Peanut Science (1983) 10:79-82

Inoculation of Peanuts on Farmers' Fields in Alabama[1]

A. E. Hiltbold*, D. L. Hartzog, R. B. Harrison, and Fred Adams[2]

ABSTRACT

Farmers occasionally put land into peanut (*Arachis hypogaea* L.) production that has not been cropped with peanuts in recent history. These fields are generally sparsely populated, if at all, by peanut rhizobia and may be responsive to application of effective peanut inoculant. To field test the need for inoculants, 13 experiments were conducted during 1980-1982 on "new" land on farms in the peanut producing region of southeastern Alabama. Treatments were (1) uninoculated control, (2) granular inoculant applied in-furrow at 3X recommended rate, and (3) ammonium nitrate at 56 kg/ha of N at planting. In 1980, an additional 56 kg N was applied at early bloom. The soil was sampled prior to treatment and numbers of rhizobia capable of nodulating peanuts determined by the most probable number (MPN) procedure. Numbers ranged from nil to 1600 rhizobia per g of soil. Although the soil at six locations contained fewer than 20 rhizobia per g, no yield responses to applied inoculant were obtained. Vine growth, leaf color, and nitrogen content of leaves were unaffected by inoculant. The inoculant provided 1 million rhizobia per seed, which is considered abundant. Lack of any yield responses to fertilizer nitrogen during the 3-year study

[1]Contribution of the Department of Agronomy and Soils, Alabama Agricultural Experiment Station, Auburn University, AL 36849.

[2]Professor, Agronomist-Peanuts, Graduate Research Assistant, and Professor, respectively. Published with the approval of the director of the Alabama Agric. Exp. Stn. as Journal Series No. 3-83432.

indicated nitrogen sufficiency in the peanut plants, derived through nodulation and nitrogen fixation by native soil rhizobia. While peanut was not a host legume for these rhizobia during the years prior to these experiments, the rhizobia apparently persisted on alternate legume hosts in the cowpea miscellany in numbers adequate for effective inoculation of peanuts.

Key Words: *Rhizobium*, rhizobia, inoculant, nitrogen fixation, *Arachis hypogaea*.

Peanut (*Arachis hypogaea* L.) and its root nodule bacteria (*Rhizobium* spp.) fix large amounts of atmospheric nitrogen. Effective rhizobia, either occurring naturally in the soil or applied at planting, are essential for nitrogen fixation and economic yield. This became apparent with early research during the expansion of peanut production to previously unplanted soils in central Alabama. Field experiments by Duggar (5) in the early 1930s showed yield responses from soaking unhulled spanish peanut seed in a suspension of rhizobia cultured from peanut nodules. Yields were correlated with the number of large nodules per plant. In further research during the early 1940s on new peanut land in Alabama, Albrecht (1) found that yield responses to fertilizer and lime applications were dependent upon seed inoculation. A more recent report from Florida (4) shows a yield increase of 750 kg/ha with inoculation. However, similar experiments on soils that had grown peanuts in the preceding 3 years showed no responses to inoculation. Reddy et al. (8) obtained yield increases up to 750 kg/ha from inoculation of peanuts planted for the first time in southern Canada. In these experiments, uninoculated controls averaged only a single nodule per plant, indicating very few soil rhizobia capable of nodulating peanut. Mahler and Wollum (6) recently examined soils in North Carolina to determine populations of peanut rhizobia. The 15 peanut fields sampled averaged 500 rhizobia per g of soil, while fewer than 10 rhizobia per g were found in orchard and forest soils. It has long been known that rhizobia numbers in soil increase in the presence of the legume host (10). The question of how many is enough has not been answered.

Farmers occasionally bring long-term pasture, forest, or idle land into peanut production. These soils generally contain few rhizobia; peanuts planted in them should be most responsive to applied inoculant. To test this hypothesis, field experiments were conducted on farms where peanuts were planted after extended periods without peanuts, or where peanuts had not been previously grown.

Materials and Methods

Isolated fields in the peanut-producing area of southeastern Alabama were located that were to be planted in peanuts for the first time in recent history. Neither were there nearby fields with recent histories of peanut plantings. Crop needs for lime, phosphate, and potash were determined by the Auburn University Soil Testing Laboratory. The farmer applied lime, fertilizer, and gypsum as needed for that field. Experimental treatments were (a) granular peat inoculant at 16.8 kg/ha (three times manufacturer's recommended rate) placed in-furrow with the seed, (b) uninoculated control, and (c) ammonium nitrate (56 kg/ha of N at planting each year, with additional 56 kg/ha at early bloom in 1980). Inoculant was applied through calibrated applicators on the farmers' planters. Florunner peanut seed were planted at a rate of 112 kg/ha in rows spaced 91 cm apart. Treatments were in randomized complete block design with 4 replications; plots were 4 rows wide and 27 m long with 1-m alleys between blocks. Recommended pest control practices were followed by the farmers as needed. Peanuts were dug and harvested with each farmer's equipment. The two inner rows of each plot were harvested; yields were adjusted to 10% moisture. Percent sound mature kernels (SMK) was determined by standard procedures.

In 1982, peanut leaves were sampled in each plot on July 7 and August 4. Total nitrogen content of dried, ground samples was determined by the Kjeldahl procedure.

Two experiments were harvested in 1980, five in 1981, and six in 1982. New sites were selected each year. Prior to application of inoculant, the soil was sampled to plow depth for determination of peanut rhizobia. Moist samples were screened through a 4-mm mesh, sterilized screen. Moist soil, equivalent to 10 g oven-dried soil, was serially diluted in sterile water in 10-fold steps. One-ml aliquots were applied to surface-sterilized peanut seed in plastic growth pouches containing minus-N nutrient solution (9), using five pouches at each of four dilutions selected to bracket the expected number of rhizobia. Roots were examined for nodulation at 3 weeks after inoculation. A single nodule was sufficient to render a pouch positive. From the frequency of positives and the dilution ratio, the most probable number (MPN) was obatined (2).

In 1980 and 1981, additional estimates were made of rhizobia in soil from the field sites by growing peanut plants from surface-sterilized seed in undiluted soil in pots. After 3 weeks, the roots were washed free from soil and their nodules counted.

Results and Discussion

Soil type, soil pH, and recent history are listed for each site in Table 1. All soils were coarse-textured and acid. No peanuts had been grown on these soils for at least 15 to 20 years. MPN determinations, however, showed all soils to contain rhizobia capable of nodulating peanuts, ranging from about 1 to 1600 per g of soil. While some fields were relatively sparse in rhizobia, similar fields contained hundreds per g of soil. The occurrence of this many rhizobia probably is associated with alternate legume hosts in these fields. The cowpea miscellany, to which peanut belongs, includes a wide variety of cross inoculating legumes. Carroll (3) listed 63 species in 10 genera of legumes in the cowpea group. Since rhizobia can persist for extended periods in soil in the absence of their legume hosts, it is also possible that the observed rhizobia descended from those in earlier, unrecorded peanut crops. Nutman (7) found rhizobia capable of nodulating alfalfa, clover, peas, and lupine in field plots at Rothamsted that had grown wheat continuously for 125 years.

Nodules developed on peanut seedlings grown in potted soil containing as few as 6 rhizobia per g of soil (Table 1). In general, nodule numbers increased with increasing concentration of soil rhizobia. Four other experiments in 1980 (lost to drought) had soils with MPN of 4, 8, 20, and 170 rhizobia per g that produced 0, 2, 4, and 7 nodules per plant, respectively, on peanuts in potted soil. Theoretically, a single cell of the appropriate *Rhizobium* in the rhizosphere of the seedling is sufficient to induce nodulation.

Treatment effects were not visually apparent in any stage during growth of peanuts in any experiment. Color of leaves and vigor of plants were generally good, with little incidence of leaf or stem diseases. Drought restricted growth at times. Determination of nitrogen contents of foliage at two dates in 1982 showed no differ-

Table 1. Number (MPN) of indigenous rhizobia in field samples and number of nodules produced by them in peanuts planted in potted soil.

Field and soil type	Soil pH	Previous crop	Peanut rhizobia MPN per g soil	Nodules per plant[1]
		1980		
Deloney I				
Lucy ls (Arenic Paleudult)	5.7	bahiagrass (15 y)	6	1
Deloney II				
Altavista fsl (Aquic Hapludult)	5.5	woods, soybeans 1979	11	3
		1981		
Deal I				
Dothan ls (Plinthic Paleudult)	5.5	idle (20 y)	1300	10
Deal II				
Dothan ls (Plinthic Paleudult)	5.5	idle (20 y)	1100	14
Trawick I				
Troup ls (Grossarenic Paleudult)	5.4	woods	12	9
Price				
Lucy ls (Arenic Paleudult)	5.5	bahiagrass (36 y)	32	8
Harden				
Red Bay ls (Rhodic Paleudult)	5.4	planted pines (28 y)	490	4
		1982		
Deloney III				
Dothan ls (Plinthic Paleudult)	4.6	row crops, no peanuts	9	-
Deloney IV				
Gritney ls (Typic Hapludult)	5.1	woods, watermelon 1981	1	-
Trawick II				
Troup ls (Grossarenic Paleudult)	5.6	bahiagrass (20 y)	1600	-
Deal III				
Lucy ls (Arenic Paleudult)	5.0	idle (20 y)	450	-
Deal IV				
Fuquay ls (Arenic Plinthic Paleudult)	4.9	idle (20 y)	17	-
Pope				
Dothan ls (Plinthic Paleudult)	5.0	planted pines	680	-

[1]6 plants per pot

Table 2. Total nitrogen content of peanut leaves in experiments on farmers' fields, 1982.

Field	Total N[1] July 7	Total N[1] August 4	Coefficient of Variation
	%	%	%
Deloney III	2.57	2.31	7
Deloney IV	2.62	2.20	8
Trawick II	2.19	2.77	5
Deal III	1.82	2.01	-
Deal IV	1.91	2.26	6
Pope	3.00	2.50	15

[1]Significant difference ($P < .05$) between dates at each field; no differences attributable to inoculation or ammonium nitrate. Values shown are averages across 3 treatments and 4 replications.

ences attributable to inoculant or fertilizer N (Table 2).

Peanut yield did not respond to inoculation in any of the 12 experiments (Table 3). The four Deloney experiments, on soils containing only 1 to 11 rhizobia per g, would be expected to be most responsive to inoculation. While their numerical responses ranged from 130 to 590 kg/ha, none was significant at $P = .05$. Other experiments showed inoculated peanuts yielded as much as 310 kg/ha above or below the uninoculated control, without statistical significance. The precision of the experiments is indicated by the coefficients of variation, ranging from 4 to 22%. These CVs are typical of field plot research.

Table 3. Peanut yield and quality, inoculation experiments on farmer-fields in 1980, 1981, and 1982.

Field	Treatment[1] None	Treatment[1] Inoculant	Treatment[1] am. nit.[2]	Coefficient of variation	SMK[1]
	kg/ha	kg/ha	kg/ha	%	%
		1980			
Deloney I	3260	3400	2910	18	70
Deloney II	2910	3500	2930	14	72
		1981			
Deal I	3550	--	3930	22	74
Deal II	4400	4710	4860	11	76
Trawick I	4690	4710	4460	10	73
Price	4520	4210	4400	15	74
Harden	4670	4840	4490	4	79
		1982			
Deloney III	5310	5470	4350	12	71
Deloney IV	5170	5300	5120	9	74
Trawick II	4130	3830	3880	5	72
Deal III	4360	4560	--	9	75
Deal IV	2910	2690	--	9	74
Pope	3010	2920	2310	15	74

[1]No significant differences ($P < .05$) in peanut yields or SMK attributable to treatments in any year or field. Values shown are averages across 4 replications.

[2]In 1980, 112 kg/ha of N was applied as ammonium nitrate, split 56 at planting and 56 at early bloom. In 1981 and 1982, 56 kg/ha of N was applied at planting only.

In 1982, the viability of the granular inoculant was determined by serial dilution and MPN to contain 1.7×10^7 peanut rhizobia per g. At the rates of seeding and inoculant applied, more than 1 million rhizobia were provided per seed, which is considered to be more than adequate. Nodulation of plants in the field was not evaluated; however, vegetative growth suggested no nitrogen deficiency. Peanut yields (Table 3) did not respond to fertilizer nitrogen in any experiment, supporting the conclusion that nodulation and nitrogen fixation by indigenous rhizobia in the controls were sufficient for maximum yield under these field conditions. Rhizobia that are effective on peanut are apparently widely distributed in soils of the peanut producing region of southeastern Alabama. They are not restricted to fields currently in production but occur in forest, pasture, and noncrop areas. Their populations are frequently sparse after long periods without the peanut host; nonetheless, they are effective in nodulating and fixing N in peanut, as demonstrated by the lack of response to applied inoculant or fertilizer nitrogen.

Literature Cited

1. Albrecht, H. R. 1943. Factors influencing the effect of inoculation of peanuts grown on new peanut lands. Soil Sci. Soc. Am. Proc. 8:217-220.
2. Alexander, M. 1965. Most-probable-number method for microbial populations. *In* C. A. Black, Ed. Methods of soil analysis, Part 2, Chemical and microbiological properties. Agronomy 9. Am. Soc. Agron., Madison, WI.
3. Carroll, W. R. 1934. A study of *Rhizobium* species in relation to nodule formation on the roots of Florida legumes: I. Soil Sci. 37:117-134.
4. Cobb, L. C., and E. B. Whitty. 1973. Response of peanuts to inoculation with nitrogen-fixing bacteria. J. Am. Peanut Res. Educ. Assoc. Inc. 5(1):211-212.
5. Duggar, J. F. 1935. The effects of inoculation and fertilization of spanish peanuts on root nodule numbers. J. Am. Soc. Agron. 27:128-133.
6. Mahler, R. L., and A. G. Wollum, II. 1981. Seasonal fluctuation of peanut rhizobia in coastal plain soils. Peanut Sci. 8:1-5.
7. Nutman, P. S. 1969. Symbiotic nitrogen fixation: Legume nodule bacteria. Rothamsted Report for 1968, part 2, pp. 179-181.
8. Reddy, V. M., J. W. Tanner, R. C. Roy, and J. M. Elliot. 1981. The effects of irrigation, inoculants, and fertilizer nitrogen on peanuts (*Arachis hypogaea* L.). II. Yield. Peanut Sci. 8:125-128.
9. Weaver, R. W., and L. R. Frederick. 1972. A new technique for most probable number (MPN) counts of rhizobia. Plant Soil 36:219-222.
10. Wilson, J. K., and T. L. Lyon. 1926. Growth of certain microorganisms in planted and unplanted soil. Cornell Univ. Agric. Exp. Stn. Memoir 103.

Accepted October 21, 1983