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EFFECT OF IRRIGATION AND RHIZOBIUM JAPONICUM STRAIN 110 UPON YIELD
AND NITROGEN ACCUMULATION AND DISTRIBUTION OF DETERMINATE SOYBEANS^{1/}

KEY WORDS: Leaf N concentration, leaf xylem potential, drought stress,
soil water, Norfolk loamy sand

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ABSTRACT

In the southeastern Coastal Plains determinate soybean [Glycine max (L.) Merr.] is grown on soils with low water-holding capacities and N contents. Therefore, N fixation and management of water are important in soybean production. Several studies were conducted to increase the understanding of the interaction of these two factors on soybean growth and yield. Experiments were conducted in 1976, 1978, and 1979 on a Norfolk loamy sand (Typic Paleudult) with soybean of maturity groups VI, VII, and VIII grown under irrigated and nonirrigated conditions each year with soil water monitored to 60-cm depths. In 1979 the plots were split by inoculating with Rhizobium japonicum strain 110 at a rate of 10^8 organisms/cm of row. Serological analyses indicated that strain 110 infection of nodules was near 0% in the noninoculated plots and 7 to 21% in the inoculated plots. Irrigation water increased

leaf-N concentration and seed yield during the severe droughts of 1976 and 1978; yields were >3 and <1.5 t/ha for irrigated and nonirrigated soybean, respectively. In 1979 there was only a short-term drought which did not affect the total N accumulation, nor leaf-N content for the noninoculated soybean. However, the drought was associated with a significant reduction in leaf-N content of the inoculated soybean, 4.74 and 4.36%, irrigated and nonirrigated, respectively. Additionally, there was a significantly more negative leaf xylem potential for inoculated than for noninoculated soybean plants under drought. These differences, coupled with varying maturities, contributed to a significant variety x irrigation x R. japonicum strain 110 interaction for yield. These results show that water management affects soybean N accumulation, distribution, and yield and that R. japonicum strain 110 may intensify or diminish the impact of water regimes on determinate soybean growth and yield.

INTRODUCTION

In the southeastern United States soybean are frequently exposed to short-term drought and plant water stress due to the low water-holding capacity of soils and erratic rainfall patterns^{5,12,6}. The soils of this region are also generally low in N and are subject to leaching and denitrification of N. Consequently, determinate soybean grown in this area must fix larger amounts of N to support both vegetative growth and seed production than soybean grown in soils with higher N contents. Total accumulations of 112 to 450 kg/ha of N have been reported under nonirrigated conditions^{7,2}. Hunt et al.⁸ reported a total N accumulation of 450 kg/ha with 'Bragg' soybean under irrigated conditions on a Norfolk loamy sand. Boote et al.³ reported that the photosynthetic rates in soybean were correlated to leaf-N concentration. Leaf N is generally believed to decline during podfill. However, Nelson and Weaver¹³ suggest that soybean can fix and accumulate enough N to supply the needed N for seed while maintaining leaf-N content under good growing conditions. Drought is one of the conditions that will obviously affect photosynthesis and N fixation.

Hunt et al.⁸ reported that both total accumulation of N and the leaf-N concentration were affected by R. japonicum strain in 'Bragg' soybean. They reported that a R. japonicum strain 110 x soil-water status interaction existed. Introduction of strain 110 R. japonicum

into 10 to 25% of the nodules increased biomass and total N accumulations, as well as leaf-N concentration, under irrigated conditions. The reverse was true under nonirrigated conditions. This suggests two things for Bragg soybean (1) that in addition to soil-water status, the R. japonicum strains that occupy the soybean nodules must be known and considered when N accumulation and distribution are investigated and managed and (2) that "superior" strains may only exhibit superior fixation, growth, and yield for soybean under optimal environmental conditions and, in fact, may not perform as well as indigenous strains when environmental conditions are suboptimal.

The objectives of this research were: (1) to determine the N accumulation and distribution patterns of several selected determinate soybean varieties under irrigated and nonirrigated conditions in sandy Coastal Plain soils and (2) to more clearly define the effects of R. japonicum strain 110 upon N accumulation and distribution and yield of these soybean varieties under these soil-water conditions.

MATERIALS AND METHODS

Soybean [Glycine max. (L.) Merr.] was grown on a Norfolk loamy sand (Typic Paleudult) in 1976, 1978, and 1979. Soil nutrient status was determined by standard Georgia soil testing procedures⁹. Experimental designs varied between years and are discussed in chronological order.

A modified randomized block design with four replicates was used in 1976. Each plot consisted of four rows, 102 cm wide and 16.4 m long. The plots were prepared by applying 336 kg/ha of 0-20-20 fertilizer on 22 March followed by plowing to a depth of 25 cm, and 560 kg/ha of 8-24-24 fertilizer was applied and disked in on 6 May. Treflan¹ (Trifluralin) was incorporated into the soil at the rate of 2.3 liter/ha prior to planting. Four varieties, 'Ransom', 'Coker 136', 'FFR666', and 'Bragg', were planted on 21 May 1976 (Julian day 142). Water was applied through Bi-wall trickle irrigation tubing when the soil matric potential reached -0.2 bar at the 15-cm depth. Tensiometers were installed in groups in irrigated and nonirrigated plots of Ransom and Coker 136 varieties at the 15-, 30-, 46-, 61-, 91-, 122-, 153-, and 175-cm depths. Plant sampling consisted of collecting 30 of the uppermost trifoliates from each plot starting on 9 July and continue on 7-to 10-day intervals until harvest. Samples were separated

into leaflets and petioles, dried at 70 C, and ground to pass a 1 mm sieve. Total Kjeldahl N (TKN) was determined by the Macro-Kjeldahl method⁴.

In 1978 a completely randomized block design with 3 replicates was used. Each plot consisted of six rows, 96 cm wide and 10.7 m long. Fertilizer application consisted of 220 kg/ha of 0-14-22, and Treflan was applied at a rate of 0.3 l/ha prior to disking. Three varieties, Ransom and Bragg (maturity group VII) and Coker 338 (maturity group VIII), were planted on 12 June (Julian day 163). Precipitation was recorded by a rain gauge. Irrigation was applied by Bi-wall trickle tubing when the soil water matric potential reached -0.25 bar at the 60-cm depth. Vacuum gauge tensiometers, 'Soil Moisture', were installed at 30-, 60-, 90-, 120-, and 150-cm depths.

In 1979 a completely randomized split plot design was used. Each whole plot was the same size as in 1978, but the row length was divided in half by an inoculation split. Inoculation was with strain IB110 R. japonicum at a rate of 10^8 organisms/cm of row applied directly to the seed in a liquid form. Rhizobium japonicum was supplied in a frozen concentrate by Ag Labs, Columbus, Ohio. Fertilizer application consisted of 202 kg/ha of 0-20-20. Treflan application, soil water tensiometer measurements, rainfall measurements, and trickle tube irrigation were the same as in 1978. Four varieties; Lee (maturity group VI), Ransom and Bragg (maturity group VII), and Coker 338 (maturity group VIII), were planted on 24 May 1979 (Julian day 144). Xylem potentials for the uppermost, fully expanded trifoliolate were measured on days 80 and 81 after planting in 1979 at presunrise and midday times using a pressure chamber²⁰.

Whole plant samples were collected 46, 72, 92, and 114 days after planting in 1978 and 39, 61, 74, 91, 105, 123, and 140 days after planting in 1979. Plant samples were randomly selected from 30 cm of each plot and separated into leaves, stems, petioles, and pods. Samples were washed, dried at 70 C, and ground to pass a 0.841 mm sieve in 1978 and a 0.500 mm screen in 1979. Total Kjeldahl N was determined by digesting 0.25 g of plant tissue with 7 ml of acid (97 g H_2SeO_3 + 4.04 L of H_2SO_4) and 3 ml of 30% H_2O_2 at 400 C for one hour. The digest was diluted to 75 ml and analyzed on a Technicon Auto-Analyzer II. Nodule samples were obtained from a 25-cm diameter cylinder driven to a depth of 30 cm. Serological analyses were performed on 24 randomly selected

nodules as described by Hunt et al.⁸

Yields were measured from the four center rows of each plot, and the seeds were analyzed for oil and protein content in 1978 and 1979¹⁹.

RESULTS AND DISCUSSION

Environmental Conditions

Rainfall patterns for the three years of the study were different (Fig. 1). Both 1976 and 1978 had significant periods of drought. In both growing seasons, rainfall was adequate in most of the vegetative phases, but drought began during the late vegetative or flowering stages of growth depending upon maturity group. In 1976 the total rainfall was 501 mm, and the total irrigation was 359 mm; but only 101 mm of rainfall came between Julian day 190 and 290. In 1978 the total rainfall was 403 mm, but only 82 mm fell after Julian day 234. Total irrigated water was 202 mm. In 1979 there was only a short mid-season drought followed by an extended wet period. Precipitation was 185, 83, and 454 mm during the predominant vegetative, flowering, and pod fill growth stages, respectively. Only 244 mm of irrigation was required.

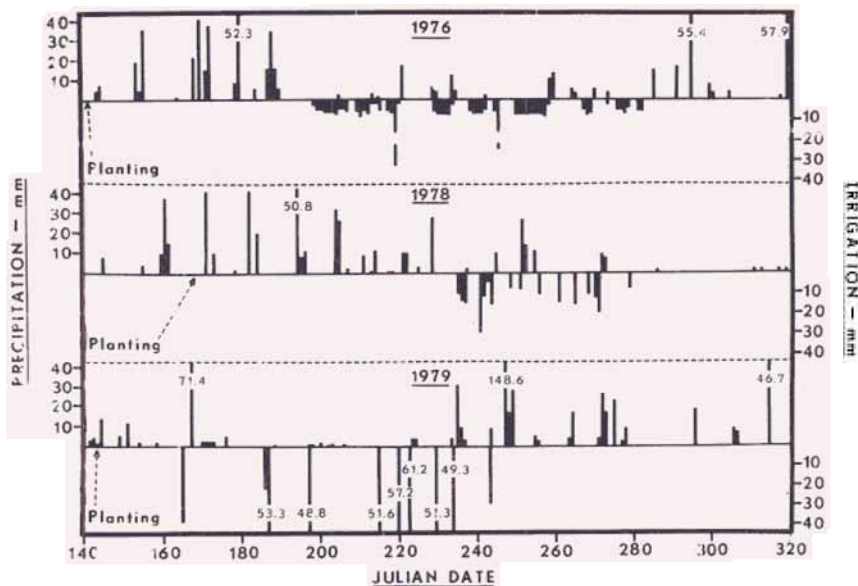


Fig. 1. Summary of precipitation and irrigation for the 1976, 1978, and 1979 growing seasons.

TABLE 1

Infection of Strain 110 R. japonicum for soybean under inoculated and irrigated treatments for 1979.

| Management | Variety | | | |
|--------------------------------|----------|---------|----------|-----------|
| | Lee* | Bragg | Ransom | Coker 338 |
| | -----% | | | |
| Nonirrigated- Noninoculated | 0 e | 0 e | 0 e | 4.2 d |
| Nonirrigated- Inoculated | 12.5 abc | 18.8 ab | 12.5 abc | 11.1 abc |
| Irrigated- Noninoculated | 0 e | 0 e | 0 e | 0 e |
| Irrigated- Inoculated | 11.3 abc | 6.9 dc | 8.3 bc | 20.8 a |

* Means followed by the same letter are not different at the 0.05 level by the Duncan Multiple Range Test.

for plant parts in each treatment in Figs. 2 and 3 are additive. The top line is the whole plant total N accumulated.)

The irrigation-induced differences were generally similar for the three varieties and were exhibited mostly in the leaves and pods. There was a linear slope of the three-variety mean for total N accumulation vs. time under irrigated conditions. However, there was an abrupt change in the slope of total N accumulation vs. time after drought stress in the nonirrigated treatment. Leaf-N accumulation was significantly higher ($P < 0.05$) in the irrigated than the nonirrigated treatment 92 days after planting. Pod N accumulation was higher for the irrigated treatment ($P < 0.05$) on both days 92 and 114 after planting. The petiole- and stem-N accumulations were not significantly different for varieties or irrigation on any date. Maximum whole plant total N accumulations were about 250 kg/ha; these somewhat low values were most likely due to the late planting date.

In 1979 the four-variety mean for whole plant total accumulated N was slightly higher for noninoculated than inoculated soybean under either irrigated or nonirrigated conditions (Fig. 3), but irrigation had no effect. By day 140 after planting, the pod N was about 200 and 150 kg/ha for irrigated and nonirrigated conditions, respectively,

1978

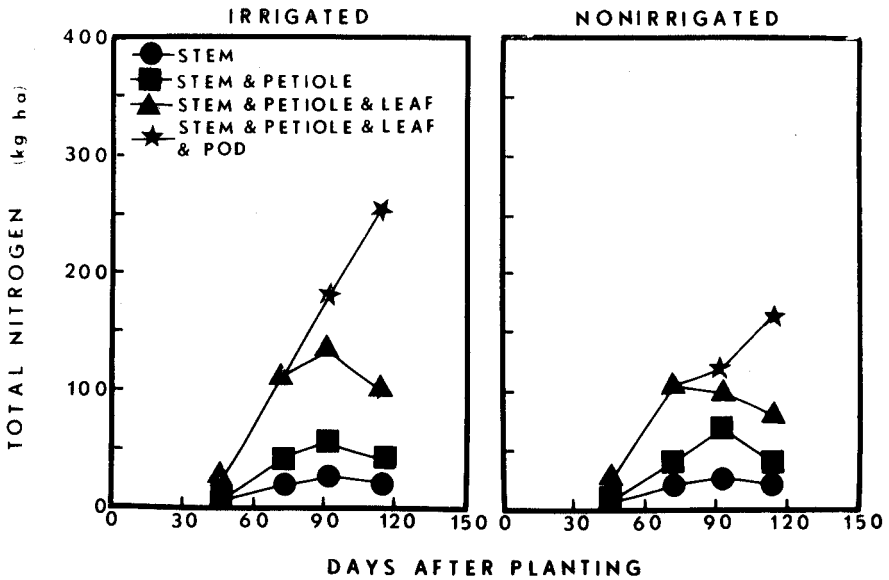


Fig. 2. Total N of irrigated and nonirrigated soybean in 1978.

regardless of inoculation. However, the leaf N declined earlier in the inoculated-irrigated treatments and resulted in its lower total N accumulation (<300 kg/ha). The Lee nonnodulating soybean were also grown in this experiment, and they accumulated 25 and 140 kg/ha of whole plant total N for irrigated and nonirrigated conditions, respectively, (Matheny and Hunt, unpublished data). Subtracting the total N accumulated by nonnodulating Lee from the mean of the N-fixing cultivars suggested that >250 kg of N came from biological N fixation under irrigated and <150 kg/ha of N came from biological N fixation under nonirrigated conditions.

Nitrogen Concentrations

The effect of soil water regime on leaf-N concentration also varied considerably each year (Tables 2, 3). The leaf-N concentrations in 1978 and 1979 were whole canopy means, while the 1976 values are upper trifoliolate means. In 1976 all varieties had significantly higher leaf-N concentration for irrigated than nonirrigated treatments. In 1978 Ransom had significantly higher leaf-N concentrations for irri-

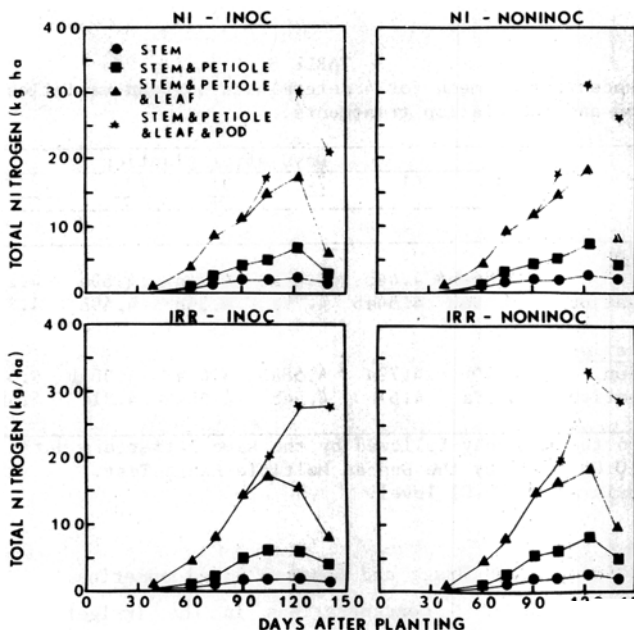


Fig. 3. Total N of irrigated and nonirrigated soybean with and without inoculation of strain 110 *R. japonicum*.

TABLE 2

Leaf-N concentration for various determinate soybean cultivars grown on a Norfolk loamy sand under irrigated and nonirrigated conditions.

| Year | Days after Planting | Cultivars | | | | | | | |
|------|---------------------|-----------|------|-------|------|-----------|------|-----------|------|
| | | Ransom | | Bragg | | FFFG66 | | Coker 136 | |
| | | I | NI* | I | NI | I | NI | I | NI |
| 1976 | 67 | 6.1a | 5.6b | 5.7a | 5.3b | 5.7a | 5.3a | 5.9a | 5.5a |
| | 82 | 5.2a | 4.9a | 5.0a | 4.5b | 5.1a | 4.6a | 5.1a | 4.8a |
| | 96 | 5.3a | 5.1b | 5.0a | 4.6b | 5.0a | 4.5b | 4.9a | 4.6b |
| | 110 | 4.8a | 4.4b | 4.5a | 4.0b | 4.5a | 3.7b | 4.3a | 3.7b |
| | 123 | 4.0a | 3.8a | 4.2a | 3.4b | 3.9a | 3.1b | 3.4a | 2.3b |
| | 137 | 3.7a | 2.9b | 3.2a | 2.3b | 3.0a | 2.5b | 2.4 | - |
| | | Ransom | | Bragg | | Coker 338 | | | |
| | | I | NI | I | NI | I | NI | I | NI |
| 1978 | 46 | 3.5a | 3.3a | 3.2a | 3.0a | 3.3a | 3.3a | | |
| | 72 | 4.8a | 5.3a | 4.1a | 4.6a | 4.5a | 4.7a | | |
| | 92 | 4.9a | 3.9b | 4.3a | 4.0a | 4.2a | 3.8a | | |
| | 114 | 4.0a | 3.3b | 3.5a | 3.1a | 3.5a | 3.4a | | |

* Means on the same day followed by the same letter are not different at the 0.1 level by the Duncan Multiple Range Test.

TABLE 3

Leaf-N concentration mean for 4 determinate soybean varieties with irrigation and inoculation treatments.

| Treatment | Days after planting | | | | | | |
|-----------------------|---------------------|--------|--------|--------|--------|-------|-------|
| | 39 | 61 | 74 | 91 | 105 | 123** | 140 |
| <hr/> | | | | | | | |
| <u>Inoculation</u> | | | | | | | |
| Irrigation | 4.56ab* | 4.44b | 4.67ab | 4.74a | 4.69a | 4.23b | 3.23a |
| Nonirrigation | 4.49ab | 4.54ab | 4.77a | 4.36b | 4.49b | 4.25b | 3.21a |
| <hr/> | | | | | | | |
| <u>Noninoculation</u> | | | | | | | |
| Irrigation | 4.40b | 4.72a | 4.58ab | 4.61a | 4.56ab | 4.52a | 3.27a |
| Nonirrigation | 4.67a | 4.51b | 4.54b | 4.56ab | 4.31c | 4.48a | 3.19a |

* Means on the same day followed by the same letter are not different at the 0.05 level by the Duncan Multiple Range Test.

**Day 123 is at the 0.01 level.

gated treatments, and Bragg and Coker 338 had numerically (though not significantly) higher N concentrations in the irrigated treatments. Differences for leaf-N concentrations in both years were large enough to have caused differences in effective photosynthesis according to Boote et al.³. Thus, the water stress could have affected both the fixation and photosynthetic efficiency.

In 1979 the drought did not reduce leaf-N concentration under noninoculated conditions until 105 days after planting (Table 3). However, the inoculated treatments showed sensitivity to drought earlier. Leaf-N values 91 days after planting were 4.74 and 4.36% vs. 4.61 and 4.56% for irrigated and nonirrigated treatments of inoculated vs. noninoculated soybean, respectively.

Rainfall after 105 days after planting in 1979 eliminated drought and caused some excess soil-water (Julian days 248-280). Net radiation was also low during this period (<100 ly/day). Under these environmental conditions, the leaf-N difference between irrigated and non irrigated treatments disappeared. However, the leaf-N concentration of inoculated soybean was lower on day 123 under both irrigated and non irrigated conditions. Additionally, petiole and stem-N concentration of inoculated soybean were also lower during this wet, low net radiation period (Tables 4, 5). These data again suggest that strain 110 functioned less efficiently than indigenous strains when the plants were subjected to unfavorable environmental conditions.

TABLE 4

Petiole N concentration of 4 determinate soybean varieties with irrigation and R. japonicum inoculation.

| Treatment | Days after planting | | | | | | |
|-----------------------|---------------------|--------|-------|-------|-------|-------|--------|
| | 39 | 61 | 74 | 91 | 105 | 123 | 140 |
| | -----% | | | | | | |
| <u>Inoculation</u> | | | | | | | |
| Irrigation | 1.81bc* | 1.71ab | 1.38c | 1.79a | 1.80a | 1.72b | 1.30b |
| Nonirrigation | 1.86b | 1.47b | 1.40c | 1.77a | 1.71a | 1.59c | 1.30b |
| <u>Noninoculation</u> | | | | | | | |
| Irrigation | 1.75c | 1.77a | 1.59b | 1.87a | 1.77a | 1.90a | 1.41a |
| Nonirrigation | 1.99a | 1.76a | 1.71a | 1.88a | 1.70a | 1.78b | 1.34ab |

* Means on the same date followed by the same letter are not different at the 0.1 level by the Duncan Multiple Range Test.

TABLE 5

Stem-N concentration of 4 determinate soybean varieties with irrigation and R. japonicum inoculation.

| Treatment | Days after planting | | | | | | |
|-----------------------|---------------------|-------|-------|-------|--------|-------|-------|
| | 39 | 61 | 74 | 91 | 105 | 123 | 140 |
| | -----% | | | | | | |
| <u>Inoculation</u> | | | | | | | |
| Irrigation | 1.83b* | 1.51a | 1.62a | 35b | 1.29bc | 1.27b | .85b |
| Nonirrigation | 1.89ab | 1.52a | 1.43a | 54a | 1.20c | 1.21b | .84b |
| <u>Noninoculation</u> | | | | | | | |
| Irrigation | 1.69c | 1.48a | 1.30a | 1.54a | 1.42a | 1.50a | 1.00a |
| Nonirrigation | 1.97a | 1.46a | 1.36a | 1.64a | 1.35ab | 1.43a | .95a |

* Means followed by the same letter are not different at the 0.1 level by the Duncan Multiple Range Test.

Predawn leaf xylem pressure potential (Ψ_x) mean for 80 and 81 days after planting in 1979 was also affected by inoculation with strain 110 (Fig. 4). At the peak of the drought, predawn Ψ_x values of nonirrigated, inoculated plants were more negative than those of noninoculated plants ($P < 0.05$), but at midday they were not statistically different. Under irrigated conditions, the opposite relationship existed. In early morning with no stress, there was no difference, but at midday the inoculated-irrigated treatment had less negative Ψ_x values ($P < 0.05$). This relationship was not the simple expression of a larger biomass

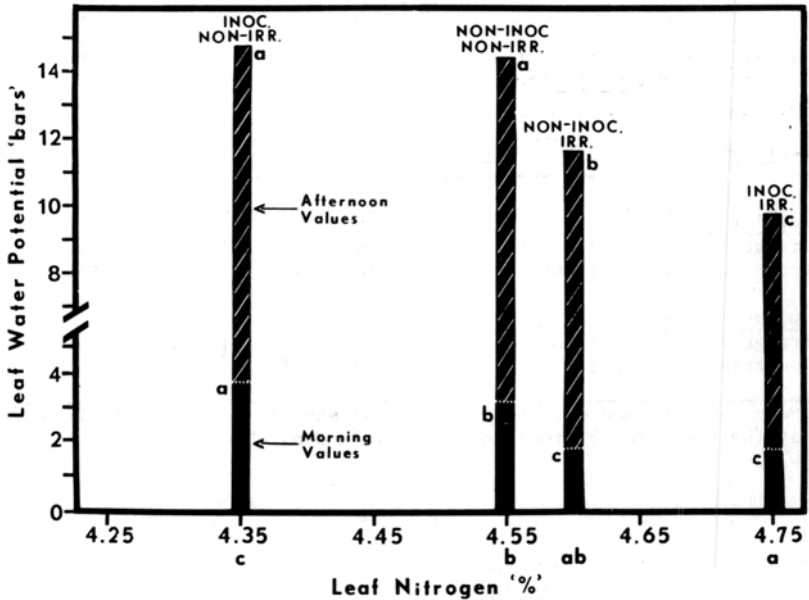


Fig. 4 Leaf water vs. leaf N (Values on a horizontal level followed by the same letter are not different at the 0.1 level by the Duncan's Multiple Range Test.)

associated with the inoculated treatments. As discussed earlier, the Lee variety had the smallest biomass with nonirrigated-inoculate conditions, and the other varieties were not significantly different for inoculation.

Leaf N has been shown to be related to water potential in cotton¹ 18,16,15. They have shown leaf N to affect water loss per unit change in water potential and water content at a given water potential. Additionally, they reported leaf-N content to affect stomatal sensitivity to water stress in cotton. Both N nutrition and water stress appear to interact to control abscisic acid accumulations and processes regulated by that accumulation. It is possible that a leaf N x water status relationship exists in soybean. However, subsequent research has failed to establish a clear and repeatable relationship of Ψ_x either leaf-N content or *R. japonicum* strain (Hunt, P. G., unpublished data).

Seed Yield

Yields for the three years of experimentation are reported

Table 6; they reflect the rainfall pattern, growth conditions, and N status of the seasons. In 1976 and 1978 the irrigated treatments averaged over 3 t/ha while the nonirrigated averaged less than 1.5 t/ha. In 1979 when only the short drought occurred, the irrigated and nonirrigated treatments yielded 2.0 and 1.6 t/ha, respectively, under noninoculated conditions. However, the variety x inoculation x irrigation interaction for yield was significant at the $P < 0.05$ level. Specifically, under noninoculated conditions, Lee yields were not increased by irrigation ($P < 0.1$), but Bragg, Coker 338, and Ransom yields were increased ($P < 0.1$, 0.05, and 0.01, respectively). Under inoculated conditions, Ransom, Coker, and Lee yields were increased by irrigation ($P < 0.05$, 0.05, and 0.01, respectively); but Bragg was unaffected ($P < 0.1$). The introduction of strain 110 caused a significant reduction in yield ($P < 0.1$) under nonirrigated conditions for the Lee variety. Neither oil nor protein percent was greatly different for any treatments.

The 1979 yields were related to the interaction of weather, R. japonicum strains, and maturity groups. Lee, (group VI) was farther into podfill when the drought of 1979 occurred and caused a reduction in leaf N and greater water stress for the inoculated-nonirrigated plants (Figs. 1 & 4, Table 3). For Lee, irrigation allowed a positive response to 110 infection while drought stress associated with the nonirrigated treatment caused a negative yield response to strain 110.

Ransom and Bragg (group VII) had much of their podfill period under excess moisture and low net radiation. Under these conditions, the infection of strain 110 was related to lower plant N concentrations.

Coker 338, (group VIII) had a period of good soil-water conditions and elevated solar radiation during its later podfill period, and its yields were increased by irrigation under both inoculated and non-inoculated conditions.

SUMMARY AND CONCLUSIONS

Weather patterns and water management have dramatic effects on the N accumulation and yield of soybean in the southeastern Coastal Plains. In seasons where drought was a significant factor, soybean leaf-N concentration was higher in the irrigated treatments. In 1978 total N accumulated throughout the growing period at a near constant rate in irrigated soybean. There was, however, a sharp reduction in the rate

TABLE 6

Yield of selected determinate soybean cultivars grown with and without irrigation on a Norfolk loamy sand.

| Year | Inoculation/ Water Management | Cultivar | | | | |
|------|-------------------------------------|--------------|---------------|------------------|------------------|------------------|
| | | t/ha | | | | |
| | | <u>Bragg</u> | <u>Ransom</u> | <u>Coker 338</u> | <u>FFR666</u> | <u>Coker 136</u> |
| 1976 | <u>Noninoculated</u> | | | | | |
| | Irrigated | 2.92a* | 3.19a | - | 3.31a | 2.98a |
| | Nonirrigated | 1.12b | 1.35b | | 1.02b | 1.04b |
| 1978 | <u>Noninoculated</u> | | | | | |
| | Irrigated | 3.07a | 3.52a | 3.07a | - | - |
| | Nonirrigated | 1.59b | 2.02b | 1.59b | - | - |
| | | <u>Lee</u> | <u>Bragg</u> | <u>Ransom</u> | <u>Coker 338</u> | |
| 1979 | <u>Inoculated</u> | | | | | |
| | Irrigated | 2.03a- | 1.82a | 2.11a | 1.78a | |
| | Nonirrigated | 1.37c+- | 1.72a | 1.80b | 1.46bc | |
| | <u>Noninoculated</u> | | | | | |
| | Irrigated | 1.80ab | 1.96a+ | 2.32a- | 1.73ab | |
| | Nonirrigated | 1.65bc+ | 1.67a+ | 1.74b- | 1.44c | |

*Means followed by the same letter are not different at the 0.05 level by the Duncan Multiple Range Test. Letters with a + indicate a difference at the 0.1 level and letters with a - indicate a difference at the 0.01 level.

of total N accumulation at the beginning of drought in the nonirrigated soybean. Mean yields during the two drought-stressed seasons of 1976 and 1978 were more than doubled by irrigation, <1.5 to >3.0 t/ha.

In 1979 there were short periods of both drought and excess soil water. Soybean yields were reduced by R. japonicum strain 110 infection when either environmental stress occurred during podfill. During the period of drought stress the mean leaf-N content of all varieties in the inoculated-nonirrigated treatment was significantly lower than any other treatment. Additionally, inoculated-nonirrigated treatments had lower xylem potentials. When soils were wet; leaf-, petiole-, and stem-N concentrations were lower in the inoculated than noninoculated soybean. These results demonstrate that R. japonicum strain 110 may intensify or diminish the impact of water deficits or excesses on N accumulation, N distribution, and yield of Lee, Bragg, Ransom and Coker 338 soybean.

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