Forage Research in Texas, 1992

Evaluation of New Inoculation Techniques for Clovers

G. W. Evers, J. M. Moran, and J. L. Gabrysch

Summary

HiStick and other new clover inoculants were compared with Pelinoc-Pelgel, uninoculated seed, and uninoculated seed + 60 lb nitrogen/A on 'Yuchi' arrowleaf clover (*Trifolium vesiculosum* Savi). The arrowleaf clover was overseeded on a 'Coastal' bermudagrass [*Cynodon dactylon* (L.) Pers.] sod in October of 1990 and 1991. Nodulation and forage production from HiStick-inoculated seed equaled or exceeded that from seed inoculated with Pelinoc. Early forage production was related to nodules/seeding at 6 weeks after planting. Nodulation of the uninoculated seed treatments by native rhizobia the second year prevented any significant yield differences among treatments.

Introduction

Clover growth and use of nitrogen (N) from the air depends on early infection through the root hairs by specific types of bacteria (Ball et al., 1991). These bacteria form nodules on clover roots and transform gaseous N from the air into ammonium (NH_4), which can be used by the clover plant. Many strains of *Rhizobium* bacteria infect legumes, and the amount of N they fix varies. Inoculum containing specific strains are applied to the clover seed to make certain the best strains are present when the clover seed germinates.

Work by Waggoner et al. (1979) showed that the Pelinoc-Pelgel inoculation procedure was superior to applying regular peat inoculant to the seed with water. New inoculation products for clovers are entering the market. A study was conducted for 2 years at the Overton Research and Extension Center to compare the performance of these new inoculation products to Pelinoc on Yuchi arrowleaf clover.

Procedure

Treatments were (1) uninoculated seed, (2) uninoculated seed plus 60 lb N/A at planting, (3) Pelinoc-Pelgel inoculant, (4) HiStick inoculant, (5) HiStick-experimental inoculant, and (6) HiCote preinoculated seed. Arrowleaf seed was shipped to the inoculant company, where the HiCote treatment was applied and then shipped back to Overton, where the preinoculated seed was stored in a refrigerator until planting. All other inoculation treatments were applied immediately before planting.

Experimental design was a randomized complete block with four replications and a plot size of 5 x 15 ft. The seed was drilled into a short Coastal bermudagrass sod on October 11, 1990, and October 3, 1991. Both years, 80 lb phosphorus (P_2O_5), 80 lb potassium (K_2O), and 1 lb boron (B) per acre were surface-applied. Approximately 6 weeks after emergence, 10 seedlings were removed from each plot to determine weight, leaves, and nodules per seedling. Dry matter production was determined by harvesting the plots twice during the growing season.

Results and Discussion

In the first year, all treatments except the uninoculated treatment resulted in similar number of leaves per seedling (Table 1). Seedlings from the four inoculation techniques ranged from 11 to 15 nodules per seedling. The two uninoculated treatments averaged less than two nodules per seedling, which indicates that few native rhizobia at the test site were capable of infecting arrowleaf clover seedlings. Weight of seedlings from the HiCote and HiStick-experimental treatments were equivalent to the seedlings from the uninoculated + 60 lb N at planting treatment. Weight of seedlings from the HiStick and Pelinoc treatments were slightly less. Arrowleaf seedlings from the uninoculated treatment were stunted because of N deficiency.

Table 1. Influence of inoculation treatments on leaves, nodules, and weight of Yuchi arrowleaf clover seedlings, 6 weeks after emergence in autumn, 1990.

Treatment	Leaves	Nodules	Dry weight
			.........g.........
HiCote	3.6 a*	13.6 ab	0.0485 a
Pelinoc	3.1 b	11.8 b	0.0334 c
HiStick	3.2 ab	13.2 ab	0.0356 bc
HiStick (exp.)	3.5 ab	15.1 a	0.0440 ab
Uninoculated	2.5 c	1.1 c	0.0208 d
Uninoculated + N†	3.4 ab	1.7 c	0.0455 ab

*Values within a column followed by the same letter are not significantly different at the 0.05 level, Waller-Duncan Multiple Range Test.

†On day after planting, 60 lb N/A applied to soil surface.

Keywords: clovers / inoculation / N_2-fixation.

Seedling differences among treatments were small the second year (Table 2). Although clover had not been grown on the site, various native legume species volunteered in and near the study. Good nodulation of the arrowleaf seedlings in the uninoculated treatment indicated that native rhizobia were present and capable of infecting and nodulating arrowleaf clover. The HiStick-experimental treatment produced significantly more nodules/seedling than the other treatments, and the uninoculated treatment had significantly less. Seedling weight was highest from the HiStick-experimental treatment, uninoculated + N treatments were the heaviest, and seedlings from the uninoculated plot were the smallest.

In the first year, dry matter yields at the first harvest (Table 3) were directly related to nodules per seedling at 6 weeks after planting. The Pelinoc treatment was less productive than HiCote, HiStick, and HiStick-experimental treatments on April 2.

Table 2. Influence of inoculation treatments on leaves, nodules, and weight of Yuchi arrowleaf clover seedlings, 6 weeks after emergence in autumn, 1991.

Treatment	Leaves	Nodules	Dry weight
			........g........
HiCote	2.8	5.8 b*	0.0178 bc
Pelinoc	2.8	6.1 b	0.0166 bc
HiStick	3.0	5.8 b	0.0172 bc
HiStick (exp.)	3.1	7.2 a	0.0200 ab
Uninoculated	2.7	4.1 c	0.0149 c
Uninoculated + N†	3.1	6.2 b	0.0231 a

*Values within a column followed by the same letter are not significantly different at the 0.05 level, Waller-Duncan Multiple Range Test.

†On day after planting, 60 lb N/A applied to soil surface.

Table 3. Influence of inoculation treatments on dry matter yield of Yuchi arrowleaf clover in spring, 1991.

Treatment	April 2	June 5	Total
	....	lb/A	....
HiCote	1394 a*	3220	4614 ab
Pelinoc	1034 b	3152	4186 b
HiStick	1363 a	2944	4307 ab
HiStick (exp.)	1377 a	3337	4714 a
Uninoculated	388 c	2887	3275 c
Uninoculated + N†	525 c	2937	3462 c

*Values within a column followed by the same letter are not significantly different at the 0.05 level, Waller-Duncan Multiple Range Test.

†On day after planting, 60 lb N/A applied to soil surface.

Forage yield of the uninoculated treatment was only 10% of the better inoculation treatments. Nitrogen applied at planting met the N needs of the clover seedling for the first 6 weeks in the uninoculated + 60 lb N treatment. But by the first harvest, clover growth was severely restricted by insufficient N because of few nodules per seedling at 6 weeks. Yield differences among treatments by the second harvest were not significant. It is not uncommon for large differences in nodulation at the seedling stage to decrease during the clover growing season. Rhizobia in poorly nodulated seedlings multiply rapidly with increasing temperature in the spring, which improves nodulation. Differences among treatments in dry matter production during the second year were not significant (Table 4). Production from the uninoculated treatments was good because seedlings were nodulated at 6 weeks (Table 2).

Table 4. Influence of inoculation treatments on dry matter yield of Yuchi arrowleaf clover in spring, 1992.

Treatment	March 11	May 14	Total
	....	lb/A	....
HiCote	1003	2866	3869
Pelinoc	1121	3173	4294
HiStick	1054	2723	3777
HiStick (exp.)	1143	3254	4397
Uninoculated	736	2421	3157
Uninoculated + N†	971	2660	3631

†On day after planting, 60 lb N/A applied to soil surface.

HiStick inoculant was sold in certain areas in the autumn of 1991. It should be available throughout the southeastern United States beginning in the autumn of 1992. Data from this study indicates that HiStick performed as well as the Pelinoc-Pelgel inoculation system. HiStick is easier to use because it is a two-step process, whereas Pelinoc requires four-steps. A sterilized, peat-based inoculant is used in HiStick, which gives it a shelf life of 2 years if not exposed to high temperatures.

Literature Cited

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