and 2020.

I-NG treatment over those of both grazed and UG nurse crop treatments as well as the NNC-NI-NG (Figure 5).

Similar trends were observed for individual species with the exception of LBS, which had an interaction with both treatment and year (Table 5).

In 2019 the LBS population was greatest within the NNC-I treatment, while in 2020 no differences between treatments were observed (Table 5).

Post-grazing WR was influenced only by treatment with the highest LAI found in NNC-NI averaging double that of the

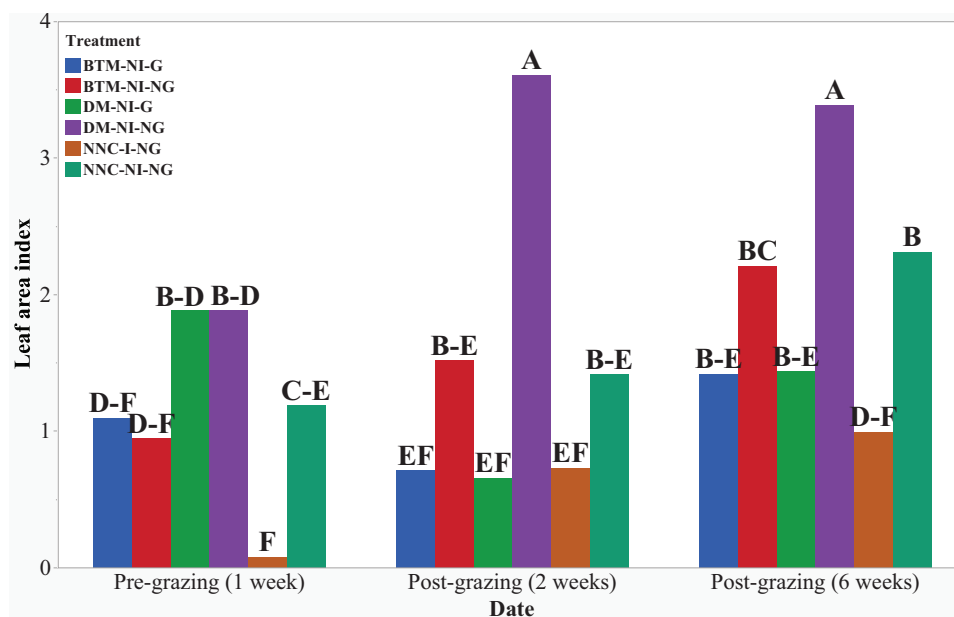


FIGURE 4 Mean leaf area index (LAI) (m^{-2} of leaf cover per m^{-2} of ground) per collection date (August 3, 2020 [7 week after planting [WAP], pre-grazing], August 28, 2020 [10 WAP, 2 weeks post-grazing], and September 18, 2020 [15 WAP, 6 weeks post-grazing]). Non-grazed (NG) treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Grazed (G) treatments were BTM-NI-G and DM-NI-G. Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2019 and 2020. Means sharing the same letter do not differ ($p < 0.05$).

TABLE 4 Analysis of variance (ANOVA) results, week 12 post-planting, total native warm-season grasses (Total), big bluestem (BBS), Indiangrass (IG), little bluestem (LBS), plant densities as well as a visual rating of weed canopy cover (WR). The time factor was year (2019 or 2020). Non-grazed (NG) treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Weed rating based on visual percent weed canopy cover per plot to the nearest 10%. Lewis Township native warm-season grass (NWSG) establishment trials, Brown County, Ohio, 2019 and 2020. * < 0.05 , ** < 0.01 , and *** < 0.001 .

Effect	Total	BBS	IG	LBS	WR
Year	0.02*	0.03*	0.04*	0.00**	0.09
Treatment	$<0.01^{**}$	0.01*	0.00**	0.00**	$<0.01^{**}$
Year \times treatment	0.21	0.22	0.68	0.01*	0.82

other treatments (Table 5). Weed rating did not statistically differ between the other treatments, notably between either nurse crop (Table 5).

4 | DISCUSSION

Research evaluating the use of nurse crops during NWSG establishment is limited, and studies examining specific nurse crops and associated grazing paradigms for NWSG establishment in the eastern United States have not been fully explored. Research conducted by Hintz et al. (1998) found success when using corn as nurse crop, and Keyser et al. (2016a) evaluated the use of harvestable small grains during SG establishment. Lawrence et al. (1995) compared the weed control capabilities of grazing versus atrazine (6-chloro-

N-ethyl-N'-(methyl-ethyl)-1,3,5-triazine-2,4-diamine) during BBS establishment but did not look at combinations of the two. Recently, Richwine et al. (2021) utilized BTM as a nurse crop when establishing BBS and SG, but their study evaluated BTM under mechanical harvest, not grazing.

The results of our study indicate that neither BTM nor DM appears to reduce early-season germination rates of the three native grass species, which is consistent with results observed by Hintz et al. (1998) when using corn as a warm-season nurse crop. However, in contrast with Hintz et al. (1998), but in line with our hypothesis, total and BBS populations did begin to decrease mid-season, and by season's end, all three species were negatively affected by both nurse crops in a comparable way to NNC-NI. This difference was likely due to a combination of the delayed grazing that allowed prolonged canopy closure of both nurse crops and weeds as well as the quick

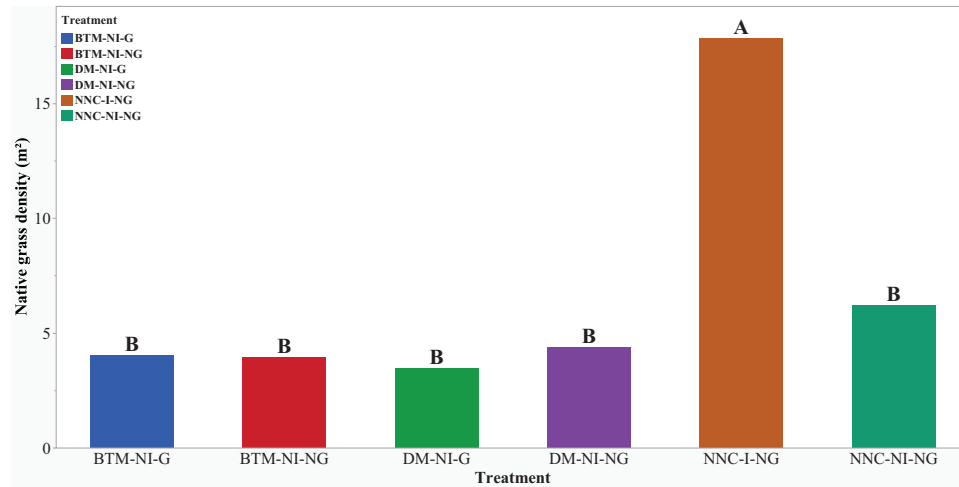


FIGURE 5 Mean density (m^{-2}) week-12 post-planting for total native warm-season grasses post-grazing. Non-grazed (NG) treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Grazed (G) treatments were BTM-NI-G and DM-NI-G. Plant populations sharing the same letter do not differ ($p < 0.05$). Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2019 and 2020.

TABLE 5 Mean density (m^{-2}), big bluestem (BBS), Indiangrass (IG), and little bluestem (LBS), as well as a visual rating of weed canopy cover (WR) at 12 week after planting (WAP). The time factor was year (2019 or 2020). Non-grazed (NG) treatments were no nurse crop imazapic (NNC-I-NG), no nurse crop no-imazapic (NNC-NI-NG), browntop millet without imazapic (BTM-NI-NG), and diversity mix without imazapic (DM-NI-NG). Grazed (G) treatments were BTM-NI-G and DM-NI-G. For clarity, letter groupings for three-way interactions are presented as combinations of week and treatment within year. Plant populations sharing the same letter do not differ ($p < 0.05$). Weed rating based on visual percent weed canopy cover per m^{-2} plot to the nearest 10%. Lewis Township native warm-season grass (NWSG) establishment trial, Brown County, Ohio, 2019 and 2020.

NWSG	Year	Treatment	Density	NWSG	Year	Treatment	Density
IG	2019	–	4.4 ± 0.9A	BBS	2019	–	6.0 ± 1.4A
IG	2020	–	1.0 ± 0.9B	BBS	2020	–	0.5 ± 1.4B
IG	–	NNC-NI-NG	0.3 ± 0.4B	BBS	–	NNC-NI-NG	3.0 ± 1.5B
IG	–	BTM-NI-G	0.6 ± 0.4B	BBS	–	BTM-NI-G	2.5 ± 1.5B
IG	–	DM-NI-G	0.2 ± 0.4B	BBS	–	DM-NI-G	1.9 ± 1.5B
IG	–	NNC-I-NG	2.5 ± 0.4A	BBS	–	NNC-I-NG	8.6 ± 1.5A
IG	–	BTM-NI-NG	0.07 ± 0.4B	BBS	–	BTM-NI-NG	1.9 ± 1.5B
IG	–	DM-NI-NG	0.07 ± 0.4B	BBS	–	DM-NI-NG	1.6 ± 1.5B
LBS	2019	NNC-NI-NG	0.5 ± 0.5B	Total	2019	–	11.7 ± 2.4A
LBS	2019	BTM-NI-G	1.2 ± 0.5B	Total	2020	–	1.6 ± 2.4B
LBS	2019	DM-NI-G	0.5 ± 0.5B	WR	–	NNC-NI-NG	85.8 ± 6.4A
LBS	2019	NNC-I-NG	4.6 ± 0.5A	WR	–	BTM-NI-G	46.3 ± 6.4B
LBS	2019	BTM-NI-NG	0.2 ± 0.5B	WR	–	DM-NI-G	36.3 ± 6.4B
LBS	2019	DM-NI-NG	1.0 ± 0.5B	WR	–	NNC-I-NG	29.8 ± 6.4B
LBS	2020	NNC-NI-NG	0.0 ± 0.5B	WR	–	BTM-NI-NG	47.4 ± 6.4B
LBS	2020	BTM-NI-G	0.0 ± 0.5B	WR	–	DM-NI-NG	37.4 ± 6.4B
LBS	2020	DM-NI-G	0.0 ± 0.5B				
LBS	2020	NNC-I-NG	0.5 ± 0.5B				
LBS	2020	BTM-NI-NG	0.0 ± 0.5B				
LBS	2020	DM-NI-NG	0.0 ± 0.5B				

regrowth of the nurse crops and weed species post-grazing. Interestingly, Hintz et al. (1998) observed that corn used as a nurse crop, even at the highest planting rates, did not negatively affect NWSG seedling establishment. This is likely because the corn in their study did not achieve a closed canopy until 8–10 WAP compared to the 5–6 weeks observed in this study.

In our study, BBS was negatively affected by the nurse crops earlier in the season compared to IG or LBS. An increase in competition for light during BBS's peak germination window may have suppressed recruitment compared to recruitment when using NNC-I. This reduction in NWSG recruitment due to increased competition is well understood as their early growth does not keep up with that of most summer annuals (Barnes et al., 2004; Martin et al., 1982; Vaughn et al., 2015). Comparatively, by 6 WAP in 2019, IG and LBS treated with imazapic had already achieved their seasonal peak population (19 and 4 plants m^{-2} , respectively) and, in the case of IG, began to gradually lose population density over the remainder of the season (8 plants m^{-2} by 12 WAP). LBS populations remained steady over the course of the season both years with NNC-I. The BBS populations treated with imazapic, however, increased 300% between 6 and 9 WAP going from 4 to 16 plants m^{-2} , while changing more modestly in the other treatments in the same period. This indicates that the bulk of the potential BBS germination took place while both nurse crops were at the point of thickest canopy shortly before grazing took place.

Extending the delay between the planting of NWSGs and the nurse crop by more than 2 weeks is not typically advisable under normal conditions as the NWSGs should have begun germination by 3 WAP. However, in situations where NWSG germination is expected to be delayed, planting the nurse crop on a similar delay may be advisable if predominantly BBS stands are desired. In addition, more developed NWSG seedlings may be more resilient to trampling, which has been a concern when grazing is employed during the seedling year (Launchbaugh, 1976; Stoddart, 1946).

Neither BTM nor DM appeared to benefit NWSG recruitment early on and actually began to overwhelm the seedlings later in the season. The nurse crops grew aggressively and greatly reduced early weed populations but, in doing so, took their place and, by 9 WAP, began to directly compete with the NWSG seedlings. Contrary to our hypothesis, both nurse crops resulted in similar NWSG densities by the end of the season. While detrimental to the NWSG populations, the nurse crops did not eliminate them. When establishing these NWSGs for forage, target end-of-season plant density is 11 plants m^{-2} (Keyser et al., 2016b). In 2019, total NWSG population density within BTM and NNC-NI remained sufficient at 9 WAP (11 plants m^{-2}) but their populations decreased by nearly half (5 plants m^{-2}) by the 12 WAP data collection, indicating that a timelier reduction of nurse crop canopies may

have allowed acceptable stands to persevere until the end of the season.

Weed canopy cover did not vary between nurse crops either year until 12 WAP. In 2019, both nurse crops had 20% more weed pressure than NNC-I but 40% less weed pressure than NNC-NI. In 2020, BTM reduced weed pressure by 35% compared to NNC-NI, while both DM and NNC-I reduced it by around 40%, suggesting that both nurse crops are capable smother crops even when planted at a reduced rate. These findings are in contrast with others that observed that nurse crops do not substantially reduce weed pressure (Hintz et al., 1998; Keyser et al., 2016a).

Grazing was effective, based on 2020 LAI data, at reducing canopy cover with the reduction persisting for up to 6 weeks after grazing. However, this same grazing (between 8 and 10 WAP) did not have an appreciable effect on NWSG density. This suggests that moderate grazing of these nurse crops earlier in the season, before the NWSG seedlings begin to be suppressed, could have provided a lasting advantage with minimal drawbacks. Grazing (especially in 2019) took place later and was over a longer period of time than initially intended. Even so, grazing was carried out in a way that may be analogous to a situation a producer might face under similar circumstances. During initial analysis, steps were taken to quantify any variability that may have been induced by the extended duration of the grazing treatments by including block as a fixed effect in the model as well as analyzing each block individually. Based on these preliminary tests, the differences in the timing of grazing from the early to latter part of the grazing period appeared to be largely captured by blocking (included as a random variable in the final model) and did not change our conclusions regarding grazing impacts. While delayed grazing would not be recommended, this study sheds light on what may happen if such circumstances are unavoidable.

At the time of grazing, BTM and DM were approximately 50–75 and 90–120 cm tall, respectively. Earlier grazing or grazing down to a height closer to that of the NWSG seedlings (23–31 cm tall) would have allowed the seedlings to maintain a better competitive growth position within the sward. Such prolonged and/or greater access to the canopy could allow greater seedling development. Additionally, the canopy in 2019 was already dense at 6 WAP (and 8 WAP in 2020) and did not materially change from the start to the completion of each year's grazing period (>75% in both cases). On the other hand, plant maturity did. At the onset of grazing in 2019, BTM was at anthesis while DM (the sorghum component) was at the late boot stage. In 2020 both BTM and DM were in the early boot stage at the onset of grazing. By the end of grazing 2019, BTM had reached the dough stage, while DM had entered anthesis. By the end of grazing in 2020, BTM was in the dough stage and DM was at anthesis or beyond.

At 2 weeks post-grazing, DM had a greater disparity in LAI between the grazed and NG treatments when compared to BTM or the two NNC treatments. At 2 weeks post-grazing, BTM-NG and both NNC-NGs were typically 20–30 cm taller than their grazed counterparts (76 vs. 46 cm), whereas the DM-NG was more than 120 cm taller than its grazed counterpart (213 vs. 92 cm). This is most likely due to the rapid regrowth observed in several species within the DM, with the most prominent being sorghum × sudangrass, which quickly began to reform a canopy well above tallest NWSG seedlings. Grazing did not appear to affect NWSG plant density at 2 weeks post-grazing but did affect WR, reducing it by 10% over the treatment. In this study, grazing provided adequate forage, maintained an open canopy, and did not damage or reduce NWSG recruitment. While grazing did not directly enhance NWSG recruitment, as seen by other studies (Doll et al., 2011; Launchbaugh, 1976; Lawrence et al., 1995), future trials should focus on more timely grazing to determine if that is indeed more beneficial. Other directions for study could be further reducing the seeding rates of the nurse crops used in this study or looking for other possible nurse crops with minimal regrowth potential.

5 | CONCLUSIONS

This study was designed to explore a potential solution to a major deterrent to NWSG adoption: lack of first-year grazing. Therefore, options that mitigate the forage gap would make adoption of NWSGs into forage programs more practical. During this study, BTM and DM reduced NWSG plant densities compared to NNC-I but did not eliminate them. Grazing of both nurse crops reduced WR and LAI but did not further reduce NWSG plant density. Both factors suggest that a balance between nurse crop density and grazing intensity has the potential to provide suitable forage during the establishment year while suppressing weed cover and allowing the recruitment of a suitable stand of perennial NWSG forage. Further studies are required to better elucidate the best balance between these factors. The timing of grazing likely plays a critical role in the effectiveness of weed suppression, but more data are required to better understand its influence. This approach has shown some potential but requires critically timed planning and grazing to achieve the best outcome.

AUTHOR CONTRIBUTIONS

Keagan J. Swilling: Conceptualization; data curation; formal analysis; investigation; methodology; project administration; supervision; validation; visualization; writing—original draft. **Eric Bisangwa:** Data curation; investigation; project administration; supervision; writing—review and editing. **Chad Bitler:** Conceptualization; data curation; funding acquisition; project administration; resources; super-

vision; writing—review and editing. **Xiaojuan Zhu:** Formal analysis; methodology; validation; writing—review and editing. **Virginia Sykes:** Conceptualization; formal analysis; funding acquisition; methodology; validation; visualization; writing—review and editing. **Patrick Keyser:** Conceptualization; formal analysis; funding acquisition; methodology; project administration; resources; supervision; validation; writing—review and editing.


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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

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