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Rhizobium japonicum Nodular Occupancy, Nitrogen Accumulation, and Yield for Determinate Soybean Under Conservation and Conventional Tillage[1]

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ABSTRACT

Nitrogen fixation and accumulation are important facets of soybean (*Glycine max* L.) production. Three determinate soybean cultivars were grown on a Norfolk loamy sand (Typic Paleudult) with either conservation or conventional tillage. Plots were split for inoculation with strain 311b 110 of *Rhizobium japonicum*. Dinitrogen fixation was estimated by the difference between total shoot N for nodulating and nonnodulating 'Lee' isolines, and net N returned to the soil was estimated by the difference in dinitrogen-fixed N and seed N. Drought was significant the first year; only 19.5 cm of rain fell during the podfill period. Rainfall and soil water were adequate the second year. Tillage, cultivar, and inoculation did not consistently affect the percentage of nodules formed by particular rhizobial strains, individually, but specific treatment combinations significantly affected nodular occupancy by certain strains. Inoculated 'Coker 338' had the highest shoot total N accumulation in both years; however, the difference between years overshadowed tillage, cultivar, or inoculation effects. For the 2-y period, percentages of N supplied by N_2 fixation were estimated to be 58 to 67% and 49 to 65% under conservation and conventional tillage, respectively. Yields generally did not differ significantly for tillage or inoculation; however, Coker 338 soybean was lowest in seed yield even though it had the highest total N accumulation in the shoots. Estimates of net N returned to the soil varied more between years than between tillage treatments; values ranged from 14 to 40 kg ha^{-1} in 1980 and 57 to 123 kg ha^{-1} in 1981.

Additional index words: Dinitrogen fixation, Strains, Inoculation, Nonnodulation, Rainfall pattern, Soil nitrogen, Acetylene reduction, Nodulation.

NITROGEN accumulation and fixation are very important to soybean (*Glycine max* L.) growth and yield. Total N accumulations of > 200 kg ha^{-1} are common (Bhangoo and Albritton, 1976; Bezdicek et al., 1978; Kohl et al., 1980; Hunt et al., 1981; Patterson and LaRue, 1983). Net N returned to the soil (N accumulated from N_2 fixation minus N removed in seed) is also very important to crops that follow soybean in rotation. Corn yields are generally increased following soybean as opposed to continuous corn. These increases have been attributed to both net soil N increases and rotational effects. In the Midwest or other areas with high N content soils, the annual balance of N returned to the soil from soybean is likely to be negative (Welch et al., 1973; Johnson et al., 1974, 1975). However, Kohl et al. (1980) and Patterson and LaRue (1983) found N_2 fixation to account for 50 to 84% of accumulated N as measured by both ^{15}N dilution and difference methods when large amounts of carbonaeous materials were used to immobilize soil N. In the southeastern Coastal Plain, N_2 fixation has accounted for > 75% of accumulated N in soybean grown on a loamy sand under irrigated conditions (Matheny and Hunt, 1983). Under these conditions of high N_2 fixation and total N accumulation, the annual return of N to the soil may be positive.

Soil N levels, acidity, and temperature are known to affect nodulation and N_2 fixation (Weber, 1966a, 1966b; Bezdicek et al., 1974; Bhangoo and Albritton, 1976; Keyser and Munns, 1979a, 1979b; Lindemann and Ham, 1979). These soil conditions differentially affect growth and symbiotic performance of various *Rhizobium japonicum* strains (Lindemann and Ham, 1979; Mahler and Wollum, 1980, 1981). Different populations of *R. japonicum* strains are likely to cause different nodule occupancy, and this, in turn, may affect the N accumulation and/or soybean yield (Munevar and Wollum, 1981; Hunt et al., 1981, 1983; Williams and Phillips, 1983; Morris and Weaver, 1983).

There is currently increased interest in tillage practices that conserve natural resources while optimizing profitable crop yield. Since 1973, conservation tillage usage in the southeastern USA has increased nearly 80% (Christensen and Magleby, 1983). Conservation tillage has been shown to affect the soil environment (Bennett et al., 1973; Blevins et al., 1977, 1978, 1983; Doran, 1980a, 1980b; Tyler et al., 1983). It would, therefore, seem reasonable that conservation tillage in the southeastern Coastal Plain could affect *R. japonicum* nodular occupancy levels in determinate soybean as well as N accumulation, N_2 fixation, and yield. These factors would also likely affect soil N carryover to the subsequent crop. Lindemann et al. (1982) concluded that conservation tillage did not greatly affect nodulation, acetylene reduction, or seed yield of indeterminate soybean grown in Minnesota. Yet, these effects have not been documented for determinate soybean grown on low N sandy soils. The purposes of this study were to assess the impact of conservation tillage and *R. japonicum* inoculation on (i) nodular occupancy and nodulation patterns of introduced and indigenous *R. japonicum* strains, (ii) total N accumulation, N_2 fixation, and the net N returned to the soil, and (iii) soybean seed yield.

MATERIALS AND METHODS

A split-split plot design with tillage as the main plot was used. Soybean cultivars were the subplots, and inoculation with *R. japonicum* was randomized within each subplot with three or four replications in 1980 and 1981, respectively. The soil was a Norfolk loamy sand (Typic Paleudult), and fertilization for both years consisted of 28, 24, 46, and 56 kg ha^{-1} of N, P, K, and S, respectively. Dolomitic lime was applied both years at a rate of 1120 kg ha^{-1}. Tillage treatments consisted of conventional vs. conservation tillage. The conventional treatment was disked before planting, but the conservation tillage treatments had no inversion of the soil surface before planting. Planting was accomplished by the

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use of a Brown and Harden Superseeder.[3] The "superseeder" has a cutting colter and a subsoil shank that precedes the planter. Thus, this form of conservation tillage allowed in-row subsoiling. Winter weeds were the vegetative cover for both tillage systems. The conservation tillage system was sprayed with paraquat (1,1′-dimethyl-4,4′-bipyridinium) during the planting operation in order to kill the surface vegetation. Other pesticides were applied at recommended rates for conventional soybean production. Four soybean cultivars ('Lee' and 'Lee-nonnodulating', group VI; 'Ransom', group VII; and 'Coker 338', group VIII) were planted on 29 May 1980 and 14 May 1981. Split plots had eight 12-m rows on 96-cm spacings in 1980 and eight 14-m rows on 76-cm spacings in 1981 for each cultivar. Rows were oriented northwest by southeast in both years. Half of each split plot was inoculated with strain 3I1b 110 of *R. japonicum* at a rate of 10^8 organisms per centimeter of row. The inoculum was in an aqueous form and was applied directly to the furrow and seed at planting (Weaver and Frederick, 1974). The indigeneous population was 10^3 rhizobia per gram of soil. Conventionally tilled soybean plots were cultivated when necessary for weed control, but conservation tillage plots received no cultivation.

Plant populations for Lee, Lee non-nod, Ransom, and Coker 338 were 26, 30, 25, and 27 plants m^{-2}, respectively, in 1980; and 20, 17, 33, and 25 plants m^{-2}, respectively, in 1981. Plant tops and roots were sampled from 30 cm of row during the pod development stage (approximately R4). Sampled plant tops were dried at 70 °C, weighed, ground to pass a 0.4-mm screen, digested with 3 mL of 30% H_2O_2 and 7 mL of H_2SO_4, and analyzed for total Kjeldahl N (TKN) on a Technicon Auto Analyzer using industrial method 334-74 W/B (Technicon Industrial Systems, 1977). Total shoot N is hereafter referred to as total N. Roots were assayed for N_2 fixation by the acetylene (C_2H_2)-reduction method (Hardy et al., 1968). Ethylene (C_2H_4) concentrations were determined on a Varian 940 gas chromatograph. Roots with attached nodules were placed in self-closing plastic bags and frozen until the nodules could be picked, cleaned, counted, and weighed. All nodule weights are reported as fresh weights. Soil samples were taken from the Ap, E, and B horizons at approximately the R9 growth stage of soybean in 1981. They were air-dried, ground, digested, and analyzed for TKN. Rainfall was measured with a recording rain gauge, and soil water was measured with tensiometers at 30- and 120-cm depths.

Nodular occupancy of *R. japonicum* strains was determined by the microagglutination test on nodule suspensions with antisera against USDA strains 3I1B24, 3I1b31, 3I1b46, 3I1b76, 3I1b94, 3I1b110, 3I1b122, 3I1b125, Brazil 587, and North Carolina 1004. The 3I1b, Brazil, and North Carolina prefixes will not be used in the remainder of the text. Twenty-four nodules from the root system of each plot were analyzed as described by Hunt et al. (1981). Due to cross agglutination reactions between strains 31 and 587 and between strains 122 and 1004, these data were combined and presented as 31 and 122, respectively.

Nodular occupancy of rhizobia was statistically analyzed on individual strains by using square root transformation, analysis of variance, and least significant difference (LSD) as outlined by Steel and Torrie (1960). Strains were analyzed separately because of nonhomogenous variances. Other parameters were also analyzed on yearly basis by analysis of variance and LSD for planned comparisons.

RESULTS AND DISCUSSION

Rainfall Patterns

Rainfall patterns for 1980 and 1981 were quite different (Fig. 1). In 1980 only 19.4 cm of rain fell during

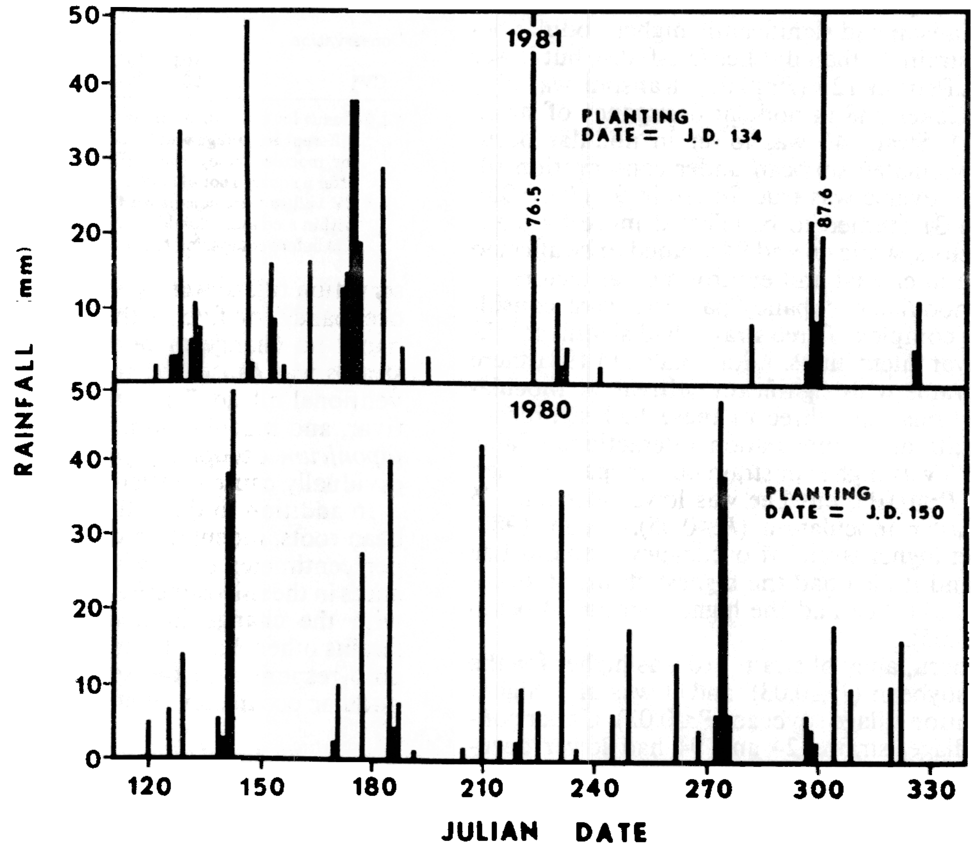

Fig. 1. Rainfall distribution during the 1980 and 1981 soybean-growing season.

the podfill period, and soil matric potential at the 30-cm depth exceeded −100 kPa during the first 40 days of pod development. In 1981, 31.5 cm of rainfall provided ample water prior to pod set, but the distribution of 13.3 cm of water during podset constituted a mild, late-season drought. Prior to this time, the soil matric potential did not exceed −100 kPa at the 30-cm depth.

Rhizobium japonicum Nodular Occupancy

In 1980 and 1981, nodules formed by unidentified strains were 53 and 42%, respectively. The large number of unidentified strains in this and in most other studies conducted in the southeastern USA is a deterrent to a clear understanding of *R. japonicum* nodular occupancy patterns and the response of soybean growth and yield (Annual Reports of Southern Regional Projects S-112 and S-170, A. G. Wollum II, Chairman). Yet, certain trends can clearly be seen. Tillage affected the nodular occupancy of strain 125 in 1980 and strain 110 in 1981 (*F* significant at 99 and 97% levels, respectively), but tillage alone did not have a major effect on nodular occupancy. Similarly, inoculation alone was not a major factor in nodule occupancy, but inoculation interacted at *F* significance levels > 90% with tillage for strains 46 and 94 in 1980 and 24, 31, and 94 in 1981. Cultivar was an important factor for nodular occupancy in 1980, while the interaction of cultivar and inoculation seemed to be important in 1981. Strains 24 and 94 had higher nodular occupancy rates in 1981 than in 1980 (Table 1). This result would be expected based on reports that these strains are particularly susceptible to moisture stress (Mahler and Wollum, 1980, 1981).

In 1980 Ranson had significantly higher nodular occupancy of strain 31 than did Lee ($P \leq 0.05$), but lower occupancy of strain 125 ($P \leq 0.01$). Ransom was also lower than Coker 338 in nodular occupancy of strain 94 ($P \leq 0.01$). Strain 46 was lower in nodular occupancy for inoculated soybean under conservation tillage, but the reverse was true for strain 94 ($P \leq 0.05$). Thus, strain 31 seemed to be affected more by plant selection factors, while 46 and 94 seemed to be affected more by the microbial and environmental factors.

In 1981 nodular occupancy patterns were considerably more complex. Three strains had significant tillage × cultivar interactions. Additionally, in 1981 there were five strains with significant cultivar × inoculation interactions, and three of these had significant tillage × cultivar × inoculation interactions (Table 1). Coker 338 was higher in strain 46 occupancy when inoculated ($P \leq 0.01$), but Lee was lower in strain 76 occupancy after inoculation ($P \leq 0.05$). As in 1980, Ransom had higher strain 31 occupancy than did Lee ($P \leq 0.05$), and it also had the highest strain 46 occupancy ($P \leq 0.01$). Lee had the highest strain 24 occupancy ($P \leq 0.05$).

Nodular occupancy of strain 110 was higher for the inoculated soybean ($P \leq 0.03$), and it was also higher for conservation tillage soybean ($P \leq 0.03$). Under conservation tillage, strains 24 and 94 had lower occupancy for inoculated soybean ($P \leq 0.05$). Strain 31 had increased occupancy in inoculated soybean conventional tillage ($P \leq 0.05$).

Inoculated and noninoculated Coker 338 under conservation tillage were significantly different in nodular occupancy for five of the eight strains. On the other hand, no change in nodular occupancy of the eight strains was caused by inoculation of Lee under conventional tillage. Thus, it is apparent that tillage, cultivar, and inoculation interacted to cause different *R. japonicum* occupancy patterns, but they did not individually cause consistent major effects.

Table 1. Nodular occupancy by eight *Rhizobium japonicum* strains as affected by soybean cultivar, tillage, and inoculation with strain 110 of *R. japonicum*.

Tillage	Inoculation	24	31	46	76	94	110	122	125
		%							
1980									
		Lee							
Conventional		3	7	4	3	3	7	8	2
		2	11	1	8	5†	4	8	0
Conservation		1	6	1	8	10‡	5	3	6†
		1	13	5	7	0†‡	0	6	1†
		Ransom							
Conventional		1	22	4	7	0	12	6	0
		0	16	2†	7	0	9	4	0
Conservation		3	21	4	6	0	7	5‡	0
		0	28	9†	4	0	3	0‡	0
		Coker							
Conventional		1	10	8	12	9	3	4	2
		0	19	6	8	14	4	4	0
Conservation		0	13	3	11	11	2	11	1
		0	16	9	7	10	2	4	1
CV¶		77	35	39	31	44	56	48	59
1981									
		Lee							
Conventional		9	8	6†	1‡	9‡	2	13	0
		13	6	5	4‡	20‡	0	8	0
Conservation		11	4	0†‡	0§	20	4‡	6	0
		20	2	4‡	4§	10	0‡	7	0
		Ransom							
Conventional	+	7	14‡	6	1†‡	14	2	3†‡	0
	−	5	5‡	7	5‡	26	0	11‡	0
Conservation	+	9	14	6	6†	11	0‡	8†	0
	−	12	8	6	6	12	3‡	9	0
		Coker							
Conventional	+	13†	7†	6‡	3	13	1†	11	1
	−	15	6†	1‡	2	18	0	6	0
Conservation	+	15	6†	7§	8‡	26	6†‡	10	0
	−	10‡	15†‡	1§	3‡	16	2‡	8	0
CV¶		36	30	41	44	31	60	28	29

†,‡,§ Means for the same year, serogroup, and cultivar followed by an † are different for tillage and those means followed by a ‡ or § are different for inoculation by the LSD test at the 0.10 level when analyzed after a square root of (mean + 0.5) transformation.

¶ CV values were calculated from transformed data; therefore, means within a column should be transformed by a square root of (mean + 0.5) before comparing to CV values.

In addition to the actual nodular occupancy of soybean roots, inoculation of 10^8 organisms of strain 110 per centimeter of row probably caused some differences in the rhizosphere microbiology. This effect, along with the change in nodular occupancy patterns of strains other than 110, may help to explain significant crop responses to the seemingly small differences in nodular occupancy of strain 110.

Nodulation and Acetylene Reduction

Nodule number and weight were greater in 1981 than in 1980 (Table 2). Although the yearly effect predominated, some responses to tillage and inoculation

Table 2. Nodulation and N_2 fixation (acetylene-reducing activity) as affected by soybean cultivar, tillage, and inoculation with strain 110 of *R. japonicum*.

Cultivar	Inoculation	Conserv.†	Conv.†	Conserv.†	Conv.†	Conserv.†	Conv.†	Conserv.†	Conv.†
		Nodules plant⁻¹‡				Nodule weight plant⁻¹ (g)			
		1980		1981		1980		1981	
Coker 338		34	51	98	112	0.32	0.55	1.80	1.46
		56	47	183	117	0.72	0.50	3.86	2.27
Ransom		54	30	101	111	0.42	0.19	1.14	1.43
		53	39	92	104	0.40	0.23	1.05	1.54
Lee		30	55	73	138	0.27	0.58	0.87	2.10
		43	38	118	93	0.52	0.67	1.47	1.18
LSD (0.10)		21		55		0.53		1.11	
		Total nodule activity (μmol C_2H_4 h⁻¹ plant⁻¹)				Specific nodule activity (μmol C_2H_4 hr⁻¹ g⁻¹)			
		1980		1981		1980		1981	
Coker		7	3	19	15	113	5	11	10
		2	1	28	23	2	2	7	12
Ransom		1	7	5	12	2	82	4	9
		2	2	8	9	8	7	7	9
Lee		1	1	7	11	4	2	8	6
		1		5	9	5	1	3	
LSD (0.10)		3.8		11.6		90.4			

† Conserv. = conservational and Conv. = conventional tillage.
‡ LSD comparisons are for the same year (2.8 and 17.0 are to be used to compare inoculation and tillage, respectively, for specific nodule activity in 1981).

Table 3. Total N accumulated by soybean grown on a Norfolk loamy sand with different tillage and *R. japonicum* inoculation.

Tillage	Inoculation	Coker	Ransom	Lee	Lee NN
		Cultivar			
		g plant⁻¹			
1980					
Conservation		0.66	0.49	0.53	
		0.46	0.60	0.66	0.22
Conventional		0.88	0.45	0.68	–
		0.49	0.57	0.65	0.33
LSD (0.10) 0.34					
(0.05) 0.42					
1981					
Conservation		1.97	1.58	1.57	
		1.86	1.71	1.65	0.63
Conventional		2.35	1.13	1.65	–
		1.78	1.17	1.43	0.58
LSD (0.10) 0.36					
(0.05) 0.44					

Table 4. Yield for soybean grown on a Norfolk loamy sand with different tillage and *R. japonicum* inoculation.

Tillage	Inoculation	Coker	Ransom	Lee	Lee NN
		Cultivar			
		Mg ha⁻¹			
1980					
Conservation		0.80	1.02	1.04	
		0.77	0.92	1.16	0.56
Conventional		0.66	0.94	0.83	–
		0.78	0.89	0.86	0.49
LSD (0.10) 0.26					
1981					
Conservation		1.52	2.00	2.20	
		1.71	1.80	1.59	1.67
Conventional		1.78	1.94	2.05	
		1.91	2.01	2.04	1.36
LSD (0.05) 0.35					

occurred. Lee soybean, when inoculated, had a higher number of nodules under conventional than conservation tillage in both years. Nodule weights were also numerically higher in 1980 and statistically higher in 1981, but these differences were not expressed in either total or specific C_2H_2–reduction activity. However, noninoculated Lee grown under conservation tillage had the lowest specific C_2H_2–reduction activity and the lowest seed yield for that cultivar in 1981. Ransom had no consistent differences between inoculated and tillage treatments for nodule numbers or weights in either year. Inoculated Ransom under conservation tillage had the lowest total and specific activity for this cultivar in both years, but this was not expressed in either total N per plant (Table 3) or seed yield (Table 4). Coker 338 soybean had no differences between treatments for nodule number or weight in 1980. In 1981, inoculated plants had fewer nodules, lower weights, and higher specific C_2H_2–reduction activity than the noninoculated plants under conservation tillage. Specific C_2H_2– reduction activity for Coker 338 under conservation tillage was increased by inoculation in both years. These differences in Coker 338 were expressed in total N accumulation, but not in yield. This was probably related to the fact that Coker 338 is a group VIII soybean that can accumulate large amounts ($\geq$ 6 MG ha^{-1}) of dry matter before pod set (Karlen et al., 1982)

Total Shoot Nitrogen Accumulation

No significant treatment effects for inoculation, cultivars, or tillage for total N per plant occurred in 1980 (Table 3), but a significant ($P \leq 0.03$) inoculation by cultivar interaction existed. Inoculated Coker 338 soybean plants contained more total N than noninoculated plants. Inoculated, Coker 338 was also higher in total N accumulation than Ransom or Lee under either tillage treatment, but the reverse was true for noninoculated Coker.

Based on the differences between Lee and Lee nonnod in 1980, 58 to 67% of the N accumulated by Lee soybean was supplied by N_2 fixation under conservation tillage, while 49 to 51% was supplied by N_2

fixation under conventional tillage. Vasilas and Ham (1984) found the difference method and isotope dilution method to be very similar for estimation of soybean N_2 fixation. These differences between tillage treatments would be expected as a result of increased immobilization and slower mineralization of N in conservation-tilled soil compared with conventionally tilled soil. Similarly, a generally low accumulation per plant would be expected from the very dry soil conditions of 1980.

Total N accumulations by cultivars were significantly different ($P \leq 0.01$) in 1981, but differences due to tillage or inoculation were not significant. Coker 338 was numerically higher than Lee or Ransom for all inoculation and tillage combinations, but only with inoculation were the differences clearly significant. As in 1980, the inoculated, conventionally tilled Coker 338 had the highest total N accumulation.

Dinitrogen fixation in 1981 was estimated to supply 60 to 62% of the total N for Lee soybean under conservation tillage, and it was estimated to supply 59 to 65% under conventional tillage. Thus, tillage did not significantly impact the percentage of N supplied by N_2 fixation in 1981. Total and fixed N, however, were both much higher in 1981 than in 1980.

Yield

In the dry year, 1980, soybean yielded an average of 0.89 Mg ha^{-1}, and in the more plentiful rainfall year, 1981, soybean yielded an average of 1.88 Mg ha^{-1} (Table 4). Yields for these years were different at the $P \leq 0.01$ level. The 2-y mean yield for conventional tillage, 1.47 Mg ha^{-1}, was not significantly different from the conservation tillage mean of 1.44 Mg ha^{-1}. These findings are generally consistent with the results reported by Campbell et al. (1984) for soybean grown on other treatments and soil in the South Carolina Coastal Plain. However, yields for conservation tillage were higher by 0.12 Mg ha^{-1} in 1980 and lower by 0.15 Mg ha^{-1} in 1981, when compared with conventional tillage yields ($P \leq 0.1$). Yield of the Lee nonnodulating soybean were higher under conservation than conventional tillage in both 1980 and 1981. This was probably a result of conservation of moisture by the residue and a lower consumptive use of water from the smaller conservation tillage plants in 1980.

Two-year mean yields were not significantly affected by inoculation; yields were 1.48 and 1.47 Mg ha^{-1} for the noninoculated and inoculated treatments, respectively. The mean yield for Coker 338 was significantly lower ($P \leq 0.05$) than that of Lee or Ransom. This lower yield can be partially explained by the fact that Coker 338, a group VIII soybean, needed water longer into the growing season to support pod set and seed development. The lack of seed yield response to higher N accumulation in soybean was consistent with previous reports (Hunt et al., 1981; Morris and Weaver, 1983). A lack of correlation between total accumulated N in the soybean plant and seed yield makes generalizations about the net N returned to the soil difficult.

The rainfall pattern in 1981 allowed for the expression of more subtle treatment differences in yield. For instance, with conservation tillage, inoculation increased the yield of Lee soybean by 0.61 Mg ha^{-1}. This yield difference could have been caused by the direct effect of superior N_2 fixing performance of strain 110 relative to the indigenous population under conservation tillage. However, N_2 fixation data do not support this hypothesis, and the hypothesis is also weakened by the fact that nodule occupancy by strain 110 was not greatly different for the inoculated and noninoculated Lee soybean, 4 and 0%, respectively. The response of yield to inoculation may have been due to a shift in the nodular occupancy pattern of other strains; the inoculated Lee under conservation tillage had 10% more occupancy of strain 94 as well as 9, 4, and 4% less occupancy of strains 24, 46, and 76; respectively.

Nitrogen Returned to the Soil

One can estimate the net N returned to the soil from the shoot after removal of the seed from Lee soybean by use of Eq. [1].

Net shoot N returned to the soil,

$$\text{kg ha}^{-1} = A - B - C, \qquad [1]$$

where

A = total N [(kg plant N^{-1}) (plant population $m^{-2} \times 10^4$)]

B = soil supplied N, [(kg plant N^{-1}, Lee non-nod)(plant population $m^{-2} \times 10^4$)]

C = seed N, [seed yield (kg ha^{-1}) $\times$ (0.065)]

Plant densities for Lee and Lee non-nod were 26 and 30 m^{-2} in 1980 and 20 and 17 m^{-2} in 1981. When mean data for Lee soybean are used, Eq. [1] provides estimated ranges of 14 to 40 kg ha^{-1} in 1980 and 57 to 123 kg ha^{-1} in 1981 for net shoot N returned to the soil. The 123 kg ha^{-1} was caused by the low yield of the noninoculated, conservation-tilled Lee soybean. Major differences in net shoot N return to the soil were associated with seasonal and rainfall pattern rather than tillage or inoculation. Coker soybean, group VIII, had higher shoot N accumulation per plant, higher plant density, and a lower seed yield than Lee. This would likely produce a much greater net N return to the soil.

Values of soil N concentrations were obtained from samples composited across inoculation and cultivar treatments. The total profile N content was not different for tillage treatment, but distribution was significantly different. At the 0- to 5-cm depth, conservation-tilled soil contained over 100 $\mu g\ g^{-1}$ more N than the conventionally tilled soil (503 vs. 396 $\mu g\ g^{-1}$). Other workers have also reported a higher concentration of N as well as a lower pH in the surface of conservation tillage soils (Blevins et al., 1977, 1978). This could have deleterious effects on herbicide effectiveness and crop production. Soil pH values in the upper 10 cm of soil in the spring of 1982 were 5.7 and 5.3 for conventional and conservation tillage, respectively. These reductions in pH could probably be controlled by an adjustment of annual liming rates.

CONCLUSIONS

1. Seasonal differences, presumably due to rainfall, affected nodule occupancy by *R. japonicum* strains, N accumulation, and yield of determinate soybean under either conventional or conservation tillage on a Norfolk loamy sand.

2. Individually, tillage, cultivar, and inoculation with strain 110 *R. japonicum* did not greatly change nodular occupancy by indigeneous strains, N accumulation, or yield. However, specific combinations of tillage, inoculation, and soybean cultivar affected all three parameters.

3. Estimates of net shoot N returned to the soil were positive for 1980 and 1981 under both tillage systems with or without inoculation with strain 110 of *R. japonicum*.

4. More research is needed to improve our understanding of N accumulation and yield in soybean as well as N management in tillage systems for sandy, low N soils of the Southeast.

REFERENCES

Bennett, O.L., E.L. Mathias, and P.E. Lundberg. 1973. Crop responses to no-till management practices on hilly terrain. Agron. J. 65:488–491.

Bezdicek, D.F., D.W. Evans, B. Abede, and R.W. Witters. 1978. Evaluation of peat and granular inoculum for soybean yield and N fixation under irrigation. Agron. J. 70:865–868.

———, R.F. Mulford, and B.H. Magee. 1974. Influence of organic nitrogen on soil nitrogen, nodulation, nitrogen fixation, and yield of soybeans. Soil Sci. Soc. Am. Proc. 38:268–273.

Bhangoo, M.S., and D.J. Albritton. 1976. Nodulating and non-nodulating Lee soybean isolines response to applied nitrogen. Agron. J. 68:642–645.

Blevins, R.L., L.W. Murdock, and G.W. Thomas. 1978. Effect of lime application on no-tillage and conventionally tilled corn. Agron. J. 70:322–326.

———, G.W. Thomas, and P.L. Cornelius. 1977. Influence of no tillage and nitrogen fertilization on certain soil properties after 5 years of continuous corn. Agron. J. 69:383–386.

———, ———, M.S. Smith, W.W. Frye, and P.L. Cornelius. 1983. Changes in soil properties after 10 years continuous non-tilled and conventionally tilled corn. Soil Tillage Res. 3:135–146.

Campbell, R.B., R.E. Sojka, and D.L. Karlen. 1984. Conservation tillage for production of soybean in the U.S. Southeastern Coastal Plain. Soil Tillage Res. 4:531–541.

Christensen, L.A., and R.S. Magleby. 1983. Conservation tillage use. J. Soil Water Conserv. 38:156–157.

Doran, J.W. 1980a. Microbial changes associated with residue management with reduced tillage. Soil Sci. Soc. Am. J. 44:518–524.

———. 1980b. Soil microbial and biochemical changes associated with reduced tillage. Soil Sci. Soc. Am. J. 44:765–771.

Hardy, R.W.F., R.D. Holsten, E.K. Jackson, and R.C. Burns. 1968. The acetylene-ethylene assay for N_2 fixation: Laboratory and field evaluation. Plant Physiol. 43:1185–1207.

Hunt, P.G., A.G. Wollum II, and T.A. Matheny. 1981. Effects of soil water on *Rhizobium japonicum* infection, nitrogen accumulation, and yield in Bragg soybeans. Agron. J. 73:501–505.

———, T.A. Matheny, D.C. Reicosky, A.G. Wollum II, R.E. Sojka, and R.B. Campbell. 1983. Effect of irrigation and *Rhizobium japonicum* strain 110 upon yield and nitrogen accumulation and distribution of determinate soybeans. Commun. Soil Sci. Plant Anal. 14:223–238.

Johnson, J.W., L.F. Welch, and L.T. Kurtz. 1974. Soybeans' role in nitrogen balance. Illinois Res., Univ. of Ill., Agric. Exp. Sta. 16(3):6–7.

———, ———, and ———. 1975. Environmental implications of N fixation by soybeans. J. Environ. Qual. 4:303–306.

Karlen, D.L., P.G. Hunt, and T.A. Matheny. 1982. Accumulation and distribution of K, Ca, and Mg by selected determinate soybean cultivars grown with and without irrigation. Agron. J. 74:347–354.

Keyser, H.H., and D.N. Munns. 1979a. Effects of calcium, manganese, and aluminum on growth of rhizobia in acid media. Soil Sci. Soc. Am. J. 43:500–503.

———, and ———. 1979b. Tolerance of rhizobia to acidity, aluminum, and phosphate. Soil Sci. Soc. Am. J. 43:519–523.

Kohl, D.H., G. Shearer, and J.E. Harper. 1980. Estimates of N_2 fixation based on differences in the natural abundance of ^{15}N in nodulating and non-nodulating isolines of soybeans. Plant Phyiol. 66:61–65.

Lindemann, W.C., and G.E. Ham. 1979. Soybean plant growth, nodulation, and nitrogen fixation as affected by root temperature. Soil Sci. Soc. Am. J. 43:1134–1137.

———, G.W. Randall, and G.E. Ham. 1982. Tillage effects on soybean nodulation, N_2 (C_2H_4) fixation, and seed yield. Agron. J. 74:1067–1070.

Mahler, R.L., and A.G. Wollum II. 1980. Influence of water potential on the survival of rhizobia in a Goldsboro loamy sand. Soil Sci. Soc. Am. J. 44:988–992.

———, and ———. 1981. The influence of soil water potential and soil texture on the survival of *Rhizobium japonicum* and *Rhizobium leguminosarum* isolates in the soil. Soil Sci. Soc. Am. J. 45:761–766.

Matheny, T.A. and P.G. Hunt. 1983. Effects of irrigation on accumulation of soil and symbiotically fixed N by soybean grown on a Norfolk loamy sand. Agron. J. 75:719–722.

Morris, D.R., and R.W. Weaver. 1983. Mobilization of ^{15}N from soybean leaves as influenced by rhizobial strains. Crop Sci. 23:1111–1114.

Munevar, F., and A.G. Wollum II. 1981. Effect of high root temperature and Rhizobium strain on nodulation, nitrogen fixation, and growth of soybeans. Soil Sci. Soc. Am. J. 45:1113–1120.

Patterson, T.G., and T.A. LaRue. 1983. Nitrogen fixation by soybean: Seasonal and cultivar effects and comparisons of estimates. Crop Sci. 23:488–492.

Steel, R.G.D., and J.H. Torrie. 1960. Principles and procedures of statistics. McGraw-Hill Book Co., Inc., New York.

Technicon Industrial Systems. 1977. Industrial method no. 334-74 W/B$^+$. Individual/simultaneous determination of N and/or P in BD acid digest. p. 1–7. Technicon Industrial Systems, Tarrytown, NY.

Tyler, D.C., J.R. Overton, and A.Y. Chambers. 1983. Tillage effects on soil properties, disease, cyst nematodes, and soybean yields. J. Soil Water Conser. 38:374–376.

Vasilas, B.L., and G.E. Ham. 1984. Nitrogen fixation in soybean: An evaluation of measurement techniques. Agron. J. 76:759–764.

Weaver, R.W., and L.R. Frederick. 1974. Effect of inoculum rate on competitive nodulation of *Glycine max* L. Merrill. II. Field studies. Agron. J. 66:233–236.

Weber, C.R. 1966a. Nodulating and nonnodulating soybean isolines: I. Agronomic and chemical attributes. Agron. J. 58:43–46.

———. 1966b. Nodulating and nonnodulating soybean isolines: II. Response to applied nitrogen and modified soil conditions. Agron. J. 58:46–49.

Welch, L.F., L.V. Boone, C.G. Chambliss, A.T. Christiansen, D.L. Mulvaney, M.G. Oldham, and J.W. Pendleton. 1973. Soybean yields with direct and residual nitrogen fertilization. Agron. J. 65:547–550.

Williams, L.E., and D.A. Phillips. 1983. Increased soybean productivity with a *Rhizobium japonicum* mutant. Crop Sci. 23:246–250.