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Prairie Remnant Soil as a Source of Mycorrhizal Inoculum

Ricky D. Kemery[1] and Michael N. Dana[2]
Department of Horticulture, 1165 Horticulture Building, Purdue University, West Lafayette, IN 47907-1165

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Abstract. **Soil from four native prairie remnant sites was used as inoculum in pot culture to achieve vesicular–arbuscular mycorrhizal (VAM) infection of Sudangrass [*Sorghum sudanense* (Piper) Stapf]. The prairie sites varied in their management histories and degradation levels. Sudangrass plants that became infected with VAM grew better than those grown in standard pasteurized greenhouse mix or those grown in a pasteurized greenhouse–prairie soil mix. Soil from prairie remnants may serve as a beginning source of inoculum that can be increased via Sudangrass pot culture for inoculation of prairie plant seedlings in nursery production.**

Vesicular–arbuscular mycorrhizae (VAM) are of interest for ecological reasons and their potential value in crop and landscape plant culture. Associated with increased phosphorus uptake, increased growth, and an extended nutrient uptake zone of roots of many species (Hayman, 1983), VAM have naturally associated with tallgrass prairie species (Hetrick et al., 1987). VAM-colonized transplants may have a better chance of survival and successful establishment in the landscape than noninfected seedlings (Davies and Call, 1987). Native prairie remnants may be good sources of inoculum for use in production of VAM-colonized seedlings of prairie species that associate with VAM. However, Moorman and Reeves (1979) have suggested that previous site disturbance decreases the amount of inoculum present at prairie sites. Several prairie remnants exist with differing site histories in west-central Indiana within an 80-km radius of West Lafayette, Ind. Sudangrass has been used often as a nonspecific host for VAM fungi in greenhouse studies (Claassen and Zasoski, 1992).

Our purpose was to determine 1) if VAM inoculation of Sudangrass with soil from four Indiana prairie remnants would result in VAM infection and 2) to inquire whether VAM infections would result in differential growth of the Sudangrass in relation to the site management histories of the inoculum sources.

Materials and Methods

Samples were collected from the upper 0.5 m of soil from each of four tallgrass prairie remnant sites in Indiana during Nov. 1991. Dhillion and Anderson (1993) have reported a decline in spore count during summer months, though seasonal variation in VAM spore abundance may vary from year to year (Cooke et al., 1992). The dominant grass species on all four sites were big bluestem (*Andropogon gerardii* Vitman) and indiangrass [*Sorghastrum nutans* (L.) Nash]. Ten 30-g samples from random locations within each remnant were collected and combined to create one composite sample from each site. The soil was stored in separate plastic bags at 3C.

The prairie sites where the soil was collected were 1) Ambia-Talbot RR Prairie, Ambia, Ind.—a 4 km × 20 m railroad right-of-way and an adjoining strip of land with little apparent disturbance and high species diversity, including closed gentian (*Gentiana andrewsii* Griseb.) [soil type, Chalmers silt loam (a fine silty, mixed, mesic Typic Haplaquoll)]; 2) Shiloh Cemetery, Thorntown, Ind.—a 0.25-ha, 19th century, pioneer cemetery with evidence of past disturbance, including introduced plantings of sugar maple (*Acer saccharum* Marsh.) and daylily [*Hemerocallis fulva* (L.) L.] [soil type, Reesville silt loam (a fine-silty, medium, mesic Aeric Ochraqualf)]; 3) Smith Cemetery, Cayuga, Ind.—a 0.1-ha, 19th century pioneer cemetery with a history of mowing management but containing several highly conservative species, including royal catchfly (*Silene regia* Sims) [soil type, Shipshe loam (a loamy-skelatal mixed, mesic Typic Argiuddel)]; and 4) Spinn Prairie, Reynolds, Ind.—a 40-ha, Nature Conservancy-owned property of prairie and oak savanna that was a site of swine production [soil type, Seafield fine sandy loam (a coarse-loamy, mixed, mesic Udollic Ochraqualf)].

A quantitative ranking of the sites by disturbance level is impossible. However, based on the qualitative observations of field naturalists who are well-acquainted with all the sites (L. Casebere, M. Homoya, and D. Zay, 1994, Naturalists, Indiana Dept. of Natural Resources, Division of Nature Preserves; personal communication), Ambia-Talbot is the least disturbed and least degraded site of the four included in this study. The Shiloh site is the most degraded; the Smith site is mostly undisturbed, but of intermediate naturalistic value due to its small size and low species diversity. Based on its large size and the many species present, naturalists value the Spinn site highly. However, it is regarded as substantially degraded due to the past hog production on the site. In recent years, the Spinn site has benefited from migration of native species from an adjacent railroad right-of-way, which was not included in the soil collection from the Spinn location.

Our preliminary research showed a 1 raw soil : 3 greenhouse medium mixture (v/v) maximized VAM infection. Soil from each prairie site was uniformly mixed at the 1:3 ratio with a steam-pasteurized (82C) greenhouse potting medium [20% topsoil, 40% peat, 40% perlite (by volume)] and placed in 10-cm square containers (250 ml). A noninoculated medium (source A) contained the greenhouse potting mix only. A second control (source B) contained a prairie soil inoculum blend created by combining equal quantities of soil from all four prairie sites; it was mixed at a 1:3 ratio with greenhouse potting mix and then autoclaved for 2 h at 82C to kill any existing VAM. Uniform, 1-week-old seedlings of 'Sudex' Sudangrass were transplanted from peatlite-filled seedling flats into the containers and were allowed to grow for 2 months. There were 10 replicate containers of each treatment. Pots were arranged on a greenhouse bench in a completely randomized design and were hand-watered daily with a mixture containing 201N–200K–46P (in mg·liter^{-1}).

At the conclusion of the growing period, total plant height was measured. Plant crowns were excised and dehydrated in an oven (24 h, 55C). Total combined shoot plus leaf dry weights were recorded. Root samples, 0.5 g each, were randomly collected from five locations from each plant. All the root samples within each treatment then were combined and stained to assess the presence of VAM using the container system of Claassen and Zasoski (1992) with the staining procedures of Koske and Gemma (1989). Plant height and dry-weight data were subjected to analysis of variance (ANOVA) using SuperANOVA (Abacus Concepts, Berkeley, Calif.). Mean comparisons were obtained using Fisher's protected test of least significant differences.

Results

VAM infection. Staining revealed that VAM infection occurred in all Sudangrass plants inoculated with soil from the prairie sites. Infection was absent in the root samples taken from either sources A or B.

Plant height. Sudangrass inoculated with soil from the Ambia-Talbot, Shiloh, and Smith sites was significantly taller than plants from either source A or Treatment B (Table 1). For example, plants with Ambia-Talbot inoculum were an average of 17.3 cm taller than plants grown in standard greenhouse mix (source A). Plant height for source A and B (noninoculated

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[1]Graduate Assistant.
[2]Associate Professor.

and steam-pasteurized) were similar. Sudangrass inoculated with soil from the Spinn site, which had been highly degraded, was not significantly taller than grass from either source A or B.

The Ambia-Talbot and Shiloh soils produced plants of similar height and plants from either source were significantly taller than those from the Spinn source.

Dry weight. Shoot plus leaf dry weights of Sudangrass plants grown in medium inoculated with soil from any of the prairie remnant sites exceeded those from source A or B (Table 1). Plants grown with Smith inoculum weighed at least 40% more than plants from either source A or B.

The Smith inoculum produced plants with dry weights significantly higher ($P < 0.02$) than any of the other inoculations. Sudangrass grown with inoculum from the Spinn site weighed significantly less than plants from the Ambia-Talbot source, but not less than plants from the Shiloh source. Shiloh plants weighed less than plants from the Ambia-Talbot treatment ($P \leq 0.05$).

Table 1. Plant height and shoot plus leaf dry weight of Sudangrass grown for 2 months in media with or without prairie remnant soil as vesicular–arbuscular mycorrhizae inoculum. Values are means of 10 replicates.

Inoculum treatment	Relative site disturbance level	Plant ht (cm)	Shoot plus leaf dry wt (g)
Ambia-Talbot	Low	85.5 a[z]	23.1 b
Shiloh Cemetery	High	83.2 ab	21.7 c
Smith Cemetery	Low	78.6 b	24.6 a
Spinn Prairie	High	73.7 c	21.6 c
Source A	NA	68.1 c	13.0 d
Source B	NA	71.6 c	14.2 d
$LSD_{0.05}$		6.2	1.3

[z]Mean separation in columns by Fisher's protected test.

Discussion

This study shows the promotional effect of prairie soil inoculum on growth of Sudangrass compared to plants grown without prairie soil inoculum. It also demonstrates that prairie soil inoculum that has been autoclaved to kill microorganisms (source B) does not confer a similar growth promotional effect. This growth promotional effect is most likely due to VAM infection. The study also demonstrated that raw prairie soil can serve as VAM inoculum.

The interpretation of the growth responses resulting from the various prairie remnant inoculum treatments is less straightforward. It is complicated by the absence of a quantitative measure of the degree of degradation of the remnant sites. The two measures of growth in this study showed no consistent relationship to the apparent disturbance levels. However, among the shoot plus leaf dry weight data alone, inoculum from the less disturbed sites (Ambia-Talbot and Smith Cemetery) produced plants with significantly higher dry weights than inoculum from the sites with relatively greater disturbance levels (Shiloh Cemetery and Spinn Prairie).

Thus, at least with respect to the dry weight data, our results are consistent with those of Moorman and Reeves (1979), who indicated that soil inoculum from more disturbed sites provides less growth promotion than inoculum from less disturbed sites.

J. Morton and S. Bentiveiga at the International Vesicular–Arbuscular Mycorrhizae Collection (INVAM) at West Virginia Univ. examined samples from all four remnant sites and identified the fungal species from the Ambia-Talbot site as *Glomus geosporum* (Nicholson & Gerdemann) Walker, *Glomus mosseae* (Nicol. & Gerd.) Gerdemann & Trappe, and another undescribed *Glomus* species (personal communication).

Only a small amount of prairie soil is needed initially to produce large quantities of inoculum using the INVAM pot culture method (Bentiveiga, 1992, personal communication). Few undisturbed and nondegraded prairie sites exist in Indiana. If VAM inoculation is used in plant production to enhance establishment or survival of prairie species, nurseries could use VAM inoculum obtained via pot culture initiated by soil from limited field collection.

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