

METHODS

Experiment 1 Design and Setup:

In preparation for experiment 1 a 70 x 120 m rectangular area of the field was sprayed with glyphosate herbicide summer 2000 and then plowed and disked March-April 2001. In mid April 2001 we established 96 4 x 5 m experimental plots, laid out in a 12 x 8 plot grid (Fig 1). The plot array was divided into

6 rectangular blocks of 16 plots, arranged to control for spatial effects associated with a slight field slope. Each block was further divided into two clusters of 8 plots. Blocks were separated by 12m buffers, clusters nested within blocks were separated by 10 m buffers, and plots nested within clusters were separated by 2 m buffers. To the array of plots we applied 16 unique treatment combinations ($n = 6$) in a $4 \times 2 \times 2$ factorial nested design. Nested in each cluster of each block, the first two factors were fully crossed in a 4×2 configuration: nitrogen (N) fertilizer added to plots as ammonium-nitrate at four levels (0, 4, 8 and $16 \text{ g N m}^{-2} \text{ yr}^{-1}$); and Phosphorus (P) added as super-phosphate at two levels (0 and $8 \text{ g P m}^{-2} \text{ yr}^{-1}$). In each block, one of the two

clusters of eight plots was randomly selected to receive multi-species seed additions (sowing treatment) to experimentally enhance the pool of species available to plots. To accomplish sowing, seeds of 50 native and naturalized species were sown in April 2002 and 2003. All sown species are members of the regional pool, chosen to represent a range of life histories, functional guilds, historical origins and habitat affinities. Seeds were sown at rates ranging from $10\text{-}320 \text{ seeds m}^{-2} \text{ yr}^{-1}$ year; depending on availability. To maintain plots as grassland, a schedule of spring prescribed burns was initiated in 2005 on a return frequency of once every three years. To date the plots have been burned twice (2005 and 2008).

Experiment 1 Data Collection:

Core data for Experiment 1 consists of plant species % cover and plant biomass data collected during annual vegetation surveys/harvests of all experiment plots. The annual

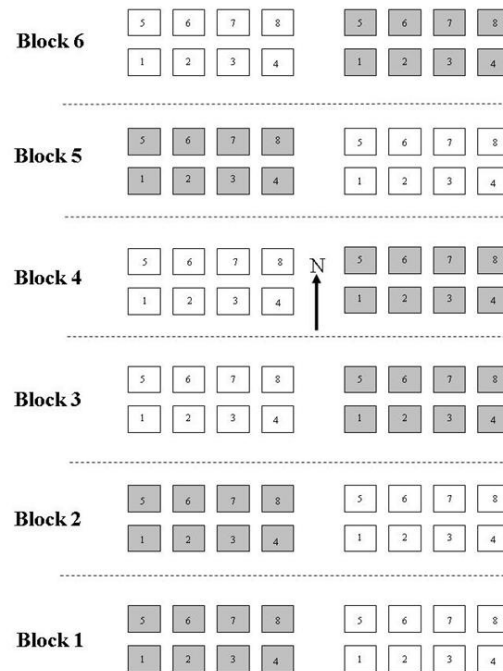


Figure 1. Experiment 1 plot layout. Each block contains two clusters of eight 4 x 5 m plots. To the plots in each cluster, a 4×2 factorial array of N and P fertilization treatments was applied. Shaded clusters received experimental seed additions.

core data time-series currently spans 2001-2009. After an additional decade the core data set will document plant species abundances and plant productivity in this experiment over a period of 19 years (2001-2019).

Percent cover - Cover surveys of vegetation are conducted annually in late June - early July by taking ocular estimates of percent cover for each species rooted within in two permanently-marked 1 x 1 m sampling quadrats in each plot (192 quadrats in all). Over the next decade we will continue the plant cover surveys as described above. However, beginning summer 2010 we will assess plant cover annually in each plot in two additional 1 x 1 m sampling quadrats (increasing the total number of quadrats per plot from two to four). These extra quadrats will allow us to better address new questions concerning community divergence and dispersion discussed above (Hypotheses 6-7).

Plant biomass - Biomass harvests of all plots are conducted annually in mid-late July. From each plot, one 0.1 x 2 m strip of aboveground plant material is harvested with electric clippers. Biomass is clipped at ground level and all litter collected. Samples are separated into live and litter fractions. All biomass fractions are dried to constant mass at 70°C in a forced-air oven and then weighed. Over the next decade we will continue the biomass harvests as described above.

Experiment 2 Design and Setup:

In March of 2000 we established a 4 x 4 grid of 16, 10 x 20 m plots at the site (Fig 2). Plots were separated by 3 m buffer strips. Each 10 x 20 m plot was divided into two contiguous 10 x 10 m subplots (yielding a total of 32 subplots). A 2 x 2 x 2 factorial set of treatments was assigned to the plots and subplots in a split-plot design: two levels of nutrient fertilization (fertilized; not fertilized) applied as the whole-plot factor; two levels of native prairie seed sowing (seed of 41 species sown; seed not sown) applied as a whole plot factor, and two levels of haying (hayed; not hayed) applied as the split-plot factor. There are four replicates of each treatment combination.

NPK fertilizer (29-3-4) was spread by hand to each 10 x 20 m whole-plot at a rate of 14-16 g N m⁻² per year. This application rate is at the high end of what is typically applied to production cool-season hayfields of the region. From 2002 to 2006, one subplot in each whole-plot was hayed annually in mid June as is typical for the cool-season hayfields of the region. In 2007 we decided to alter haying management in the non-fertilized plots by shifting the time of haying to late July - early August, the

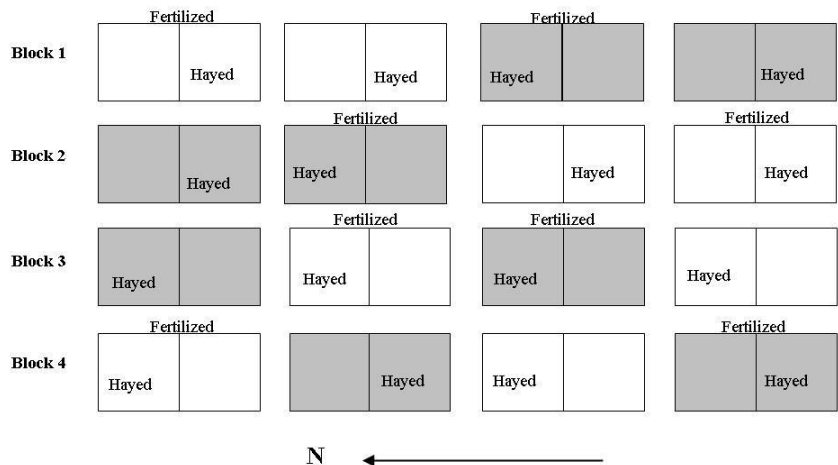


Figure 2. Experiment 2 plot layout. Each block contains four 10 x 20 m plots, each divided into two 10 x 10 m subplots. Shaded plots received experimental seed additions.

typical time of harvest for native warm-season hay meadows in our area. This was done because although fertilized plots remain dominated by cool-season grasses, non-fertilized plots have shifted to dominance by warm-season grasses. Over the next decade we plan on maintaining haying treatments in the following way: fertilized plots hayed in June, typical for cool-season hay fields; non-fertilized plots hayed in late July - early August in accordance with native hay meadow management. Seeds of 41 native prairie species, common to nearby native prairie and prairie hay meadows, were sown by hand into 8 of the 16 whole-plots in February of 2003 and 2004 at an average rate of 47 seeds per m² for each species.

Experiment 2 Data Collection:

Core data for Experiment 2 consists of plant species biomass and % cover data collected annually in all subplots. The dataset currently spans 2001-2009. After an additional decade of research, the core data set will document species abundances and productivity over a period of 19 years (2000-2019).

Plant biomass - Each year we sample aboveground biomass in all subplots, just prior to mid June haying when cool-season grasslands are at peak production. Beginning in 2007, after shifting the haying time for the non-fertilized plots to late summer, we initiated a second round of annual biomass sampling in late July, just prior to late summer haying to better estimate plant production and hay yields in the non-fertilized plots that are now dominated by warm-season grasses. To sample biomass, two 0.1 x 2 m strips of aboveground plant material are harvested in each subplot with electric clippers. Biomass is clipped at ground level and all litter collected. Samples are separated into live and litter fractions, with the live fraction sorted to species.

Percent cover - Starting 2003, the first year seeds were sown, we began conducting annual plant cover surveys in all subplots. Cover surveys are conducted each June prior to June haying. Beginning this year (2008), the timing of cover surveys conducted in the non-fertilized plots has been shifted to mid July, just prior to the time that non-fertilized plots are hayed. So from 2008 on, cover surveys will be conducted in June for fertilized plots which reach peak production at that time, July for non-fertilized plots which are near peak production at that time. Cover surveys are conducted in the central 8 x 8 m area of each 10 x 10 m subplot to capture dominant plant composition at a larger scale than assessed by biomass samples. From 2009 onward we will also conduct cover surveys in each subplot within six 1 x 1 m quadrats. These new cover quadrats will support tests of hypotheses 6 and 7 regarding spatial turnover and community dispersion as discussed above.