

Improving Wildflower Longevity in Roadside Seeding Areas

Project M329

Final Report

Submitted to the Nebraska Department of Transportation on August 18th, 2017.

By

Walter Schacht and Jon Soper

Department of Agronomy and Horticulture

University of Nebraska-Lincoln

In Collaboration with

Carol Wienhold

Environmental Section

Nebraska Department of Transportation

Introduction

Re-vegetation efforts on bare roadsides of newly-constructed highways are primarily focused on the stabilization of soil to reduce rates of erosion. The Nebraska Department of Transportation (NDOT) seeds roadsides with a diverse mixture of grasses and wildflowers for site stabilization as well as to enhance the visual quality of roadsides. Although grasses dominate roadside plantings in terms of cover and density, wildflowers are largely responsible for the visual enhancement of recently-seeded roadsides. In addition to the visual component, wildflowers provide essential ecological functions on roadsides. Wildflowers improve water and nutrient cycling in the compacted roadside soils by increasing water infiltration and nutrient availability. Leguminous wildflower species increase nitrogen content of soil. The variability of wildflower leaf size, shape and orientation provides a more continuous soil cover than grass alone. The diversity of wildflower growth habits and life cycles also allows for a greater range of stand establishment and persistence when compared to sites seeded to grasses alone.

Federal guidance states that wildflowers are to be used in roadside seeding mixtures. Wildflowers compose about 10% of the total seeds in the seeding mixtures used by NDOR. Wildflower seeds are relatively expensive; therefore, despite the low percentage of wildflowers in the seeding mixtures, they represent 30% of the total cost of seed. Staff of NDOT report that wildflower density of most roadside seeding projects is acceptable in the first 2 or 3 years after seeding, but that wildflower density and cover decline in the following years. A recent study completed by the University of Nebraska-Lincoln in collaboration with NDOT (Soper et al. 2010) has shown that wildflowers compose less than 10% of the botanical composition of highway roadsides 10 years following seeding. The study showed that the mature seeded stands along Nebraska highways are clearly dominated by seeded native grasses and invasive grasses, with only a few species of seeded wildflowers appearing occasionally.

The common native grasses (e.g., *Panicum virgatum*) used in the NDOT seeding mixtures germinate and establish more readily than most native, perennial wildflowers included in the seeding mixtures (Masters et al. 2001). These grasses have higher germination rates, are well-adapted to a diversity of soil conditions, and grow rapidly following germination; therefore, they are able to out-compete most wildflowers when they are seeded in the same area. In some cases, however, wildflower presence in recently-seeded, mixed stands can be enhanced by increasing the density or availability of wildflower seeds and/or spatially segregating the grass and wildflower seeds at seeding. The density or availability of seeds can be increased by using higher seeding rates or by establishing other sources of propagules or seeds, such as wildflower islands. Wildflower islands refer to small blocks within the area to be established that are seeded to wildflowers only. These localized areas seeded to wildflowers have the potential to remain as a dense stand of wildflowers and provide a source of seeds and propagules for wildflowers in the overall stand (Foster et al. 2004). When seeding new sites, the separation of grass and wildflower seed into separate rows has been shown to improve wildflower establishment by reducing the competition for resources between seedlings (Dickson and Busby 2009).

Research in grassland ecology has shown that wildflower diversity declines over time because the grasses are more competitive when natural disturbances are removed from the ecosystem (Collins et al. 1998). In grassland systems, common natural disturbances are burning and the trampling and defoliation associated with grazing. The significance of disturbance is made evident by the relatively high plant diversity of grasslands periodically exposed to disturbances. Disturbances (e.g., grazing and/or burning) tend to favor wildflowers because they suppress grasses, thus opening areas and providing

wildflowers greater access to water, nutrients, and light interception. Burning or grazing roadsides in Nebraska is not realistic; however, mowing can be used as a reasonable substitute for grazing. The trampling effect and the patchiness of defoliation associated with grazing are difficult to replicate with mowing but the timing and intensity of defoliation can be manipulated with mowing. Research has shown that mowing or grazing at specific times and heights during the year can greatly impact plant communities by reducing the competition between grasses and wildflowers (Collins et al. 1998, Williams et al. 2007).

Objectives

The purpose of our research was to test different strategies for establishing and maintaining wildflowers in roadside stands. The objective of the first study was to determine the interacting effects of (1) wildflower seeding rates and (2) spatial segregation of wildflower seeds on establishment of wildflowers on backslopes of roadsides (Experiment 1). A second study was conducted to determine the effect of seeding backslopes with the Type A mixture, with and without subplots of high-density seeding of wildflowers (wildflower islands; Experiment 2). The objective of a third study was to determine the interacting effects of (1) interseeding wildflowers into established stands on roadside backslopes and (2) the timing of mowing of the stands as the drilled wildflowers are establishing (Experiment 3).

Materials and Methods

Study Sites

Two locations were selected for the installation of research plots in 2013. The first location was near Kearney, NE, directly north of I-80 exit 275, along the Kearney by-pass (Nebraska State Highway 10). This site was previously a crop field, but the site was used for equipment storage during construction of the by-pass. During construction, the site was allowed to grow up with weedy annual grasses and broadleaf plant species. The site was seeded in spring 2014 and, because of a high density of weeds (Figure 1), was mowed in August 2014 and 2015; however, the production of seeds of the weedy species occurred because of the lateness of mowing. The primary soil types are sandy loam and silt loam. This site is level, with very little topographic features to influence water movement across the site. Average annual precipitation for Kearney, NE is 25.2 inches per year (Figure 2). During the study, annual precipitation was near average although 2016 was below the long-term average.

The second location included two sites along US Highway 81, 2 miles north of Bruning, NE. The two sites were seeded most recently in 2002 resulting in a fully vegetated stand of warm- and cool-season grasses, with some wildflowers present as well. The primary soil types are silt loam and silt clay loam; the sites are level. Geneva, NE is the nearest weather station to the project site and average annual precipitation is 29.3 inches per year (Figure 3). During the study, annual precipitation was near average although 2014 was above the long-term average.

Wildflower Establishment (Kearney Site)

All combinations of three wildflower seeding rates and two levels of spatial segregation of wildflower seeds in Experiment 1 were arranged in a completely randomized design with six replications. The seeding rates of wildflowers were the standard NDOT rate (4.1 lbs PLS/acre), a double rate (8.2 lbs PLS/acre), or a triple rate (12.3 lbs PLS/acre) (Table 1). The amount of grass seed applied (23.5 lbs PLS/acre) was consistent for all rates of wildflowers. Oats were seeded as a cover crop at 12 lbs

PLS/acre. The two levels of spatial segregation of wildflower seeds were (1) the conventional seeding approach of uniformly mixing and seeding wildflower and grass seeds and (2) segregating the wildflower seeds from the grass seeds and planting every fourth row to wildflowers only with the intervening three rows seeded to grass only. Prior to seeding, the entire plot area was disked. A no-till drill was used to seed all seeding rate levels of the conventional seeding approach, with the drill being cleaned between different treatments. The drill row spacing was 7 inches. For the segregated approach, grass seed was planted by the no-till drill and the wildflowers were planted by hand. Using the available 15 seed tubes (rows) on the drill, grass seed was planted in a sequence of three rows with every fourth seed tube plugged so that no seed was placed in the fourth row. The blank grass rows were then hand seeded with wildflower seeds. This resulted in 3 rows of wildflowers and 12 rows of grass in each plot. Each plot was 20 X 20 feet in size. There was 30 feet of border areas between plots within a replication and 20 feet between replications. All border area was planted with a grass only mixture, at the same rate as the grass in the plots. After planting, all plots and border areas were covered with hay mulch (4000 lbs/acre) and crimped into the soil with a disk. Planting occurred in mid-May 2014.

A second set of plots at the Kearney site was arranged in a completely randomized design with eight replications (Experiment 2). Eight plots, each measuring 20 x 26 feet (520 square feet), were seeded with the standard NDOT Type A seeding mixture (23.5 lbs PLS/acre grass seed and 4.1 lbs PLS/acre wildflower seed) and the entire area of another eight plots were seeded with the standard Type A mixture along with two wildflower islands seeded (8.2 lbs PLS/acre) in each plot. The wildflower islands were 4 X 4.5 feet (18 square feet x 2 = 36 square feet) and covered 7% of each plot. The islands were seeded following the drilling of the whole plots. Pre-measured seed amounts were hand scattered and lightly raked into the soil. Prior to planting, the area was disked to prepare the seedbed. All plots were covered with hay mulch (4000 lbs/acre) that was crimped into the soil with a disk. Planting occurred in mid-May, 2014.

Plot areas were mowed in both July and September of 2014 and 2015. Mowing was done to reduce the population of annual grasses and forb species.

Wildflower Persistence (Bruning, NE Site)

All combinations of three wildflower interseeding rates and three levels of mowing were arranged in a randomized complete block design (Experiment 3). The seeding rate of wildflowers was the standard NDOT rate (4.1 lbs PLS/acre), a double rate (8.2 PLS/acre) and no seeding. The three levels of mowing were: mowing in early July, mowing in early August, and no mowing. Planting occurred in mid-May, 2014. The plots were mowed at the appropriate dates (by treatment) in 2014, 2015, and 2016. The plots were mowed to a 6 inch stubble height using a tractor and shredder, which is the standard mowing practice used by NDOT.

Vegetation Measurements

In the year of seeding (2014), seedbed preparation at the Kearney site appeared to have created conditions favorable to the growth of weedy forbs and grasses. Because of the high populations of weedy forbs and grasses, plots were not sampled in 2014.

Frequency of occurrence was sampled in Experiment 1 using a frequency-grid method (Vogel and Masters 2001). The frequency grid was a metal frame containing 25 squares (5 x 5) or cells and was

made from concrete reinforcing sheets that have 15 x 15 cm squares (Figure 4). Within each plot, the frequency grid was placed in four locations and the presence or absence of individual species was recorded for each cell. The four placements per plot were combined to give an overall frequency of occurrence for an individual species for each plot. Sampling occurred in late June/early July and mid-September of 2015 and 2016.

Frequency of occurrence was sampled in Experiment 2 using a nested-frequency frame method (Smith et al. 1986). We used the nested-frequency method instead of the frequency-grid method because of the change in seeding method. With the broadcast method, seed placement could not be expected to be regularly spaced with seeds in each cell of a frequency grid. The nested quadrat had 4 sub-quadrats and the sizes were 100 cm², 625 cm², 1225 cm², and 2,500 cm². Ten nested quadrats were randomly located in each plot. At each quadrat placement, the smallest sub-quadrat in which each of the species was first encountered was recorded. If a species was recorded in the smallest sub-quadrat, it was given a score of 10, 6 for the next largest, 3 for the next largest, and 1 if it were found only in the largest sub-quadrat. Frequency scores for each treatment were calculated for each seeded species and selected categories (e.g., all seeded perennial species and native seeded species). Frequency scores provided an index of stand establishment. Well-established seeded grasslands in the Great Plains generally have a plant density of about 20 plants/m² (about 2 plants/ft²) (Vogel and Masters 2001). Based on the area of the nested quadrats used in this study, a frequency score of 6.0 would be equivalent to about 16 plants/m² (about 1.5 plants/ft²). The broadcast seeding of the wildflower islands is not compatible with the frequency grid method, which is a better technique for row plantings. The nested-frequency frame is better at accounting for the variability of a broadcast seeding. Sampling occurred in late June/early July and mid-September of 2015 and 2016.

Frequency of occurrence was sampled in August 2014, 2015, and 2016 in Experiment 3 using the frequency rod method (Sampson and Moser 1982). The frequency rod is a 1-m long rod, which is subdivided into 20, 5-cm increments. The frequency rod is placed alongside a seeded row and the presence of the seeded species is recorded for each of the 20 increments. For each plot, seven placements of the frequency rod was used to calculate the frequency of occurrence for each seeded species. As these plots were previously vegetated, the high canopy of the grass would have made the frequency grid hard to place at sampling locations. The frequency rod fits much better alongside the seeded rows and still gives accurate estimates of frequency of occurrence.

Data Analyses

Data were analyzed using the General Linear Model in SAS (SAS Institute Inc., Cary, NC). In Experiment 1, year (2), date (2), seeding rate (3), and spatial segregation (2) were included in the model. To further evaluate the effect of seeding method (segregation of wildflower seeds) on wildflower establishment in Experiment 1, we conducted an additional analysis on comparing frequency of occurrence of wildflowers in segregated rows vs their frequency of occurrence in the rows of the conventionally-seeded plots. To perform this analysis, we calculated the frequency of occurrence of each wildflower species in the 20 cells sampled over the four grids per segregated plot that were seeded with only wildflowers and the frequency of occurrence of wildflower species in all squares of the conventionally seeded plots. In Experiment 2, year (2), date (2), and wildflower island treatment (2) were included in the model. Treatment differences were analyzed for each individual seed species, weedy forb species,

and weedy grass species. In Experiment 3, year (3), site (2), seeding rate (2), and mowing frequency (3) were included in the model. Treatment differences were analyzed for each individual seeded species.

Results

Experiment 1 (Kearney)

Wildflowers

Year and Date Effects. Wildflower species occurred infrequently (had low frequencies of occurrence, <1%) and generally did not differ between years (2015 and 2016) and dates (June and September). The frequency of occurrence of Maximilian sunflower was 2.6 times greater in 2016 compared to 2015 (Table 2). Although the frequency of occurrence of blackeyed Susan differed between years, it was an infrequently occurring plant. Frequency of weedy forbs declined over time with 72.4% in 2015 and 27.1% in 2016.

Method and Seeding Rate Effects. There was a method by seeding rate interaction for upright prairie coneflower and Indian blanket-flower (Tables 3 and 4). Frequency of occurrence of upright prairie coneflower in the 1X seeding rate was 1.6 times greater for the combined method than the segregated method but the opposite was true for the 2x and 3x seeding rates (Table 3). For Indian blanket-flower, frequency of occurrence for the 1x seeding rate by the combined seeding method was generally greater than the other two seeding rates and the segregated seeding method (Table 4).

Grasses

Year and Date Effects. The frequency of occurrence of both big bluestem and sideoats grama were 2 times greater in 2016 than in 2015 (Table 2). Although switchgrass was uncommon, its frequency of occurrence also increased in 2016. The frequency of occurrence differed between sampling dates in five seeded grass species (Table 5). Frequency of occurrence was greatest in June for Indiangrass, little bluestem, switchgrass, and western wheatgrass. Sideoats grama had a frequency of occurrence that was greatest in September.

There was a year x date interaction for Canada wildrye (Table 6). The frequency of occurrence for Canada wildrye in June 2016 was 2 times greater than June 2015. Additionally, Canada wildrye frequency in June 2016 was 4 times greater than in September 2016.

Seeding Rate and Seeding Method Effects. The impact of seeding method had limited effect on seeded grass species, except for big bluestem, which was greater in the combined method compared to the segregated method (Table 7). There was a seeding rate by seeding method interaction for two species. The frequency of occurrence of Canada wildrye in the 1X seeding rate was 2.6 times greater for the combined method than the segregated method (Table 8). The frequency of occurrence of Canada wildrye in the combined method also was 2 times greater in the 1X seeding rate than the 2X seeding rate. The frequency of occurrence for slender wheatgrass in the 1X seeding rate was nearly 2 times greater in the combined method than the segregated method (Table 9). Within the combined method, the frequency of occurrence of slender wheatgrass at the 1X seeding rate was about 2 times greater than in either the 2X or 3x seeding rate.

Seeding Rate and Seeding Method by Year Effects. None of the seeded species had a seeding rate by seeding method by year effect, but weedy grasses did. In 2015, frequency of occurrence for weedy

grasses did not differ between seeding rate and seeding method. In 2016, frequency of occurrence was different between the seeding rates and seeding methods (Table 10). Within the combined seeding method, the 2X rate was 2.6 and 1.5 times greater than the 1X and 3X seeding rates, respectively. The frequency of occurrence at the 2X rate was 1.6 times greater in the combined method than in the segregated method. The frequency of occurrence was 2.2 and 1.4 times greater in the segregated method compared to the combined method in the 1X and 3X seeding rates, respectively. Within the segregated method, the frequency of occurrence was greater in the 3X rate compared to the 2X seeding rate.

Wildflower Frequency of Occurrence Based on Segregated Rows

Method Effects. The frequency of occurrence of both Indian blanket-flower and Maximilian sunflower were greater in the segregated rows than in the combined rows (Table 11). In the segregated rows, the frequency of occurrence of Indian blanket-flower and Maximilian sunflower was about 2 times greater than in the combined rows.

Method by Seeding Rate Effect. The frequency of occurrence of upright prairie coneflower was 2 to 9 times greater in the segregated rows than in the combined rows (Table 12). Within the segregated rows, the 3x seeding rate of upright prairie coneflower had a significantly higher frequency of occurrence compared to the 1X seeding rate. Within the combined rows, the frequency of occurrence of upright prairie coneflower did not differ among the seeding rates.

Experiment 2 (Kearney)

Wildflowers

Year, Date and Treatment Effects. Maximilian sunflower was the only seeded species to have a year by date by treatment interaction (Table 13). The frequency score of Maximilian sunflower in the wildflower island plots in September 2016 (7.5) were 2.5 times greater than in the standard treatment plots in September 2016 (Table 13). The frequency score of Maximilian sunflower was also about 2.5 times greater in September (7) than in June (3) of 2016. Overall, Maximilian sunflower peaked in frequency in September 2016 in the wildflower island plots.

Weedy forbs also had a significant year by date by treatment interaction. In 2015, frequency scores of weedy forbs were high, generally 5.5 to 9.3 (Table 14). However, in 2016 weedy forb frequency scores declined to 0.8 to 1.2.

Three wildflower species had significant date by treatment interactions (Table 15). Upright prairie coneflower had frequency scores that were highest in June (2.8) for the standard seeding and in September (3.5) for the wildflower island plots. Frequency scores of blackeyed Susan and blue flax varied by month and treatment, but the frequency scores were generally low (<1.0) and not very meaningful in testing the response of these two species to the two seeding treatments.

Upright prairie coneflower had a significant year by treatment interaction. (Table 16). It had a higher frequency score in the standard seeding plots than the wildflower island plots in 2015 but the reverse was true in 2016. Similar to Maximilian sunflower, upright prairie coneflower peaked in frequency in September 2016 in the wildflower island plots.

Treatment Effects. Indian blanket-flower was the only forb where treatment as a main effect was significant. Frequency score of Indian blanket-flower was greater for wildflower island plots regardless of date and year (Table 17). Two forb species had significant treatment effects. Frequency scores of Indian blanket-flower and New England aster were both greater in the wildflower island method compared to standard method (Table 17).

Wildflowers Inside and Outside of Islands

We summarized the frequency scores of the wildflower species within the wildflower islands and the plot area outside of the wildflower islands so that we could quantify and compare the frequency of wildflowers inside and outside of the islands (Table 18). We found that the frequency scores of Indian blanket-flower, Maximilian sunflower, and upright prairie coneflower were consistently greater through the two years of data collection in the wildflower islands than in the plot areas outside the wildflower islands (Figure 5). The frequency scores of the other seeded wildflowers also tended to be greater in the wildflower islands but these species were relatively uncommon.

Grasses

Year and Date Effects. Three species, Canada wildrye, little bluestem, and slender wheatgrass had significant year by date interactions. The frequency scores of the three species peaked in either June or September 2016 (Table 19). Although frequency scores of all three species peaked in 2016, the frequency score of slender wheatgrass the only with a higher score in September 2016 than in June 2016.

Date was a significant main effect for sideoats grama and western wheatgrass. Regardless of seeding method and year, the frequency score of sideoats grama was greater in September compared to June; whereas, western wheatgrass had frequency scores greater in June than in September (Table 20). This is as expected because sideoats grama is a warm-season grass and should peak in frequency in September; whereas, western wheatgrass is a cool-season grass and should peak in frequency in June. Year was a significant main effect for big bluestem and sideoats grama. Regardless of seeding method and date, frequency scores of both species was almost 2 times greater in 2016 than 2015 (Table 21).

Treatment and Date Effects. Indiangrass had a date by treatment interaction. Frequency scores in 2015 did not differ between June and September; whereas, frequency score was greater in September than in June in 2016 (Table 22). Weedy grasses had a year by date by treatment interaction. There was no consistent response of weedy grasses to seeding method between dates and years (Table 23).

Experiment 3 (Bruning)

Site and Year Effects. Frequency of occurrence of Indian blanket-flower was greater on the North site than the South site in all years; however, its frequency declined three fold from 2014 to 2016 on the North site whereas it did not decline significantly on the South site (Table 24). Contrary to Indian blanket-flower, frequency of occurrence of purple prairieclover increased at both sites from 2014 to 2016 (Table 25). However, similar to Indian blanket-flower, the frequency of occurrence of purple prairieclover was greater at the North site than the South site. Regardless of mowing frequency or seeding rate, frequency of occurrence of upright prairie coneflower was greater at the North site (3.3%) than the South site (0.7%).

Mowing Frequency and Seeding Rate Effects. Frequency of mowing and seeding rate had no effect on frequency of occurrence of the seeded wildflower species with two exceptions. With a mowing frequency x seeding rate interaction, frequency of occurrence of blackeyed Susan was two to three times greater on the mowed plots than on the non-mowed plots at the 4 lbs PLS/acre seeding rate (Table 26). The frequency of occurrence of blackeyed Susan was generally greater at the 4 lbs PLS/acre seeding rate than at the heavier seeding rate. Frequency of occurrence of purple prairieclover also was greater at the 4.1 lbs PLS/acre seeding rate with a frequency of occurrence of 3.3% at the lower seeding rate and 0.9% at the heavier seeding rate.

Discussion

Experiment 1 (Kearney)

Wildflowers

Of the 10 wildflower species seeded, only Indian blanket-flower, Maximilian sunflower, and upright prairie coneflower were commonly occurring (>5% frequency of occurrence). Plants of blue flax, New England aster, shell-leaf penstemon, and blackeyed Susan were present in the plots but at low frequencies of occurrence (<1%). Of the three main wildflower species, the frequency of occurrence of only upright prairie coneflower was greater in the segregated plots than in the combined plots (conventionally seeded plots with the grasses and wildflowers seeded as a mixture). Overall, segregation of wildflower seeds in every fourth row of a plot did not result in increasing the frequency of wildflowers as compared to seeding the grass/wildflower mixture in all rows of a plot. However, the frequencies of occurrence of Indian blanket-flower and Maximilian sunflower were two times greater in segregated rows than in rows seeded to the grass/wildflower mixture, suggesting that wildflowers establish at a greater rate when seeded in wildflower only rows and in the absence of competition with grasses. We used a ratio of grass segregated rows to wildflower segregated rows of 3:1. Our results at the plot level likely would have been different if we would have tested a ratio of 3:2 or 2:2 because there would have been more wildflower segregated rows per plot resulting in more wildflowers. The amount of grass seed applied (23.5 lbs PLS/acre) was consistent for all rates of wildflowers

In our study with a ratio of three segregated grass rows to one segregated wildflower row, we took the wildflower seeds that would have been in the three grass segregated rows and placed them in the wildflower segregated row. Decreasing the ratio of grass rows to wildflower rows would decrease the wildflower seeds in the wildflower rows. This would have the potential of decreasing the number of wildflowers per row. As expected, increasing the seeding rate of wildflowers from 4.1 lb PLS/acre (standard NDOR seeding rate) to 8.2 lbs PLS/acre and 12.3 lbs PLS/acre resulted in increased frequencies of the main wildflower species. With ratios of grass segregated rows to wildflower segregated rows closer to 1:1 and higher seeding rates of wildflowers, we would expect greater establishment of wildflowers on backslopes of roadsides.

Grasses

All of the seeded grasses were found in the plots of the study site. Slender wheatgrass, big bluestem, Indiangrass, sideoats grama, and Canada wildrye were the most common (>5%). The segregation of grass and wildflower seeds in separate rows affected frequency of occurrence of seeded grasses at the plot level in only a few instances. We expected lower frequency of occurrence of seeded grasses in plots

with segregated rows because some of the cells of the frequency grid would be on the wildflower segregated rows and no grasses would be recorded for these cells. However, only big bluestem had greater frequency of occurrence in the combined plots than in the segregated plots, although Canada wildrye and slender wheatgrass had greater frequencies in the combined plots in a few interactions of seeding method and seeding rate. With a 3:1 ratio of grass segregated rows to wildflower segregated rows, frequency of occurrence of seeded grasses was largely unaffected. We expect that frequency of occurrence of seeded grasses would decrease appreciably as the ratio of grass segregated rows to wildflower segregated rows approaches 3:2 or 1:1.

Weedy Forbs and Grasses

The study site had a dense cover of weedy forbs and grasses in 2013, the year before the plots were established and seeded. We applied pre-emergent herbicide in spring 2014 but the weedy forbs and grasses still dominated the study site with nearly a 100% canopy cover during the initial growing season of 2014. Even in the second growing season (2015) following mowing of the plots in both 2014 and 2015, the frequency of occurrence of weedy forbs was 72.4%. However, frequency of weedy forbs declined to 27.1% in 2016. Some of the seeded wildflower species, such as Maximilian sunflower, responded positively to the decline in weedy forbs and gradually increased in frequency of occurrence over the study period. We also found an increase in a number of the seeded grasses, including big bluestem, sideoats grama, and switchgrass, over the study period. The establishment of seeded stands of perennial grasses and wildflowers commonly requires three years but the heavy weed pressure on our study site likely slowed the rate of establishment even more and perhaps reduced the wildflower response to the segregated seeding.

Experiment 2 (Kearney)

Incorporating wildflower islands into conventionally-seeded (standard) plots generally resulted in increased frequency scores of Maximilian sunflower, upright prairie coneflower, and Indian blanket-flower at the whole plot level compared to the standard plots. Even though the wildflower islands composed only 7% of an entire plot area, the addition of the broadcast wildflower seeds increased the presence of the three species at the whole plot level. Increasing the size of the wildflower islands likely would result in significant increases in the presence of the other seeded wildflower species. In the longer-term, the frequency (or density) of wildflowers should remain relatively high in the islands because of less competition with grasses. If this is the case, the wildflowers within the islands could serve as a propagule source (e.g., seeds and rhizomes) to facilitate expansion of wildflowers throughout the roadside.

Seeded grass species did not show any negative impacts from the seeding of wildflower islands within the larger planting area. If the size of the wildflower islands were to increase, it seems unlikely that the grass population would be impacted. Grasses usually out-compete wildflowers as the seeding matures, so creating larger wildflower islands should favor establishment of higher wildflower densities in the overall seeded area.

Experiment 3 (Bruning)

Our attempt to establish wildflowers in grass-dominated backslopes of roadsides by interseeding and mowing was successful for only a few wildflower species. The grass stands at the two sites were

dominated by smooth brome grass and Kentucky bluegrass. Establishing wildflowers into mature grass stands is challenging because of the difficulty of getting good wildflower seed-to-soil contact when seeding and the poor growing conditions (considerable competition for light and soil nutrients and water) for a wildflower seedling in a mature grass stand. Our approach to address these two challenges was to use methods that NDOT likely could employ without significant changes in roadside management. A no-till drill was used to insert the wildflower seeds into the soil for ensuring the likelihood of good seed-to-soil contact. Herbicides, grazing, or fire are commonly used as methods of suppressing the grasses in an established stand and of reducing competition for the interseeded species. We used mowing as a tool that could be readily implemented by NDOT or their contractors.

Blackeyed Susan, Indian blanket-flower, and purple prairieclover were the only species with frequencies of occurrence greater than 5% (but they were less than 10%). Frequencies of 5 to 10% are equivalent to 1 or 2 plants for every meter (39.4 inches) of row, which should be considered acceptable seeding success. The other seeded species had frequencies of occurrence less than 1% except for upright prairie coneflower which had a frequency of about 2.5% on the two sites.

The low presence of most of the wildflower species indicates poor seed-to-soil contact and/or low soil moisture following interseeding. More aggressive interseeding methods are likely needed to improve seeding conditions. Other practices may need to be implemented to create better conditions for seed germination and seedling establishment, such as 1. light disking to penetrate the sod and expose soil in which the seeds can be drilled, 2. some other form of soil surface disturbance such as mowing followed by severe raking to remove the plant residue and disturb the soil surface, and 3. application of foliar-active herbicides (e.g., glyphosate or paraquat) at the time of seeding to effectively suppress the sod species.

Mowing favored the establishment of blackeyed Susan and upright prairie coneflower but appeared to have no effect on the other wildflower species. Mowing as a means of opening the canopy of a roadside stand should have a positive influence. There was not any evidence that a second mowing during the growing season would have a positive effect on wildflower establishment. It is possible that our second mowing date (August) was too late in the growing season to affect wildflower growth and establishment. The lack of response to an increased seeding rate cannot be explained.

Summary

Concerning the segregation and increased seeding rate of wildflowers in Experiment 1, we conclude the following.

- Upright prairie coneflower had the greatest increase in frequency of occurrence in the plots with segregated rows compared to the combined plots that were conventionally seeded.
- Maximilian sunflower and Indian blanket-flower also increased in frequency of occurrence in the plots with segregated rows compared to the combined plots.
- The frequency of occurrence of wildflowers was greater in the segregated wildflower rows than in the combined rows.
- Our results suggest that a 3:1 ratio of grass segregated rows to wildflower segregated rows is not optimum for establishing wildflowers in roadsides. Lower ratios of grass segregated rows to wildflower segregated rows (e.g., 3:2 or 1:1) should be considered. The more even ratios could also further reduce inter-row competition between grasses and wildflowers.

- The frequency of occurrence of wildflowers was not affected by seeding rate.
- The frequency of occurrence of grasses was not reduced by either segregation or by increased seeding rates of wildflowers.
- Weedy forbs and grasses heavily infested the study location in 2014 (year of seeding) and 2015, but declined in 2016. Weed control efforts and continued development of the seeded species likely were the reasons for the decline of the weedy species over the three years of study.

Concerning the impact of establishing wildflower islands within a larger seeded area, we conclude the following.

- Maximilian sunflower, upright prairie coneflower, and Indian blanket-flower were more common in wildflower islands than in the surrounding plot area that was seeded to the Type A mixture.
- Even with the wildflower islands composing only 7% of the entire plot area, plots with wildflower islands had greater frequency of Maximilian sunflower, upright prairie coneflower, and Indian blanket-flower than did plots without wildflower islands.

Concerning the impact of increased seeding rate and mowing frequency on the establishment of interseeded wildflowers, we conclude the following.

- The frequency of occurrence of blackeyed Susan, Indian blanket-flower, and purple prairieclover responded favorably to interseeding.
- Mowing favored the establishment of blackeyed Susan and upright prairie coneflower but appeared to have no effect on the other wildflower species.
- Overall, interseeding of wildflowers into established, mature stands of perennial grasses on highway roadsides was marginally successful. Additional methods of suppressing the grass sod should be considered for improving interseeding success.

Management Implications

- The segregation of wildflowers into separate rows can be a useful method to increase wildflower densities on recently-seeded roadsides; however, only three of the ten seeded wildflower species had adequate establishment to fully evaluate the effect of row segregation of wildflower seeds on establishment. Upright prairie coneflower, Maximilian sunflower, and Indian blanket-flower were the three species that responded favorably to row segregation. In our study, the very high weed density in 2014 and 2015 likely affected the establishment of the other seeded wildflower species, e.g., blue flax, shell-leaf penstemon, and New England aster. These species tended to respond favorably to row segregation but their frequencies were so low (<1%) that we decided against reporting their results. Regardless, we measured a favorable response of wildflower establishment to row segregation at the row level and plot level. However, the plot level response was minimal even for the three more common wildflower species. Therefore, we suggest a lower grass row to wildflower row ratio (e.g., 4:2 or 3:2) if row segregation is to be considered a strategy for increasing wildflower frequency on backslopes of roadsides. Although we like the approach of row segregation to increase the presence of wildflowers on backslopes, seeding roadsides with lower grass:wildflower row ratios increases the proportion of the area that is seeded to wildflowers only. Wildflower seedings are relatively slow to establish and have a higher risk of establishment, creating challenges in minimizing the

time required to close highway construction projects' environmental permits. The cost of seeding also would increase as the proportion of wildflower seeds in the total seeding mixture increases. Another important note is that commercial seed drills would need to be modified to seed grasses and wildflowers in separate rows. Overall, the strategy of using segregated rows to improve the success of wildflower establishment appears to have much potential and should be further tested.

- The addition of wildflower islands within a larger seeding area has the potential to increase wildflower density/frequency in new roadside plantings. Our wildflower islands composed only 7% of the new seeding area, which appears to be the minimum size required to have an impact on wildflower frequency. Again only three species, Maximilian sunflower, Indian blanket-flower, and upright prairie coneflower, responded favorably to the wildflower island approach; as in Experiment 1, the weed infestation in 2014 and 2015 likely limited the establishment of the other seven species that were seeded. Establishment of wildflower islands appears to be another approach of increasing wildflower density/frequency on roadsides. The patchy appearance of wildflower islands certainly is different than the more linear, continuous appearance of segregated rows. The wildflower islands might be considered better adapted at roadway interchanges, near towns, or for creation of pollinator habitat. We were not able to assess the longer-term influence of wildflower islands on plant composition of roadsides (i.e., the whole plot). We expect that the frequency/density of wildflowers should remain relatively high in the islands because of less competition with grasses. And, if this is the case, the wildflowers within the islands could serve as a propagule source (e.g., seeds and rhizomes) to facilitate expansion of wildflowers throughout the roadside. Finally, we used a labor intensive method of broadcast seeding and hand-raking to incorporate wildflower seeds into the ground. Mechanized methods of patch seeding will need to be explored in seeding patches if wildflower islands are to be considered a roadside strategy.
- Further testing is needed of interseeding wildflowers into established stands dominated by perennial grasses on roadsides. Wildflower response to interseeding was favorable on the North site and provided the basis for identifying the factors that need further research. Providing for good seed-to-soil contact is essential and other methods of improving seeding conditions should be tested, such as raking or disking prior to drilling. Suppression of the grasses following interseeding is also essential and mowing appears to have limited impact on wildflower establishment; herbicides or more severe mowing could be tested. The longevity of the established wildflowers resulting from interseeding also needs to be documented. We expect a short life expectancy for the established wildflowers (<5 years) unless aggressive follow-up management practices are implemented that suppress the grasses.

Literature Cited

- Collins, S.L., A.K. Knapp, J.M. Briggs, J.M. Blair and E.M. Steinauer. 1998. Modulation of Diversity by Grazing and Mowing in Native Tallgrass Prairie. *Science* **280**:745-747
- Dickson, T.L. and W.H. Busby. 2009. Forb Species Establishment Increases with Decreased Grass Seeding Density in Northeast Kansas, U.S.A., Experimental Prairie Restoration. *Restoration Ecology*. **17**:597-605
- Foster, B.L., T.L. Dickson, C.A. Murphy, I.S. Karel and V.H. Smith. 2004. Propagule Pools Mediate Community Assembly and Diversity-Ecosystem Regulation Along a Grassland Productivity Gradient. *Journal of Ecology*. **92**:435-449
- Masters, R.A., D.D. Beran, and R.E. Gaussion. 2001. Restoring Tallgrass Prairie Species Mixtures on Leafy Spurge-infested Rangeland. *Journal of Range Management*. **54**: 362-369
- Sampson, J.F. and L.E. Moser. 1982 Sod-seeding perennial grasses into eastern Nebraska pastures. *Agronomy Journal* **74**:1055-1060.
- SAS Institute Inc. 2013. SAS/ACCESS 9.4. Cary, NC: SAS Institute Inc.
- Smith, B.H., M.L. Ronsheim, and K.R. Swartz. 1986. Reproductive ecology of *Jeffersonia diphylla* (Berberidaceae). *American Journal of Botany* **73**:1416-1426.
- Soper, J.M., W.H. Schacht and C.E. Wienhold. 2010. Adapting Roadside Seeding Mixtures to Locate Site Conditions in Nebraska. Annual Meeting of the Society for Range Management. Denver, CO.
- Vogel, K.P. and Masters, R.A. 2001. Frequency Grid: A Simple Tool for Measuring Grassland Establishment. *Journal of Range Management*. **54**: 653-655
- Williams, D.W., L.L. Jackson and D.D. Smith. 2007. Effects of Frequent Mowing on Survival and Persistence of Forbs and Seeded into a Species-Poor Grassland. *Restoration Ecology*. **15**:24-33

List of Figures

Figure 1. Growth of weedy forbs and grasses in September 2014 at Experiment 1 (Kearney, NE).

Figure 2. Cumulative precipitation at Kearney, NE.

Figure 3. Cumulative precipitation at Geneva, NE

Figure 4. Frequency grid used for estimating frequency of occurrence in Experiment 1 (Kearney, NE).

Figure 5. Wildflower island plot in Experiment 2 (Kearney, NE).



Figure 1. Growth of weedy forbs and grasses in September 2014 at Experiment 1 (Kearney, NE).

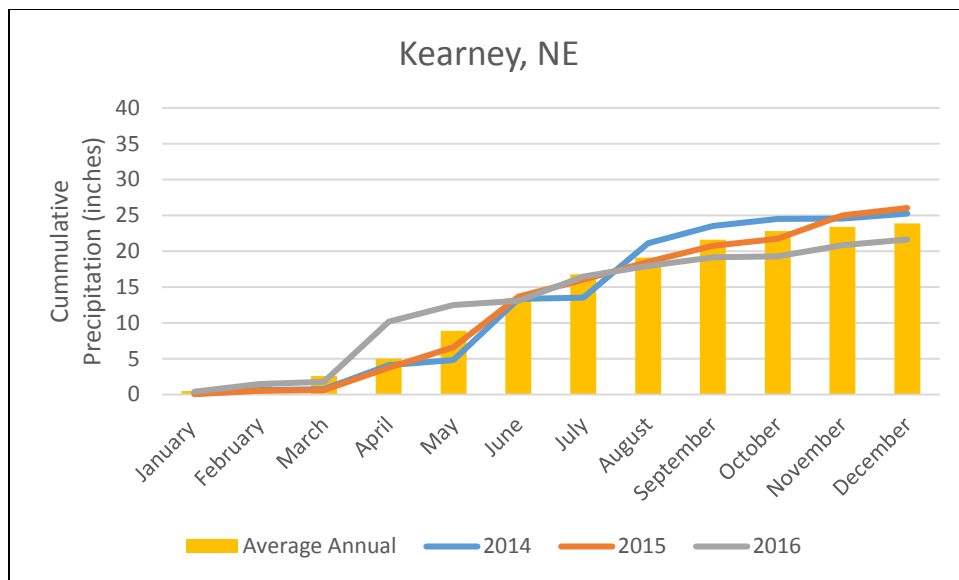


Figure 2. Cumulative precipitation at Kearney, NE.

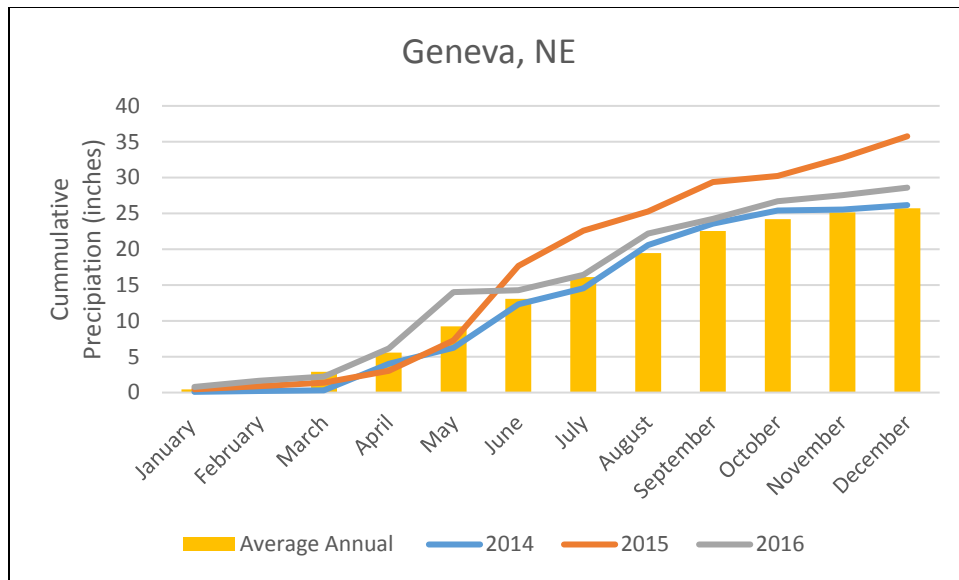


Figure 3. Cumulative precipitation for Geneva, NE.



Figure 4. Frequency grid used for estimating frequency of occurrence in Experiment 1 (Kearney, NE).



Figure 5. Wildflower island plot in Experiment 2 (Kearney, NE).

List of Tables

Table 1. Seeding mixture and seeding rate for Experiment 1 and 2 (Kearney, NE).

Table 2. Frequency of occurrence (%) for Experiment 1 in 2015 and 2016 (Kearney, NE).

Table 3. Frequency of occurrence (%) for upright prairie coneflower in response to combinations of seeding rate and seeding method for Experiment 1 (Kearney, NE).

Table 4. Frequency of occurrence (%) for Indian blanket-flower in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

Table 5. Frequency of occurrence (%) of several grasses in June and September for Experiment 1 (Kearney, NE).

Table 6. Frequency of occurrence (%) of Canada wildrye in response to date and year of sampling for Experiment 1 (Kearney, NE).

Table 7. Frequency of occurrence (%) of big bluestem, New England aster, and weedy forbs by seeding method for Experiment 1 (Kearney, NE).

Table 8. Frequency of occurrence (%) of Canada wildrye in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

Table 9. Frequency of occurrence (%) for slender wheatgrass in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

Table 10. Frequency of occurrence (%) of weedy grasses in response to seeding rate and seeding method in 2016 for Experiment 1 (Kearney, NE).

Table 11. Frequency of occurrence (%) of Indian blanket-flower and Maximillian sunflower in response to seeding method for Experiment 1 (Kearney, NE).

Table 12. Frequency of occurrence (%) of upright prairie coneflower in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

Table 13. Frequency scores of Maximillian sunflower for Experiment 2 (Kearney, NE).

Table 14. Frequency scores of weedy forbs for Experiment 2 (Kearney, NE).

Table 15. Frequency scores by seeding method and date for Experiment 2 (Kearney, NE).

Table 16. Frequency scores for upright prairie coneflower for Experiment 2 (Kearney, NE).

Table 17. Frequency scores based on seeding method for Experiment 2 (Kearney, NE).

Table 18. Average frequency scores of wildflower islands compared to the non-island area of the plot for Experiment 2 (Kearney, NE).

Table 19. Frequency scores of grasses based on year and date for Experiment 2 (Kearney, NE).

Table 20. Frequency scores based on date for Experiment 2 (Kearney, NE).

Table 21. Frequency scores based on year for Experiment 2 (Kearney, NE).

Table 22. Frequency scores of Indiangrass for Experiment 2 (Kearney, NE).

Table 23. Frequency scores of weedy grasses for Experiment 2 (Kearney, NE).

Table 24. Frequency of occurrence of Indian blanket-flower on the North and South sites from 2014 to 2016 for Experiment 3 (Bruning, NE).

Table 25. Frequency of occurrence for purple prairieclover on the North and South sites from 2014 to 2016 for Experiment 3 (Bruning, NE).

Table 26. Frequency of occurrence of blackeyed Susan for Experiment 3 (Bruning, NE).

Table 1. Seeding mixture and seeding rate for Experiment 1 and 2 (Kearney, NE).

Species	Seeding rate		
	1x	2X	3X
	—lbs. of pure live seed/acre—		
Wildflowers			
Blackeyed Susan (<i>Rudbeckia hirta</i> L.)	0.3	0.6	0.9
Blue flax (<i>Linum perenne</i> L.)	1	2	3
Indian blanket-flower (<i>Gaillardia pulchella</i> Foug.)	1	2	3
Maximilian sunflower (<i>Helianthus maximiliani</i> Schrad.)	0.25	0.5	0.75
New England aster (<i>Aster novae-angliae</i> L.)	0.2	0.4	0.6
Purple prairie clover (<i>Dalea purpurea</i> Venten.)	0.2	0.4	0.6
Rocky Mountain bee plant (<i>Cleome serrulata</i> Pursh)	0.4	0.8	1.2
Roundhead lespedeza (<i>Lespedeza capitata</i> Michx.)	0.1	0.2	0.3
Shell-leaf penstemon (<i>Penstemon grandiflorus</i> Nutt.)	0.15	0.3	0.45
Upright prairie coneflower (<i>Ratibida columnifera</i> ((Nutt.)) Wooton & Standl.)	0.5	1	1.5
Grasses			
Big bluestem (<i>Andropogon gerardii</i> Vitman)	3	3	3
Canada wildrye (<i>Elymus canadensis</i> L.)	4	4	4
Indiangrass (<i>Sorghastrum nutans</i> ((L.)) Nash)	3	3	3
Little bluestem (<i>Schizachyrium scoparium</i> ((Michx.)) Nash)	2.5	2.5	2.5
Sideoats grama (<i>Bouteloua curtipendula</i> ((Michx.)) Torr.)	3	3	3
Slender wheatgrass (<i>Elymus trachycaulus</i> (Link) Shinnery)	3	3	3
Switchgrass (<i>Panicum virgatum</i> L.)	1	1	1
Western wheatgrass (<i>Elymus smithii</i> ((Rydb.)) Gould)	4	4	4
Oats (<i>Avena fatua</i> L.)	12	12	12

Table 2. Frequency of occurrence (%) for Experiment 1 in 2015 and 2016 (Kearney, NE).

Species	<i>Year</i>	
	2015	2016
	%	
Big bluestem	12.4 ^b	25.4 ^a
Blackeyed Susan	1.2 ^a	0.2 ^b
Maximilian sunflower	4.0 ^b	10.4 ^a
Sideoats grama	3.8 ^b	9.8 ^a
Switchgrass	0.2 ^b	0.5 ^a
Weedy forbs	72.4 ^a	27.1 ^b

^{a-b} Different letters within a row indicate a significant difference ($p < 0.05$).

Table 3. Frequency of occurrence (%) for upright prairie coneflower in response to combinations of seeding rate and seeding method for Experiment 1 (Kearney, NE).

<i>Seeding rate</i>	<i>Seeding method</i>	
	Combined	Segregated
	%	
1X	5.4 ^{Aa}	3.4 ^{Ab}
2X	2.0 ^{Bb}	6.1 ^{Aab}
3X	2.8 ^{Bab}	8.0 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns (p<0.05).

Table 4. Frequency of occurrence (%) for Indian blanket-flower in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

<i>Seeding rate</i>	<i>Seeding method</i>	
	Combined	Segregated
	%	
1X	9.9 ^{Aa}	5.2 ^{Ba}
2X	4.9 ^{Ab}	6.9 ^{Aa}
3X	4.2 ^{Ab}	7.2 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns (p<0.05).

Table 5. Frequency of occurrence (%) of several grasses in June and September for Experiment 1 (Kearney, NE).

<i>Species</i>	<i>Sampling date</i>	
	June	September
	—————%—————	
Indiangrass	8.7 ^a	6.0 ^b
Little bluestem	2.5 ^a	0.9 ^b
Sideoats grama	5.0 ^b	8.4 ^a
Switchgrass	0.5 ^a	0.2 ^b
Western wheatgrass	9.7 ^a	0.9 ^b

^{a-b} Different letters within a row indicate a significant difference ($p < 0.05$).

Table 6. Frequency of occurrence (%) of Canada wildrye in response to date and year of sampling for Experiment 1 (Kearney, NE).

<i>Sample Date</i>	<i>Year</i>	
	2015	2016
	—————%—————	
June	6.3 ^{Ba}	14.1 ^{Aa}
September	5.6 ^{Aa}	3.7 ^{Ab}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns ($p < 0.05$).

Table 7. Frequency of occurrence (%) of big bluestem, New England aster, and weedy forbs by seeding method for Experiment 1 (Kearney, NE).

<i>Species</i>	<i>Seeding method</i>	
	Combined	Segregated
	—————%—————	
Big bluestem	21.8 ^a	16.1 ^b
New England aster	0 ^b	0.1 ^a
Weedy forbs	45.6 ^b	54.2 ^a

^{a-b} Different letters within a row indicate a significant difference ($p < 0.05$).

Table 8. Frequency of occurrence (%) of Canada wildrye in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

<i>Seeding rate</i>	<i>Seeding method</i>	
	Combined	Segregated
	%	
1X	12.6 ^{Aa}	4.8 ^{Ba}
2X	6.3 ^{Ab}	6.6 ^{Aa}
3X	9.3 ^{Aab}	4.1 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns (p<0.05).

Table 9. Frequency of occurrence (%) for slender wheatgrass in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

<i>Seeding rate</i>	<i>Seeding method</i>	
	Combined	Segregated
	%	
1X	30.0 ^{Aa}	17.9 ^{Ba}
2X	15.2 ^{Ab}	18.3 ^{Aa}
3X	19.6 ^{Ab}	17.2 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns (p<0.05).

Table 10. Frequency of occurrence (%) of weedy grasses in response to seeding rate and seeding method in 2016 for Experiment 1 (Kearney, NE).

<i>Seeding rate</i>	<i>Seeding method</i>	
	Combined	Segregated
	—————%—————	
1X	28.0 ^{Bb}	61.6 ^{Aab}
2X	73.6 ^{Aa}	46.5 ^{Bb}
3X	47.8 ^{Bb}	65.6 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letter indicate differences within columns (p<0.05).

Table 11. Frequency of occurrence (%) of Indian blanket-flower and Maximilian sunflower in response to seeding method for Experiment 1 (Kearney, NE).

Species	Seeding method	
	Combined	Segregated
	%	
Indian blanket-flower	6.4 ^b	16.4 ^a
Maximilian sunflower	7.2 ^b	12.4 ^a

^{a-b} Different letters within a row indicate a significant difference ($p < 0.05$).

Table 12. Frequency of occurrence (%) of upright prairie coneflower in response to seeding rate and seeding method for Experiment 1 (Kearney, NE).

<i>Seeding rate</i>	Seeding method	
	Combined	Segregated
	—————%—————	
1X	5.4 ^{Aa}	11.3 ^{Ab}
2X	2.0 ^{Ba}	19.2 ^{Aab}
3X	2.8 ^{Ba}	21.1 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns (p<0.05).

Table 13. Frequency scores of Maximilian sunflower for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
2015		
June	1.3 ^{Aa}	2.5 ^{Aa}
September	2.6 ^{Aa}	2.5 ^{Aa}
2016		
June	4.1 ^{Aa}	3 ^{Ab}
September	2.9 ^{Ba}	7.5 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns at the p=0.05.

Table 14. Frequency scores of weedy forbs for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
2015		
June	5.5 ^{Bb}	9.3 ^{Aa}
September	8.6 ^{Aa}	8.8 ^{Aa}
2016		
June	1.2 ^{Aa}	0.6 ^{Aa}
September	0.1 ^{Aa}	0.8 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns at the p=0.05.

Table 15. Frequency scores by seeding method and date for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
Blackeyed Susan		
June	0.3 ^{Aa}	0.2 ^{Ab}
September	0.1 ^{Ba}	0.9 ^{Aa}
Blue flax		
June	0.7 ^{Aa}	0.3 ^{Ba}
September	0 ^{Bb}	0.4 ^{Aa}
Upright prairie coneflower		
June	2.8 ^{Aa}	1 ^{Bb}
September	0.7 ^{Bb}	3.5 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns (p<0.05).

Table 16. Frequency scores for upright prairie coneflower for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
2015	2.7 ^{Aa}	0.8 ^{Bb}
2016	0.8 ^{Bb}	4.1 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns (p<0.05).

Table 17. Frequency scores based on seeding method for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
Big bluestem	2.3 ^b	4.4 ^a
Indian blanket-flower	1.3 ^b	1.7 ^a
New England aster	0 ^b	0.3 ^a

^{a-b} Different letters within a row indicate a significant difference ($p < 0.05$).

Table 18. Average frequency scores of wildflower islands compared to the whole (non-island area) plot for Experiment 2 (Kearney, NE).

	2015				2016			
	June		September		June		September	
	Islands	Whole plot	Island	Whole plot	Island	Whole plot	Island	Whole plot
Blackeyed Susan	2.2	0.1	2.8	0.6	0.7	0.0	1.1	0.0
Blue flax	3.6	0.2	2.1	0.0	1.2	0.0	0.6	0.0
Indian blanket flower	5.4	1.6	5.9	1.2	0.5	1.1	2.5	0.2
Maximilian sunflower	2.4	2.5	3.5	2.4	3.4	2.8	4.7	2.9
New England aster	0.7	0.0	0.1	0.0	0.0	0.4	0.6	0.0
Purple prairie clover	1.2	0.0	0.8	0.1	0.3	0.1	0.2	0.3
Rocky Mountain bee plant	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0
Roundhead lespedeza	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Shell-leaf penstemon	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0
Upright prairie coneflower	4.6	0.6	3.8	0.6	4.3	1.0	5.8	0.4
Weed forbs	6.7	9.5	6.9	8.9	2.8	0.4	0.4	0.4
Weed grasses	0.5	0.8	1.2	1.2	1.2	1.7	0.8	1.6

Table 19. Frequency scores of grasses based on year and date for Experiment 2 (Kearney, NE).

	June	September
Canada wildrye		
2015	2.4 ^{Aa}	0.7 ^{Bb}
2016	2.9 ^{Aa}	1.5 ^{Aa}
Little bluestem		
2015	0.9 ^{Ab}	1.2 ^{Aa}
2016	2.1 ^{Aa}	0.2 ^{Bb}
Slender wheatgrass		
2015	3.7 ^{Aa}	2.6 ^{Ab}
2016	3.8 ^{Aa}	5.6 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns (p<0.05).

Table 20. Frequency scores based on date for Experiment 2 (Kearney, NE).

	June	September
Sideoats grama	2.7 ^b	4.5 ^a
Western wheatgrass	0.9 ^a	0.2 ^b

^{a-b}Different letters within a row indicate a significant difference ($p < 0.05$).

Table 21. Frequency scores based on year for Experiment 2 (Kearney, NE).

	2015	2016
Big bluestem	2.3 ^b	4.4 ^a
Sideoats grama	2.5 ^b	4.8 ^a

^{a-b}Different letters within a row indicate a significant difference ($p < 0.05$).

Table 22. Frequency scores of Indiangrass for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
June	1.9 ^{Aa}	1.8 ^{Aa}
September	1 ^{Ba}	2.6 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns (p<0.05).

Table 23. Frequency score of weedy grasses for Experiment 2 (Kearney, NE).

	Seeding method	
	Standard	Wildflower island
2015		
June	1 ^{Ab}	0.8 ^{Aa}
September	3.6 ^{Aa}	1.2 ^{Ba}
2016		
June	2.6 ^{Aa}	1.8 ^{Aa}
September	1.6 ^{Aa}	2.3 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences with columns (p=0.05).

Table 24. Frequency of occurrence of Indian blanket-flower on the North and South sites from 2014 to 2016 for Experiment 3 (Bruning, NE).

	2014	2015	2016
	%		
North	14.3 ^{Aa}	6.8 ^{Ba}	5 ^{Ba}
South	3.6 ^{Ab}	2.3 ^{Ab}	1.5 ^{Ab}

^{A-B}Different capital letters indicate differences within rows and ^{a-b}lower case letters indicate differences within columns (p<0.05).

Table 25. Frequency of occurrence for purple prairieclover on the North and South sites from 2014 to 2016 for Experiment 3 (Bruning, NE).

	2014	2015	2016
	%		
North	1.5 ^{Ba}	2.3 ^{Ba}	6.1 ^{Aa}
South	0.1 ^{Aa}	0.5 ^{Ba}	2.1 ^{Ab}

^{A-B}Different capital letters indicate differences within rows and ^{a-b}lower case letters indicate differences within columns (p<0.05).

Table 26. Frequency of occurrence of blackeyed Susan for Experiment 3 (Bruning, NE).

	Mowing Treatment		
	0	1	2
Seeding rate	%		
North			
4 (lbs PLS/acre)	3.8 ^{Aa}	2 ^{Aa}	2.6 ^{Aa}
8 (lbs PLS/acre)	1.9 ^{Aa}	3.9 ^{Aa}	2.2 ^{Aa}
South			
4 (lbs PLS/acre)	2.6 ^{Ba}	8.6 ^{Aa}	4.7 ^{Ba}
8 (lbs PLS/acre)	1.8 ^{Aa}	2.7 ^{Ab}	4.3 ^{Aa}

^{A-B} Different capital letters indicate differences within rows and ^{a-b} lower case letters indicate differences within columns (p<0.05).

