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# MAXIMIZING NITROGEN FIXATION IN COMMON BEANS

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## ABBREVIATIONS AND DEFINITIONS

SARH - Secretaria de Agricultura of Recursos Hidraulicos (Secretary of Agricultura and Hydraulic Resources)

INIA - Instituto Nacional de Investigaciones Agricolas (National Institute of Agricultural Research)

CIAN - Centro de Investigaciones Agricolas del Norte (Agricultural Research Center of the North)

CIANOC - Centro de Investigaciones Agricolas del Norte Centro (Agricultural Research Center of the North Central)

CENAMAR - Centro Nacional de Metodos Avanzados de Riego (National Center for Advanced Irrigation Methods)

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SARH, INIA, CIAN, CIANOC.

## INTRODUCTION AND JUSTIFICATION

Dry beans (Phaseolus vulgaris) have traditionally been an important staple in Latin America and are the most important pulse in South America (1). Mexico has the third highest per capita daily supply of protein rich leguminous seeds in the world (2). Beans are the staple food of many Mexicans, providing energy as well as protein to millions of poor people.

In the northcentral plateau region of Mexico, little or no research has been performed on the nodulation of beans. No trained soil microbiologists or nitrogen fixation scientists are available. However, trained agronomists and researchers working in CENAMAR, INIA, and similar institutions could investigate nodulation and nitrogen fixation in beans if trained personnel could demonstrate the importance of nitrogen fixation and appropriate techniques for investigation.

Beans should obtain much of their nitrogen needs from nitrogen fixation. However, up to 100 kg/ha of nitrogen is commonly applied to beans on larger irrigated farms. On smaller, subsistence types of farms, little or no nitrogen is applied. Increased nitrogen fixation could increase bean yields on small farms and decrease nitrogen needs on larger farms.

Over five million hectares of beans are grown in the states of Chihuahua, Coahuila, Durango, and Zacatecas. Although the hectares in bean production are increasing in these states, Mexico has been importing beans from the U. S. for several years. There is a need for greater bean production and thus nitrogen fixation in the northcentral region of Mexico.

In keeping with the general theme of the USDA-SEA/AID BNF program, the major goal of this project was to begin nitrogen fixation research in northcentral Mexico in order to acquaint agronomists and researchers with the needs and techniques of nitrogen fixation research. The specific objective was to increase common bean yield and protein content through *R. phaseoli* strain selection.

## MATERIALS AND METHODS

### Experiment Locations

Field experiments were performed at a variety of locations. Irrigated experiments were conducted in New Mexico at the New Mexico Agricultural Experiment Station near Las Cruces in 1980 and on a farmer's field near Deming in 1981. In Mexico, irrigated experiments were conducted in both 1980 and 1981 at the CIAN experiment stations at Delicias, Chihuahua and Matamoros, Coahuila. In 1981, irrigated experiments were also conducted at the CENAMAR experiment station near Torreon, Coahuila and the Universidad Autonoma de Nuevo Leon near Monterrey, Nuevo Leon. Dryland experiments were conducted at the CIAN experiment station near Cuauhtemoc, Chihuahua and at the CIANOC Experiment Station near Victoria, Durango in both 1980 and 1981. Greenhouse experiments were conducted on the campus of NMSU.

### Experimental Design

Several types of experiments were designed. Strain trials included 10-20 strains of *R. phaseoli*, an uninoculated control, nitrogen fertilizer controls, and occasionally phosphorus fertilizer controls. Single strain peat based inoculants were prepared at NMSU and applied at twenty times the normal application rate (100 g powder inoculant/kg seed). The

experimental design was a randomized complete block with 3-5 replications depending on year and location. Individual plots were usually 10 m long with 1-3 rows per plot. Strain experiments were conducted at Victoria, Cuauhtemoc, and Monterrey.

Strain by variety interaction experiments included 3 varieties, 3 replications, and 8-10 treatments. An uninoculated control, nitrogen control, and 6-8 strains were included in the treatments. The design was a split-plot with treatment (strains) as the main plot. Strain by variety experiments were conducted at CENAMAR, Delicias, Matamoros, Las Cruces, and Deming.

Persistance experiments were conducted in 1981 by planting in the exact 1980 plot area without inoculation. Persistence experiments were conducted at Delicias, Matamoros, and Cuauhtemoc.

Two international inoculation trials were also conducted. In 1980, an IBIT (Prueba Internacional de Cepas de Rhizobium para Frijol) trial was supplied by Dr. Peter Grapham of CIAT (Centro Internacional de Agricultura Tropical). The experiment was located at Victoria. In 1981, an inoculation and fertility trial was supplied by Dr. Jake Halliday of NifTAL (Nitrogen Fixation by Tropical Agricultural Legumes). This experiment was located at CENAMAR.

A greenhouse experiment was conducted in 1981 to test the interaction of strain and variety. This experiment included 3 varieties and 15 strains with uninoculated and nitrogen fertilizer controls. Table 1 lists all experiments and locations.

Inoculants and Rhizobium Strains

Single strain inoculants were prepared at NMSU with sterile peat. Powdered, sterile (gamma irridiated) peat was supplied by Nitragin Inc.

Table 1. Location and design of experiments in 1980 and 1981.

| Location | Year | Design |
|---|---|---|
| Las Cruces (field) | 1980 | Strain x variety (field) |
| Las Cruces (greenhouse) | 1981 | Strain x variety (greenhouse) |
| Deming | 1981 | Strain x variety |
| CENAMAR | 1981a | Strain x variety |
| CENAMAR | 1981b | NifTAL |
| Delicias | 1980 | Strain x variety |
| Delicias | 1981 | Persistence |
| Matamoros | 1980 | Strain x variety |
| Matamoros | 1981 | Persistence |
| Cuauhtemoc* | 1980 | Strain trial |
| Cuauhtemoc* | 1981a | Strain trial |
| Cuauhtemoc* | 1981b | Persistence |
| Victoria* | 1980 | IBIT |
| Victoria* | 1981 | Strain trail |
| Monterrey | 1981 | Strain trail |

*Indicates dryland conditions, all other locations were furrow irrigated.

Individual polyethylene bags of peat (125 g) were injected with 60 ml of a 3-4 day old YEM culture (3) containing 9.4 g of $CaCO_3$. The bags were kneaded daily for one week at room temperature before refrigeration at 4°C.

Following these directions, the addition of 60 mls of culture (approximately $2 \times 10^9$ cells/ml) resulted in an initial peat inoculant containing approximately $8 \times 10^8$ cells/g. During the week of incubation, the numbers increased to approximately $5 \times 10^9$ cells/g and then declined slightly to between $2 \times 10^9$ - $4 \times 10^9$ cells/g. After refrigeration, the numbers remained steady between these limits for at least 3 months.

Each polyethylene bag produced approximately 190 g of inoculant with a final pH of approximately 7.1 and a moisture content of approximately 55% (dry weight basis). Occasionally bacterial contaminants were found in some of the bags during periodic checks, but the rhizobial numbers were always greater than the contaminant numbers.

In 1980, all inoculants were plated prior to field application and were found to contain between $2 \times 10^9$ - $4 \times 10^9$ cells/g. In 1981, only a few spot checks were made of the inoculants and all were found to be within these limits.

R. phaseoli strains were provided by numerous investigators (Table 2). All experimental data herein refer to the N.M. number and not numbers assigned by other individuals or institutions. N.M. strains 18-25 were all isolations made from local bean fields. Only strains 19 and 25 still are in the culture collection. The other strains reproduced poorly on agar slants and were eventually lost.

Table 2. *Rhizobium phaseoli* strains used in 1980 and 1981 experiments.

| New Mexico number | Other numbers | Source |
|---|---|---|
| NM strain 1 | 127K51 | Ham-originally from Burton |
| NM " 2 | IQ 423 | Ham |
| NM " 3 | IP 736A1 | Ham-originally from Bezdicek |
| NM " 4 | CIAT 404 | Ham-originally from CIAT |
| NM " 5 | Allen 413-2 | Ham-originally from Wisconsin |
| NM " 6 | 650-R | Ham-originally from Rhodesia |
| NM " 7 | 3644 | Ham-originally from Rhodesia |
| NM " 8 | Kim 5 | Bezdicek |
| NM " 9 | IP347 | Bezdicek |
| NM " 10 | 127K74 | Bezdicek-originally from Burton |
| NM " 11 | 127K79 | Bezdicek-originally from Burton |
| NM " 12 | NSRM 36-4 | Ham |
| NM " 13 | NSRM 36-1 | Ham |
| NM " 14 | CC511 | Ham-originally from Australia |
| NM " 15 | QA1061 | Ham-originally from Australia |
| NM " 16 | QA1062 | Ham-originally from Australia |
| NM " 17 | IP566A5 | Ham |
| NM " 18 | --- | Lindemann-Isolation near Deming |
| NM " 19 | --- | " " " " " |
| NM " 21 | --- | " " " " " |
| NM " 22 | --- | " " " " " |
| NM " 24 | --- | " " " " " |
| NM " 25 | --- | " " " " " |
| NM " 26 | 2667 | USDA |
| NM " 27 | 2668 | USDA |
| NM " 28 | CIAT 632 | USDA |
| NM " 29 | CIAT 640 | USDA |

Bean Varieties

Several pinto bean varieties (supplied by CIAN) were used (Table 3). In general, the Mexican beans were smaller, more mottled, and later in maturity than American varieties. Pinto Mexicano 80, Matamoros 64, and Delicias 71 are generally planted under irrigated conditions. The varieties used at Victoria and Cuauhtemoc are varieties adapted for dryland conditions. However, in the mountaneous regions of Durango and Chihuahua, many local varieties are grown that vary considerably in color, size, and mottling. All of the varieties used were certified seed and were not coated with fungicides.

Measurements

A variety of measurements were taken. At Las Cruces and Deming, nodulation samples were taken approximately 6-10 weeks after planting (full flowering and some pod set). A golf green cup plugger measuring 10 cm in diameter and 15 cm in depth was placed over the tap root of one or two plants. This soil core with nodule and roots was returned to the laboratory where the nodules were separated, counted, and dried. In 1980, color scores were taken on a portion of the nodules. In 1981, plant dry weights and nitrogen determinations were made on the top portion of the plants. Yield was determined by harvesting approximately 8 m of row. Nitrogen determinations were made on the ground seed. Periodic notes on growth, maturity, disease, etc., were made but are not included in this report. No strain identification of the nodules was attempted.

Nodulation and yield measurements in Mexico were similar. However, the Mexicans also took a variety of additional measurements including number of pods per plant, number of seeds per pod, plant height, etc., which are not reported.

Table 3. *Phaseolus vulgaris* varieties used in 1980 and 1981 experiments.

| Location | Year | Varieties |
|---|---|---|
| Las Cruces (field) | 1981 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| Las Cruces (greenhouse) | 1981 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| Deming | 1981 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| CENAMAR | 1981a | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| CENAMAR | 1981b | Pinto Mexicano |
| Delicias | 1980 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| Delicias | 1981 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| Matamoros | 1980 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| Matamoros | 1981 | Pinto Mexicano 80, Delicias 71, Matamoros 64 |
| Cuauhtemoc | 1980a | Mantequilla, Ojo de Cabre, Bayo Durango |
| Cuauhtemoc | 1981a | Ojo de Cabre |
| Cuauhtemoc | 1981b | Ojo de Cabre Regional |
| Victoria | 1980 | Flor de Mayo |
| Victoria | 1981 | Flor de Mayo |
| Monterrey | 1981 | -------- |

## RESULTS AND DISCUSSION

### Strain x Variety Experiments - New Mexico

Experimental locations at both Deming and Las Cruces had indigenous *R. phaseoli* populations (Table 4). The Las Cruces site had not had beans grown on the land in recent memory, but the Rio Grande valley has been farmed for several hundred years and may account for the relatively high population. At Deming, the site had no history of bean cultivation or inoculation.

No visual differences with respect to treatments were seen in the field at any time during the 1980 growing season. However, varietal differences were very obvious. Pinto Mexicana 80 was taller and greener than Matamoros 64 or Delicias 71 and was later in flowering and maturity. Several diseases attacked the plots late in the season, the most severe was bacterial blight. Although all three varieties were affected by the diseases, Pinto Mexicano 80 was the least damaged and showed the most resistance to bacterial blight. Matamoros 64 and Delicias 71 were more infested with the disease and showed less resistance than Pinto Mexicana 80 or the American pinto bean variety (Idaho 114) used as a border. Root-knot nematodes were observed on the roots only during the October harvest. No root-knots were seen during nodule sampling. The general growing conditions were otherwise ideal and the plots received more water than is usually given to beans.

The data from the first sampling date (35 days from planting) are presented in Table 5. No interaction was found between varieties and treatments, but some statistical differences were noted between varieties and/or treatments. However, in general, the data was so variable, especially the nodulation data, that no conclusions could be drawn.

Table 4. Soil characteristics of experimental sites at Las Cruces and Deming in 1980 and 1981, respectively, and Matamoros and Delicias in 1980.

| Location | pH | EC | Organic Matter | Total N | $NO_3$ N | $NaHCO_3$ P | Texture | *R. phaseoli* |
|---|---|---|---|---|---|---|---|---|
| | | mmhos/cm | % | --------ppm------- | | | | #/g |
| Las Cruces | 7.6 | 1.46 | 1.2 | 639 | 18.2 | 9.2 | Clay loam | $6.5 \times 10^3$ |
| Deming | 7.4 | 0.98 | 1.1 | 621 | 22.6 | 11.8 | Clay | $1.0 \times 10^2$ |
| Matamoros | 7.8 | 0.72 | 0.9 | 278 | 1.6 | 9.6 | Clay | ND |
| Delicias | 7.3 | 1.58 | 0.4 | 381 | 15.8 | 8.0 | Sandy clay loam | ND |

ND = not determined

Table 5a. Effect of treatment (6 R. phaseoli strains and 2 controls) on bean nodulation, dry weight, and percent nitrogen averaged over 3 varieties. First sampling date, 35 days from planting, July 22, 1980, at Las Cruces.

| Treatment | Plant dry weight | Plant nitrogen | Nodule number | Nodule dry weight | Average nodule dry weight |
|---|---|---|---|---|---|
| | g/plant | % | #/plant | mg/plant | mg/nodule |
| NM 2 | 16.2 ab[1/] | 3.1 a | 10.0 a | 7.4 a | 0.62 a |
| NM 5 | 17.4 ab | 3.2 a | 2.6 a | 0.7 a | 0.17 a |
| NM 6 | 19.2 a | 3.1 a | 4.9 a | 1.5 a | 0.38 a |
| NM 7 | 16.3 ab | 3.2 a | 12.4 a | 7.0 a | 0.51 a |
| NM 8 | 15.8 ab | 3.2 a | 33.4 a | 16.8 a | 0.46 a |
| NM 9 | 15.9 ab | 3.0 a | 31.9 a | 18.5 a | 0.89 a |
| C-No Rhizobium | 17.4 ab | 3.2 a | 4.7 a | 3.5 a | 0.47 a |
| N-40 kg/ha | 14.0 b | 3.0 a | 35.3 a | 27.9 a | 0.62 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

Table 5b. Effect of variety on nodulation, dry weight, and percent nitrogen averaged over 8 treatments. First sampling date, 35 days from planting, July 22, 1980, at Las Cruces.

| Bean variety | Plant dry weight | Plant nitrogen | Nodule number | Nodule dry weight | Average nodule dry weight |
|---|---|---|---|---|---|
| | g/plant | % | #/plant | mg/plant | mg/nodule |
| Matamoros 64 | 17.9 a[1/] | 3.3 a | 8.6 a | 13.9 a | 0.44 a |
| Delicias 71 | 16.2 ab | 3.1 b | 17.6 a | 12.2 a | 0.74 a |
| Pinto Mexicano 80 | 15.6 b | 3.0 b | 25.5 a | 5.2 a | 0.61 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

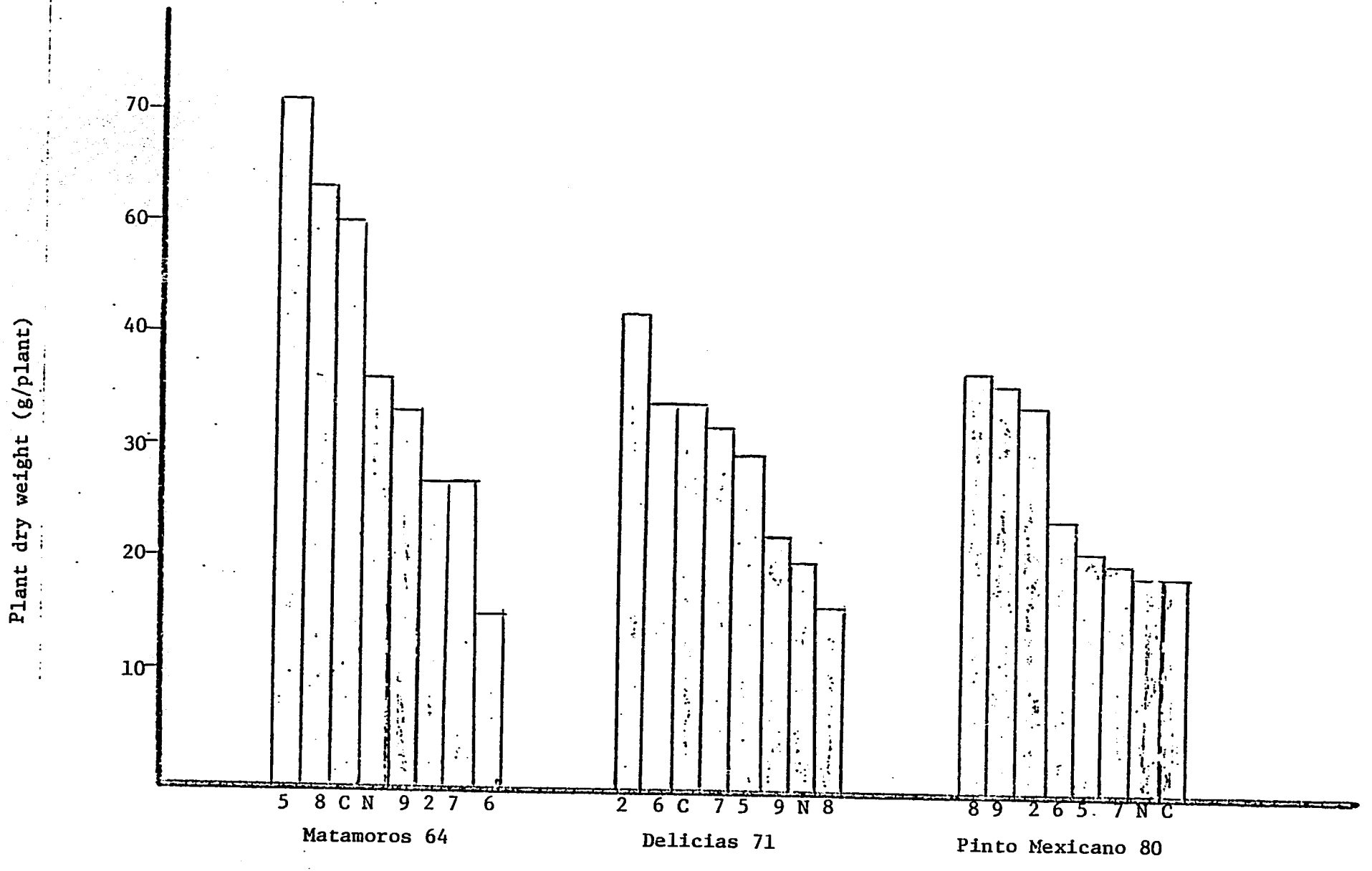


Figure 1. Interaction of 3 pinto bean varieties and 8 treatments including 6 R. phaseoli strains (NM 2,5,6,7,8,9), uninoculated control (C), and nitrogen control (N) on plant dry weight. Second sampling date, 55 days from planting, August 11, 1980 at Las Cruces.

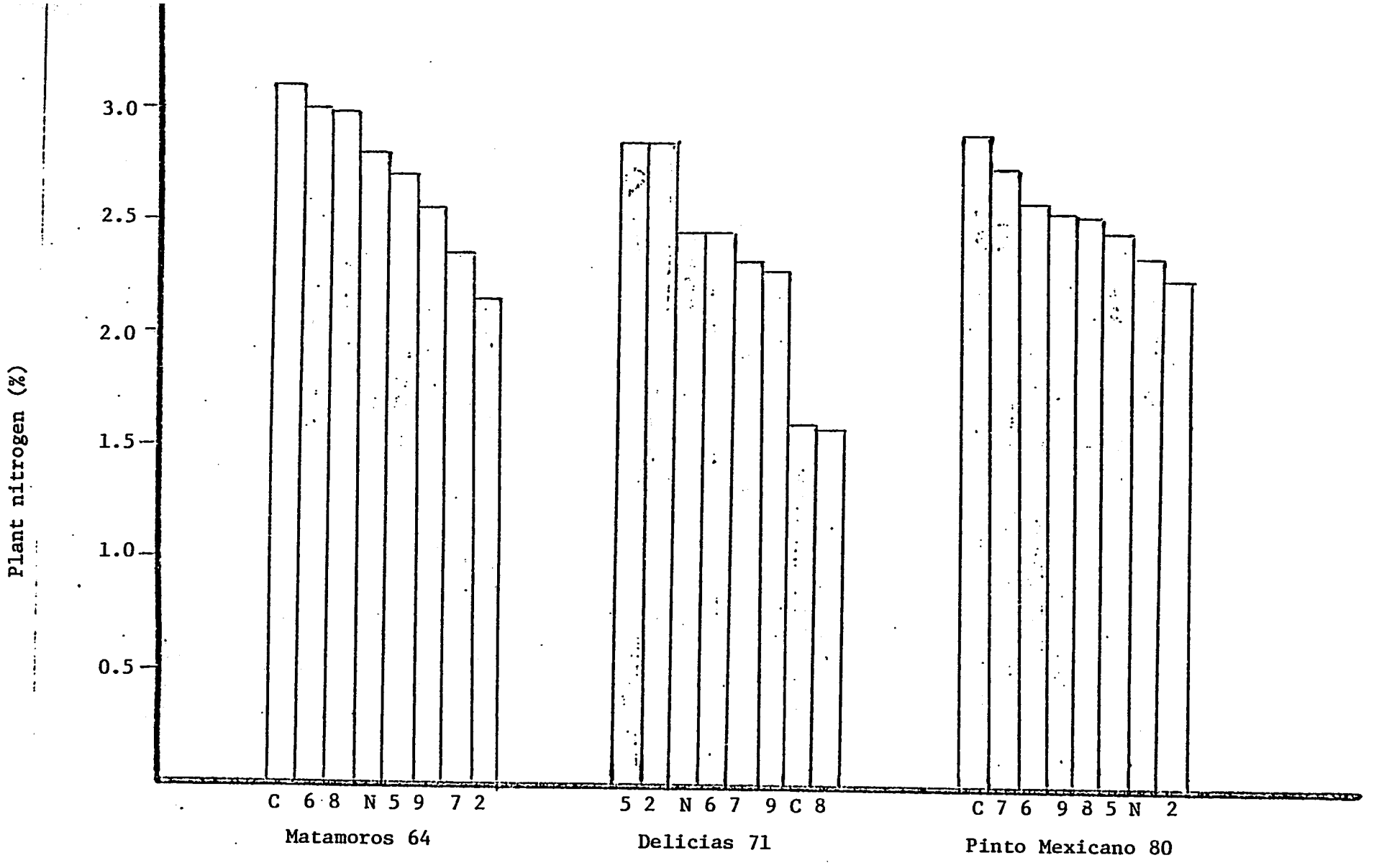


Figure 2. Interaction of 3 pinto bean varieties and 8 treatments including 6 R. phaseoli strains (NM 2,5,6,7,8,9), uninoculated control (C), and nitrogen control (N) on plant nitrogen content. Second sampling date, 55 days from planting, August 11, 1980 at Las Cruces.

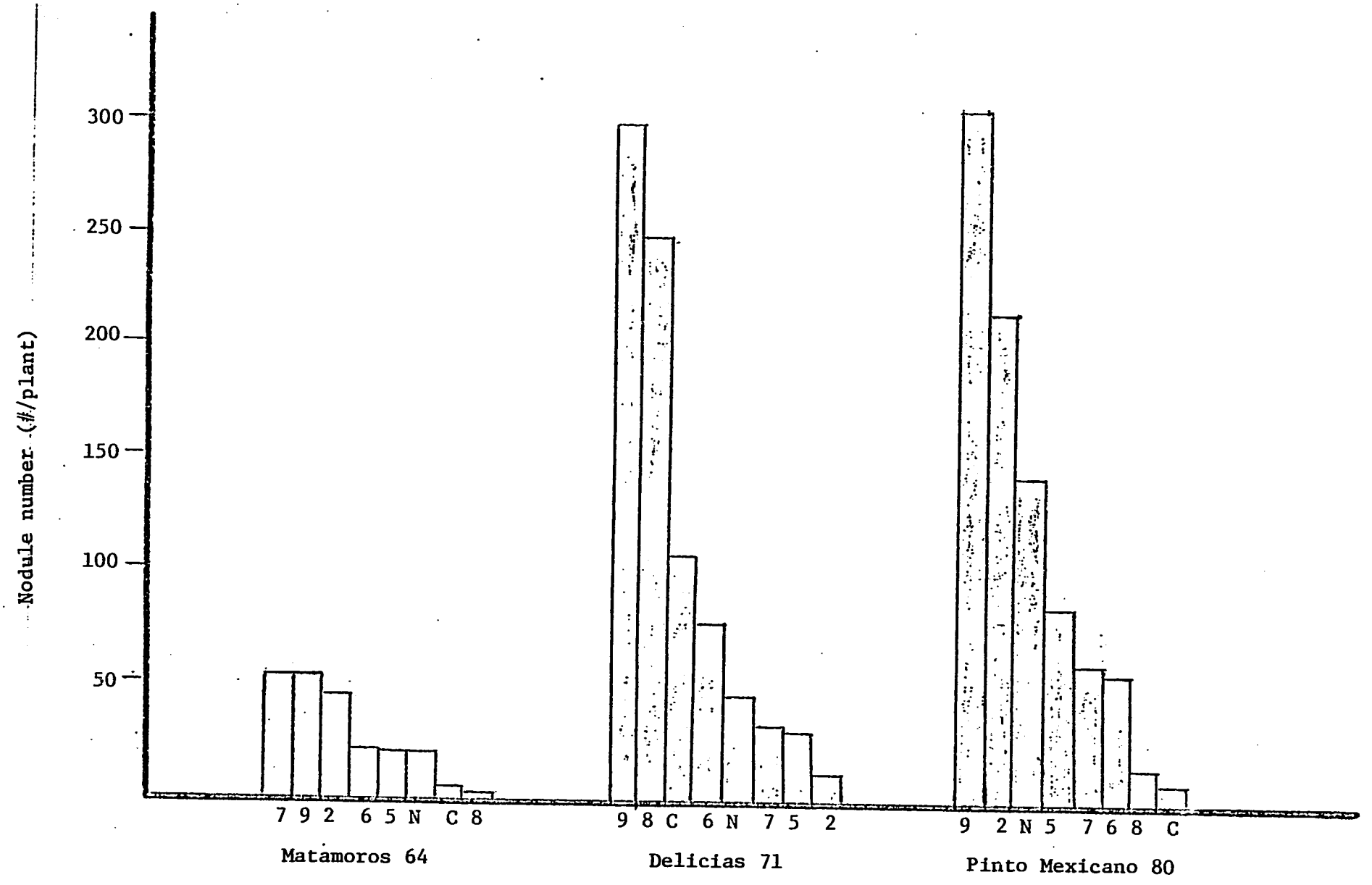


Figure 3. Interaction of 3 pinto bean varieties and 8 treatments including R. phaseoli strains (NM 2,5,6,7,8,9), uninoculated control (C), and nitrogen control (N) on nodule number. Second sampling date, 55 days from planting, August 11, 1980 at Las Cruces.

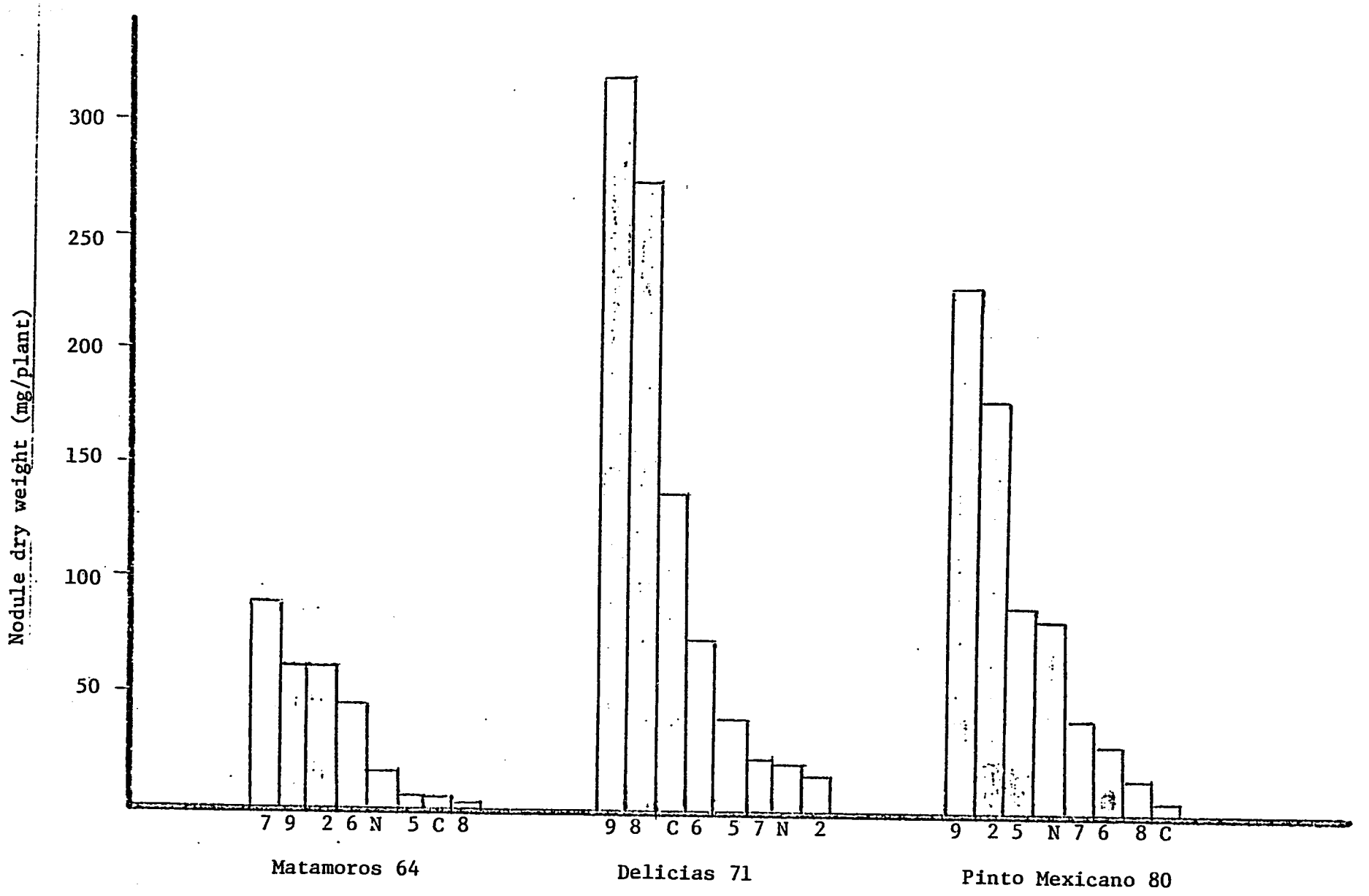


Figure 4. Interaction of 3 pinto bean varieties and 8 treatments including 6 R. phaseoli strains (NM 2,5,6,7,8,9), uninoculated control (C), and nitrogen control (N) on nodule dry weight. Second sampling date, 55 days from planting, August 11, 1980 at Las Cruces.

The second sampling date (August 11, 55 days from planting) produced much less variability in the sampling parameters. The number and weight of nodules had increased about tenfold. Highly significant variety by treatment interactions were noted for all measured parameters (except average nodule weight) from the second sampling date (Figure 1-4). However, no positive correlations was found when comparing plant dry weight or plant nitrogen with nodule number or nodule weight. In fact, correlation coefficients were generally negative when comparing nodulation parameters and plant parameters. Strain NM 9 generally produced the most nodule number and weight. However, the superior nodulation did not result in greater plant growth, plant N (Figure 1-4, or yield (Table 6). Strain 9 appears to nodulate well but not efficiently fix nitrogen. Strain 8 produced the least number and weight of nodules but produced the highest yields. Such results are not unusual in that some strains may produce many nodules but fail to efficiently fix nitrogen.

In 1981, the experiment did not receive sufficient water. Germination was sporadic over three weeks, the stand was spotty, nodulation was poor, and the plants were generally in poor shape. No visual differences were seen during vegetative growth, but at harvest the control (no Rhizobium) definitely was shorter and the bushes less full than the other treatments. There were few disease and insect problems. No statistically significant interaction was found between treatments and varieties for the variables measured.

Nodulation was poor and extremely variable and did not correlate with yield or other plant parameters (Table 7). Core samples were only taken to a 15 cm depth, yet the top 6-8 cm of soil was always dry.

Table 6a. Effect of treatment (6 *R. phaseoli* strains and 2 controls) on bean yield, protein, and seed weight averaged over 3 varieties in 1980 at Las Cruces.

| Treatment | Bean yield | Bean protein | Bean weight |
|---|---|---|---|
| | kg/ha | % | g/100 seeds |
| NM 2 | 3236 ab[1/] | 17.3 a | 27.6 a |
| NM 5 | 3401 ab | 17.9 a | 26.7 a |
| NM 6 | 2861 b | 17.9 a | 26.6 a |
| NM 7 | 3217 ab | 18.6 a | 26.6 a |
| NM 8 | 3637 a | 18.4 a | 27.0 a |
| NM 9 | 3067 ab | 17.9 a | 28.0 a |
| C-No *Rhizobium* | 3262 ab | 17.8 a | 27.0 a |
| N-40 kg/ha | 3323 ab | 18.3 a | 27.9 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Dunca's Multiple Range Test.

Table 6b. Effect of variety on bean yield, protein, and seed weight averaged over 8 treatments in 1980 at Las Cruces.

| Bean variety | Bean yield[1/] | Bean protein[1/] | Bean weight[1/] |
|---|---|---|---|
| | kg/ha | % | g/100 seeds |
| Matamoros 64 | 3026 b | 17.3 b | 28.6 b |
| Delicias 71 | 2684 b | 17.5 b | 23.0 c |
| Pinto Mexicano 80 | 4041 a | 19.2 a | 29.9 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

Table 7a. Effect of treatment (7 *R. phaseoli* strains and 2 controls) on bean nodulation, dry weight, and percent nitrogen averaged over 3 varieties on August 18, 1981, 54 days from planting at Deming.

| Treatment[1/] | Plant dry weight | Plant nitrogen | Nodule number | Nodule dry weight | Average nodule dry weight |
|---|---|---|---|---|---|
| | g/plant | % | #/plant | mg/plant | mg/nodule |
| NM 2 | 20.4 | 4.6 | 9.3 | 3.4 | 0.44 |
| NM 5 | 28.3 | 4.6 | 15.7 | 8.8 | 0.56 |
| NM 8 | 27.7 | 4.5 | 17.0 | 7.3 | 0.49 |
| NM 17 | 19.2 | 4.8 | 14.0 | 5.4 | 0.50 |
| NM 18 | 24.3 | 4.5 | 20.0 | 11.7 | 0.44 |
| NM 26 | 21.4 | 4.4 | 24.0 | 31.4 | 0.97 |
| NM 29 | 16.6 | 4.6 | 14.0 | 18.2 | 1.20 |
| C-No *Rhizobium* | 20.2 | 4.5 | 14.0 | 13.4 | 0.92 |
| N-40 kg/ha | 16.4 | 4.5 | 8.7 | 3.4 | 0.56 |

[1/] Treatment had no statistically significant effect on yield or protein.

Table 7b. Effect of variety on nodulation, dry weight, and percent nitrogen averaged over 9 treatments on August 18, 1981, 54 days from planting, at Deming.

| Bean variety | Plant weight | Plant nitrogen | Nodule number | Nodule dry weight | Average nodule dry weight |
|---|---|---|---|---|---|
| | g/plant | % | #/plant | mg/plant | mg/nodule |
| Matomoros 64 | 26.1 a[1/] | 4.5 a | 17.6 ab | 13.7 a | 0.60 a |
| Delicias 71 | 21.2 ab | 4.5 a | 7.6 b | 7.0 a | 0.75 a |
| Pinto Mexicano 80 | 17.5 b | 4.7 a | 20.9 a | 13.7 a | 0.70 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

After the preplant irrigation the top of the bed never became wet with irrigation water. The plants received irrigation water only by seepage from the furrow. Thus, the soil cores may have only sampled 6-8 cm of the nodulated root, the remainder of the core sample being primarily stem and dry soil. Future sampling will take into account furrow irrigation wetting of the root system and the depth of nodulation.

Yield was lower than normally expected at Deming but was significantly increased by inoculation (Table 8). Inoculation more than doubled yield over the uninoculated control and increased yield over the nitrogen control. However, additional fertilizer nitrogen might have been needed on the nitrogen fertilizer plots. Although no significant differences between strains were found, strain NM 5 and NM 17 produced the lowest yield. Bean protein (as well as tissue nitrogen) was higher in 1981 than in 1980. The lower protein and tissue nitrogen in 1980 was attributed to dilution.

Several explanations can be given for the yield response to inoculation without a similar response in nodule mass. As previously mentioned, the core samples may have missed the bulk of the nodules. Secondly, the inoculant strains may have been more efficient in nitrogen fixation. Nodule color scores were not taken because of the lack of good nodulation.

It may be interesting to note that another experiment adjacent to the USDA-SEA/AID experiment in the same field showed no statistcally significant response to inoculation. In this state sponsored experiment, several rates (1x, 2x, 5x normal rates) of powder and granular commercial inoculant were applied. Yields in this state experiment averaged 1239 kg/ha for the inoculated plots and 1072 kg/ha for the

Table 8a. Effect of treatment (7 *R. phaseoli* strains and 2 controls) on bean yield and protein averaged over 3 varieties in 1981 at Deming.

| Treatment | Bean yield | Bean protein |
|---|---|---|
| | kg/ha | % |
| NM 2 | 1895 a[1/] | 27.7 bc |
| NM 5 | 1338 a | 29.3 ab |
| NM 8 | 1768 a | 26.4 c |
| NM 17 | 1411 a | 29.8 ab |
| NM 18 | 1968 a | 27.5 bc |
| NM 26 | 1673 a | 28.4 bc |
| NM 29 | 1777 a | 27.4 bc |
| C-No *Rhizobium* | 770 b | 31.9 a |
| N-40 kg/ha | 1480 a | 31.8 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

Table 8b. Effect of variety on bean yield and protein average over 9 treatments in 1981 at Deming.

| | Bean yield | Bean protein |
|---|---|---|
| | kg/ha | % |
| Matamoros 64 | 1679 a[1/] | 28.4 a |
| Delicias 71 | 1579 ab | 28.2 a |
| Pinto Mexicano 82 | 1439 b | 29.9 b |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

uninoculated control. Although the American pinto bean variety (Idaho 114) outyielded the Mexican varieties and no direct comparisons can be made, several additional explanations can be considered. The American pinto beans may not respond to inoculation or the strains may not be as compatible with the American bean. Also, a large population of inoculant rhizobia may be needed to overcome the indigenous rhizobia since inoculants were added at 20x the normal rate in the USDA-SEA/AID experiment. Finally, a large amount of inoculum rhizobia may have been needed simply to nodulate the beans in 1981 when water was limiting and the seed bed was dry.

In the greenhouse experiment, sixteen treatments (fourteen strains and two controls) were tested on three varieties with five replications. The medium was sand and vermiculite (50:50, v/v), and the plants were watered with nitrogen free nutrient solution (4). The fourteen strains of R. phaseoli were N.M. 1, 5, 6, 7, 8, 10, 13, 14, 17, 18, 19, 21, 22, 24) and the three varieties were Delicias 71, Matamoros 64, and Pinto Mexicano 80. Parameters measured included nodule number, nodule dry weight, and plant dry weight.

Treatment and variety interacted significantly for nodule number and plant dry weight, but not for nodule weight. Both treatment and variety were significant for nodule weight. Pinto Mexicano 80 had the highest nodule number, nodule dry weight, and plant dry weight. The uninoculated control, nitrogen control, and strain N.M. 17 had the lowest nodulation and plant dry weights. However, since there was interaction between treatments and varieties and the data require many graphs of these interactions the data is not included in this report.

Strain x Variety Experiments - Mexico

At Delicias in 1980, nodulation samples were taken four times during the reason and an analysis of variance performed at each sampling. To concentrate the data, only an average of the four dates is presented without an analysis of variance testing. However, there was no interaction of variety or treatment for the nodulation data. The only statistically significant difference found was between varieties on the second and third sampling dates.

Nodule number increased from an average of 17 per plant on August 11, 1980 to 98 per plant on Oct. 6, 1980. Pinto Mexicano 80 had significantly more nodules than Delicias 71 and Matamoros 64 on the second and third sampling dates and when the four sampling dates are averaged (Table 9). Since no significant treatment differences were noted for each of the four sampling dates, it is doubtful that treatment had a statistically significant effect on nodulation when the average is considered (Table 9). However, nitrogen fertilization did tend to decrease the number of nodules on each sampling date and on the average. There appears to be a large indigenous *R. phaseoli* population as shown by the number of nodules on the control plots.

Inoculation increased yield an average of approximately 500 kg/ha over the uninoculated control (Table 9). As in Las Cruces in 1980, Pinto Mexicano 80 produced the highest yield. Nitrogen fertilization with 40 kg N/ha had approximately the same effect as inoculation. Thus, a tentative conclusion might be that inoculation resulted in an additional 40 kg/ha of nitrogen fixation. The lack of significantly more nodulation of the inoculated plots as compared to the control yet higher yield indicates that the inoculant strains were more efficient than the

Table 9a. Effect of treatment (6 R. phaseoli strains and 2 controls) on bean nodulation and yield averaged over 3 varieties on 1981 at Delicias. The nodule number data are an average of four sampling dates. No statistical analysis was performed on the averaged data.

| Treatment | Nodule number | Bean yield |
|---|---|---|
| | #/plant | kg/ha |
| NM 2 | 49.4 | 1616 ab[1/] |
| NM 5 | 51.9 | 1937 a |
| NM 6 | 37.5 | 1758 a |
| NM 7 | 66.5 | 1725 a |
| NM 8 | 79.8 | 1763 a |
| NM 9 | 49.7 | 1799 a |
| C-No Rhizobium | 51.6 | 1276 b |
| N-40 kg/ha | 35.1 | 1792 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

Table 9b. Effect of variety on nodulation and yield averaged over 8 treatments in 1980 at Delicias. The nodulation data is an average of 4 sampling dates No statistics were performed on the averaged data.

| Bean variety | Nodule number | Bean yield |
|---|---|---|
| | #/plant | kg/ha |
| Matamoros 64 | 36.1 b[1/] | 1508 b |
| Delicias 71 | 35.1 b | 1552 b |
| Pinto Mexicano 80 | 89.1 a | 2064 a |

[1/] Means in the same column followed by the same letter are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

indigenous strains of R. phaseoli. However, no strain identification of the nodules was performed. Also, the inoculum was applied at 20x the normal rate.

At Matamoros in 1980, only one sample for nodulation was taken for nodulation at 50% flowering. No interaction was found between variety and strain for the nodulation or yield parameters. Thus, the data (Table 10) are presented as means over treatments and varieties. There also appeared to be some problem with the statistical analysis and the setup of the analysis done at Matamoros, thus statistics are not included in the tables.

Nodulation was poor when samples were taken. As at Delicias, Deming, and Las Cruces, nodulation of the uninoculated control was almost as good as the inoculated plots. Also, nitrogen fertilizer appeared to inhibit nodulation.

Average yield on the inoculated plots was approximately 375 kg/ha higher than on the uninoculated control. However, it is doubtful if this difference was statistically significant. The yields were in general low and highly variable. Researchers at Matamoros attributed the low yields to this particular clayey soil.

At CENAMAR in 1981, nodulation was poor. Dr. Lee indicated high levels of soil nitrate remained from the previous crop. There was no statistically significant effect of treatment, variety, or treatment x variety interaction on the parameters measured except for variety on yield (Table 11). As at the other irrigated locations, an indigenous R. phaseoli exists as shown by nodulation of the three controls, but there was a trend (not statistically significant) for inoculation to increase nodulation and yield.

Table 10a. Effect of treatment (6 R. phaseoli strains and 2 controls) on bean nodulation and yield averaged over 3 varieties in 1980 at Matamoros. Statistics were performed on the original data in Mexico, but it is unclear as to the actual values and experimental statistical designs and tests need for the analysis of variance. Thus, the statistics are omitted.

| Treatment | Nodule number | Nodule fresh weight | Bean yield |
|---|---|---|---|
| | #/plant | mg/plant | kg/ha |
| NM 2 | 8.1 | 58.5 | 733 |
| NM 5 | 13.4 | 34.7 | 1005 |
| NM 6 | 4.4 | 15.6 | 801 |
| NM 7 | 24.6 | 56.8 | 962 |
| NM 8 | 20.6 | 60.9 | 1047 |
| NM 9 | 17.5 | 69.6 | 702 |
| C-No Rhizobium | 10.6 | 31.7 | 501 |
| N-40 kg/ha | 9.0 | 20.2 | 530 |

Table 10b. Effect of variety on nodulation and yield averaged over 8 treatments in 1980 at Delicias. Statistics were performed on the original data in Mexico, but it is unclear as to the actual values and experimental statistical designs and tests used for the analysis of variance. Thus, the statistics are omitted.

| Bean variety | Nodule number | Nodule fresh weight | Bean yield |
|---|---|---|---|
| | #/plant | mg/plant | kg/ha |
| Matamoros 64 | 14.4 | 53.3 | 739 |
| Delicias 71 | 11.8 | 43.8 | 855 |
| Pinto Mexicano 80 | 14.4 | 33.4 | 761 |

Table 11a. Effect of treatment (7 *R. phaseoli* strains and 3 controls) on bean nodulation, yield, and bean protein averaged over 3 varieties in 1981 at CENAMAR.

| Treatment[1/] | Nodule number | Nodule dry weight | Bean yield | Bean protein |
|---|---|---|---|---|
| | #/plant | mg/plant | kg/ha | % |
| NM 5 | 3.3 | 0.4 | 1480 | 24.7 |
| NM 6 | 3.1 | 0.8 | 1407 | 24.0 |
| NM 8 | 5.2 | 0.9 | 1604 | 24.1 |
| NM 17 | 11.1 | 8.2 | 1535 | 24.0 |
| NM 24 | 10.9 | 11.3 | 1484 | 24.8 |
| NM 25 | 1.3 | 0.2 | 1285 | 24.7 |
| NM 28 | 6.3 | 3.3 | 1401 | 24.6 |
| N + P[2/] | 2.4 | 1.5 | 1447 | 25.4 |
| N - P[3/] | 0.9 | 0.8 | 1646 | 25.2 |
| C[4/] | 1.4 | 0.7 | 1343 | 24.5 |

[1/] Treatment had no statistically significant effect on the parameters measured.

[2/] N + P plots received 40 kg N/ha and 60 kg P/ha but no *Rhizobium*.

[3/] N - P plots received 40 kg N/ha but no P or *Rhizobium*.

[4/] Control plots received no N, P, or *Rhizobium*.

Table 11b. Effect of variety on bean nodulation, yield, and protein averaged over 10 treatments in 1981 at CENAMAR.

| Variety | Nodule number | Nodule dry weight | Bean yield | Bean protein |
|---|---|---|---|---|
| | #/plant | mg/plant | kg/ha | % |
| Matamoros | 3.6 a[1/] | 2.3 a | 1338 b | 25.2 a |
| Delicias 71 | 5.0 a | 2.0 a | 1627 a | 24.2 a |
| Pinto Mexicano 80 | 5.2 a | 4.1 a | 1426 b | 24.5 a |

[1/] Means followed by the same letter within the same column are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

Strain Trials

At Cuauhtemoc in 1980, several experiments were put in the field. However, drought resulted in abandonment of all but 1 experiment. This experiment consisted of 17 treatments (14 strains and 3 controls) and 3 varieties. The design was a randomized complete block. However, for all the data except yield, the varieties were averaged together. All plots except the C1 and C2 control received 50 kg P/ha.

Nodule number and plant dry weight taken 45 days after emergence are given in Table 12. In previous annual reports, these experimental results, and those in Table 13, were reported differently because of confusion in the data from Mexico. The controls (C1, C2, and N) had no Rhizobium applied, but control C2 had sterile peat applied at the same rate of peat application as in the inoculated plots. The low yield and plant dry weight of the C2 plots may have been due to a pathogen freely multiplying in this sterile peat. In general, inoculation increased nodule mass and plant dry weight, however, some of this increase was due to phosphorus fertilization. Strain differences were apparent.

Yields (Table 13) were very low for each of the varieties. The poor yields were attributed to the drought. However, inoculation in general significantly increased yields on each of the varieties as compared to the C1 and C2 controls. For each variety, nitrogen fertilization resulted in about 100 kg/ha more yield when all 14 strains are averaged. It is interesting to note that strain N.M. 8, which was the best overall strain at the irrigated locations, was only mediocre on all the three varieties under dryland conditions.

In 1981, a similar strain trial was conducted with some of the same strains used in 1980 plus additional strains. Only one variety (Ojo de

Table 12. Effect of treatment (14 *R. phaseoli* strains and 3 controls) on bean nodulation and plant dry weight averaged over 3 varieties in 1980 at Cuauhtemoc.

| Treatment | Nodule number | Plant dry weight |
|---|---|---|
| | #/plant | g/plant |
| NM 2[1/] | 20.6 a[1/] | 135.6 a |
| NM 3 | 21.3 a | 109.5 b |
| NM 4 | 16.6 de | 55.6 g |
| NM 5 | 21.3 a | 114.8 b |
| NM 6 | 18.3 bcd | 104.4 bc |
| NM 7 | 16.6 de | 82.5 de |
| NM 8 | 16.6 de | 67.3 fg |
| NM 9 | 12.6 fg | 74.6 ef |
| NM 12 | 16.3 de | 115.7 b |
| NM 13 | 17.6 cd | 91.7 cde |
| NM 14 | 19.6 abc | 92.3 cd |
| NM 15 | 14.3 ef | 111.8 b |
| NM 16 | 12.0 fgh | 75.7 ef |
| NM 17 | 11.6 gh | 81.4 de |
| N-30 kg/ha | 11.0 ghi | 107.9 b |
| C1[3/] | 9.0 i | 89.7 cde |
| C2[4/] | 10.0 hi | 25.8 h |

[1/] All plots except controls C1 and C2 received 50 kg P/ha.

[2/] Means followed by the same letter within the same column are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

[3/] C1 was a control without added P, N, or inoculant.

[4/] C2 was a control without added P or N but was inoculated with sterile peat without *Rhizobium*.

Table 13. Effect of inoculant treatment (14 *R. phaseoli* strains and 3 controls) on bean yield on 3 bean varieties in 1980 at Cuauhtemoc.

| Treatment | Bean variety | | |
|---|---|---|---|
| | Mantequilla | Ojo de Cabra | Bayo Durango |
| | kg/ha | | |
| NM 2[1/] | 386 b[2/] | 564 a | 501 b |
| NM 3 | 306 c | 425 bc | 403 c |
| NM 4 | 329 bc | 435 bc | 375 cd |
| NM 5 | 388 b | 513 b | 548 a |
| NM 6 | 389 b | 525 b | 404 c |
| NM 7 | 315 c | 521 b | 408 bc |
| NM 8 | 304 cd | 434 bc | 305 cd |
| NM 9 | 186 d | 188 cf | 58 c |
| NM 12 | 334 bc | 454 bc | 382 cd |
| NM 13 | 327 bc | 419 bcd | 305 cd |
| NM 14 | 212 cd | 102 f | 118 c |
| NM 15 | 96 d | 279 cde | 96 e |
| NM 16 | 344 bc | 382 bcde | 257 d |
| NM 17 | 548 a | 496 b | 418 bc |
| N-30kg/ha | 443 b | 530 b | 426 bc |
| C1[3/] | 192 d | 264 de | 288 d |
| C2[4/] | 35 e | 31 g | 22 e |

[1/] All plots except controls C1 and C2 received 50 kg P/ha.

[2/] Means followed by the same letter within the same column are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

[3/] C1 was a control without added P, N, or inoculant.

[4/] C2 was a control without added P or N but was inoculated with sterile peat containing no *Rhizobium* and fertilized with P but not N.

cabre) and two controls were used in this experiment. The experiment was sampled on two dates for nodulation and plant dry weight, 42 and 56 days from seedling emergence. Only the data from 42 days from emergence and yield are reported. No statistical analysis of the nodulation and plant data was received from Cuauhtemoc.

Nodulation was poor at 42 days from planting (Table 14) and did not improve much by the second planting date (data not shown). Few nodules were found on the N and C controls on the first sampling date and no nodules were found on these treatments on the second sampling date. Regardless of strain, inoculation appears to increase nodule mass and plant growth. Yield (Table 14) was significantly greater on the inoculated plots, averaging approximately 800 kg/ha more than the control (C) and approximately 350 kg/ha more than the nitrogen (N) treatments. The nitrogen control plots probably should have received more nitrogen than the 30 kg/ha applied. Also, the control (C) plots did not receive phosphorus fertilizer as they should have. Thus, some of the yield increase between the inoculated and control (C) plots may have been due to phosphorus fertilization. Strain differences were minor.

The experiment at Monterrey in 1981 had ten treatments (8 R. phaseoli strains and 2 controls) and four replications in a randomized complete block design. This experiment will not be discussed in detail because the researcher did not follow directions. The amounts of nitrogen and phosphorus added, the bean variety, and other details were not recorded. No nodulation samples were taken, but about 20 other plant parameters were measured. Also, the researcher did not use the appropriate controls and the control treatments appear to be switched. The data (Table 15) show statistical differences between treatments. However, no conclusions can be drawn because of the problems mentioned.

Table 14. Effect of treatment (15 R. phaseoli strains and 2 controls) on bean nodulation, plant dry weight, and yield in 1981 at Cuauhtemoc. Statistical analysis was not performed on the nodulation or plant data.

| Treatment | Plant dry weight | Nodule number | Nodule dry weight | Bean yield |
|---|---|---|---|---|
| | g/plant | #/plant | mg/plant | kg/ha |
| NM 2[1/] | 9.0 a[2/] | 2.0 a | 10.4 a | 1290 a |
| NM 7 | 6.9 a | 20.4 a | 10.8 a | 1305 bcd |
| NM 8 | 10.8 a | 9.6 a | 18.3 a | 1182 cde |
| NM 10 | 8.7 a | 4.0 a | 10.8 a | 1154 de |
| NM 17 | 10.8 a | 22.6 a | 36.4 a | 1471 ab |
| NM 19 | 9.5 a | 1.0 a | 1.3 a | 1274 bcd |
| NM 21 | 10.7 a | 3.4 a | 9.4 a | 1173 cde |
| NM 22 | 11.2 a | 9.4 a | 10.8 a | 1234 bcd |
| NM 23 | 11.0 a | 4.6 a | 12.4 a | 1598 a |
| NM 24 | 10.4 a | 7.8 a | 17.0 a | 1164 cde |
| NM 25 | 11.1 a | 4.2 a | 7.0 a | 1185 cde |
| NM 26 | 8.8 a | 2.0 a | 10.2 a | 1367 bcd |
| NM 27 | 9.0 a | 17.8 a | 52.8 a | 1243 bcd |
| NM 28 | 11.6 a | 6.8 a | 24.5 a | 1343 bcd |
| NM 29 | 14.0 a | 21.6 a | 18.4 a | 1409 abc |
| N-30 kg/ha | 13.4 a | 1.0 a | 14.2 a | 955 e |
| C-No Rhizobium or P | 4.1 a | 1.0 a | 0.2 a | 485 f |

[1/] All plots except for the control (C) received 50 kg P/ha.

[2/] Means followed by the same letter are not different at the 5% probability level by Duncan's Multiple Range Test.

Table 15. Effect of treatment (8 *R. phaseoli* strains and 2 controls) on bean plant dry weight and yield in 1981 at Monterrey.

| Treatment | Plant dry weight -- 56 days | Plant dry weight -- 68 days | Bean yield |
|---|---|---|---|
| | g/plant | g/plant | kg/ha |
| NM 2 | 12.5 ab[1/] | 36.5 a | 2543 b |
| NM 5 | 14.2 ab | 36.2 a | 3027 ab |
| NM 6 | 13.2 ab | 23.8 a | 2073 b |
| NM 7 | 13.7 ab | 31.4 a | 2637 b |
| NM 8 | 16.2 a | 33.6 a | 2851 ab |
| NM 10 | 16.6 a | 38.0 a | 3173 ab |
| NM 17 | 15.[illegible] a | 39.0 a | 4276 a |
| NM 29 | 14.1 ab | 26.8 a | 3485 ab |
| N[2/] | 9.3 b | 21.9 a | 2249 b |
| C[3/] | 16.9 a | 26.2 a | 3186 ab |

[1/] Means followed by the same letter within the same columns are not statistically different at the 5% probability level by Duncan's Multiple Range Test.

[2/] Nitrogen and phosphorus fertilizer but no inoculant were applied.

[3/] No nitrogen or inoculant were applied but phosphorus fertilizer was applied at the same rate as in the nitrogen control.

The strain trial at Victoria in 1981 was never sampled and no data was recovered. Prior to sampling for nodulation, the plots were drowned with torrential rains.

Persistence Experiments

The persistence of inoculated strains was studied by planting in the same plot areas of the previous year. Thus, in 1981 the same plots from 1980 were replanted without additional inoculation at Delicias, Matamoros, and Cuauhtemoc. Additional fertilizer was added.

Nodulation and yield data from Delicias in 1981 are shown in Table 16. As in 1980, no significant treatment x variety interaction was found. Statistically significant effects were found for variety only. Average yield on the six inoculated plots was approximately 140 kg/ha greater than on the control plots and approximately 50 kg/ha greater than on the nitrogen plots. However, these comparisons were statistically insignificant. Differences between strains were small, but N.M. 8 yielded approximately 200 kg/ha more than all other treatments. N.M. 8 was previously shown to produce the highest yields in the strain x variety experiments when all locations, varieties, and years were considered. This strain was included in all experiments because of its reportedly good nitrogen fixation and competitiveness (Bezdicek, 1979--personal communication).

Nodulation was poor on the persistence study in Matamoros in 1981. However, nodulation was also poor in 1980. Researchers at Matamoros claim it is the soil type. Since nitrate and total nitrogen were low, inhibition of nodulation by excess nitrogen can be ruled out. Some of the data from the first sampling date (68 days from planting) are presented in Table 17. Little or no effect can be attributed to treatment.

Table 16a. Effect of persistence of 1980 treatments (6 *R. phaseoli* strains and 2 controls) on bean nodulation and yield averaged over 3 varieties in 1981 at Delicias.

| Treatment[1/] | Nodule number | Nodule dry weight | Bean yield |
|---|---|---|---|
| | #/plant | mg/plant | kg/ha |
| NM 2 | 76.0 | 14.6 | 1650 |
| NM 5 | 77.1 | 14.7 | 1749 |
| NM 6 | 61.2 | 9.7 | 1628 |
| NM 7 | 59.8 | 11.7 | 1567 |
| NM 8 | 94.3 | 20.2 | 1829 |
| NM 9 | 56.7 | 10.5 | 1641 |
| C-No *Rhizobium* | 76.5 | 15.8 | 1545 |
| N-40 kg/ha | 52.1 | 10.9 | 1636 |

[1/] Treatment had no significant effect on the parameters measured.

Table 16b. Effect of variety on nodulation and yield averaged over 8 persistence treatments in 1981 at Delicias.

| Variety | Nodule number | Nodule dry weight | Bean yield |
|---|---|---|---|
| | #/plant | mg/plant | kg/ha |
| Delicias 71 | 56.2 b[1/] | 14.6 a | 1644 b |
| Matamoros 64 | 83.0 a | 17.4 a | 1202 c |
| Pinto Mexicano 80 | 69.3 ab | 8.7 b | 2121 a |

[1/] Means followed by the same letter within the same columns are statistically different at the 5% probability level by Duncan's Multiple Range Test.

Table 17a. Effect of persistence of 1980 treatments (6 *R. phaseoli* strains and 2 controls) in bean nodulation and plant growth averaged over 3 varieties in 1981 at Matamoros.

| Treatment[1/] | Nodule number | Nodule fresh weight | Plant dry weight |
|---|---|---|---|
| | #/plant | mg/plant | g/plant |
| NM 2 | 4.8 | 13.1 | 11.3 |
| NM 5 | 12.0 | 39.7 | 10.9 |
| NM 6 | 4.5 | 12.5 | 12.5 |
| NM 7 | 8.1 | 41.3 | 12.2 |
| NM 8 | 10.8 | 32.3 | 12.2 |
| NM 9 | 9.6 | 51.9 | 12.0 |
| C-No *Rhizobium* | 8.5 | 24.2 | 11.8 |
| N-40 kg/ha | 8.4 | 33.6 | 12.6 |

[1/] Treatment had no statistically significant effect on the parameters measured.

Table 17b. Effect of variety on nodulation and plant growth averaged over 8 persistence treatments in 1981 at Matamoros.

| Variety[1/] | Nodule number | Nodule fresh weight | Plant dry weight |
|---|---|---|---|
| | #/plant | mg/plant | g/plant |
| Matamoros 64 | 9.5 | 37.9 | 13.9 |
| Delicias 71 | 12.5 | 56.3 | 11.8 |
| Pinto Mexicano 80 | 6.7 | 8.6 | 10.9 |

[1/] Variety had no statitically significant effect on the parameters measured.

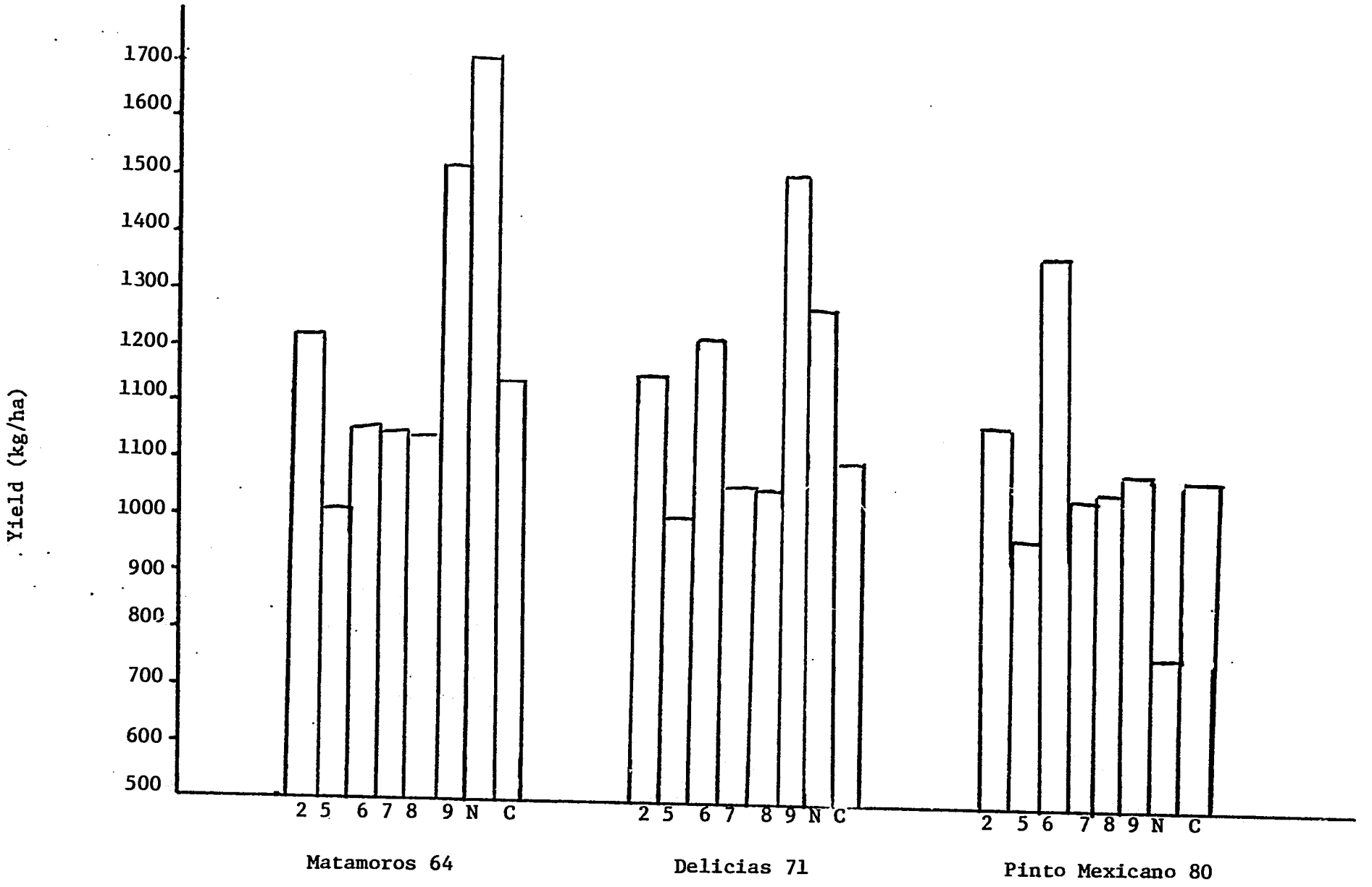


Figure 5. Interaction of 3 pinto bean varieties and 8 treatments including 6 R. phaseoli strains (NM 2,5,6,7,8,9), uninoculated control (C), and nitrogen control (N) on bean yield at Matamoros, 1981.

There was highly significant interactions between variety and strain on seed yield. The data are presented in Figure 5 to show this interaction. Although strain x variety interaction was significant, it is doubtful if any conclusions can be made. Most of this interaction occurred between the control and nitrogen fertilization treatments. Strain N.M. 6 was the poorest strain and N.M. 8 was also low. Strain N.M. 2 produced consistently higher yields than most strains. N.M. 5, which was considered to be one of the better strains in previous studies, consistently produced the lowest yields over all three varieties.

Precipitation at Cuauhtemoc in 1981 was near or above normal and yields were good (Table 18). As at Delicias and Matamoros, additional fertilizer but no additional inoculation was applied. No statistical data was received from Mexico but the treatments was replicated five times. All the plots except the control (C) received 50 kg/ha of phosphorus. The variety was Ojo de Cabre.

The inoculated plots yielded an average of 1316 kg/ha, approximately 322 kg/ha greater than the uninoculated and phosphorus fertilized plots ($C_1$ control). The data indicate that the inoculant rhizobia persisted from 1980 and that inoculation produced the same yields as the recommended rate of nitrogen fertilizer (30 kg N/ha). However, since 1981 was a good year for bean production, additional nitrogen fertilizer might have produced greater yields than the inoculated plots. Phosphorus fertilizer alone increased yields about 275 kg/ha, indicating that phosphorus fertilizer is needed. When applied together, both phosphorus fertilizer and inoculation increased yields approximately 600 kg/ha over the uninoculated and unfertilized plots. Strain differences are apparent, however except for strain 16, it is doubtful if these differences are statistically significant.

Table 18. Effect of persistence of 1980 treatments on bean in 1981 at at Cuauhtemoc.

| Treatment[1/] | Bean yield |
|---|---|
| | kg/ha |
| NM 2 | 1384 |
| NM 3 | 1224 |
| NM 4 | 1615 |
| NM 5 | 1419 |
| NM 6 | 1351 |
| NM 7 | 1371 |
| NM 8 | 1405 |
| NM 9 | 1169 |
| NM 12 | 1316 |
| NM 13 | 1395 |
| NM 14 | 1488 |
| NM 15 | 1158 |
| NM 16 | 996 |
| NM 17 | 1129 |
| N-30 kg N/ha | 1364 |
| C1 - P but no *Rhizobium* | 994 |
| C2 - no P or *Rhizobium* | 725 |

[1/] All plots except the C2 control received 50 kg P/ha.

Other Experiments

The strain trial at Victoria in 1980 was not sampled because of drought. Thus, in both 1980 and 1981, no data were received from Victoria because of drought and flooding in those years, respectively.

Yield data from the NifTAL trial at CENAMAR in 1981 are presented in Table 19. No statistical differences were found between the treatments. Yields were somewhat lower than usually expected at CENAMAR and were lower than the strain x variety experiment. In the NifTAL experiment the trend was for inoculation to reduce yields while in the strain x variety experiment inoculation tended to increase yields. Possibly the inoculant strains supplied by NifTAL were more applicable to acid soils and tropical conditions than to alkaline soils and arid conditions. However, no definitive conclusions can be drawn. Nodulation data were only taken on one replication and are not reported.

## DISCUSSION AND CONCLUSIONS

Under irrigated field conditions there was little interaction between strains of *R. phaseoli* and the three varieties of *P. vulgaris*. When the interaction of strain and variety was found to be statistically significant, mitigating circumstances occurred and the effect was primarily on nodulation and not yield. For example, at Las Cruces in 1980, the differential resistance of the three varieties to blight and nematode infection probably confounded the nodulation results on the second sampling date. At Matamoros in 1981 (persistence experiment) most of the interaction on yield occurred between the control and nitrogen treatments, not between the strain treatments. Highly significant interactions between strain and variety were found in the greenhouse

Table 19. Effect of fertilization and inoculation on yield of beans at CENAMAR in 1981. Experimental design and materials were supplied by NifTAL.

| | Treatment[1/] N | P | K | | Bean yield | Bean protein |
|---|---|---|---|---|---|---|
| | kg/ha | | | | kg/ha | % |
| F-1 | 0 | 60 | 60 | | 1143 | 26.7 |
| F-2 | 100 | 60 | 60 | | 1207 | 27.8 |
| F-3 | 0 | 60 | 60 | + inoculant | 1025 | 26.8 |
| M-1 | 0 | 100 | 100 | | 1263 | 27.5 |
| M-2 | 100 | 100 | 100 | | 1125 | 28.4 |
| M-3 | 0 | 100 | 100 | + inoculant | 1035 | 27.6 |

[1/] Treatment had no statistically significant effect on yield or protein.

experiment. However, under these ideal growth conditions and optimum nitrogen fixation conditions (nil soil nitrogen and no competing rhizobia) a significant interaction was expected. Evidently, the interaction between the strains and the varieties is small enough to be masked by field conditions.

Thus, we conclude that interaction between varieties Matamoros 64, Delicias 71 and Pinto Mexicano 80 and strains NM 2, 5, 6, 7, 8, 9, 17, 18, 24, 25, 26, 28, 29 is insignificant under irrigated field conditions. However, there often is a strain and/or variety effect on both nodulation and yield.

Differences between R. phaseoli strains were generally small both under irrigated and field conditions. Some strains showed great variability in ranking (based on yield) among other strains from year to year and location to location (for example, strains NM 2 and 9). Other strains were generally consistently high, but not necessarily the highest ranking strain in terms of yield (for example, strain NM 8). Some strains persisted better from season to season while others appeared to do better under either irrigated or dryland conditions but not both. However, all of these differences were small and, for the most part, statistically insignificant. Also, since no identification of the nodule contents was performed, it is difficult to make definitive statements about the persistence of the strains. Many of the strains were put into the field only once or twice which is not sufficient to make a determination of the best strains. Finally, inoculants were added at twenty times the normal rate to insure a high population of the inoculant strain and to compensate for minor differences in numbers in the single strain inoculants.

When all the seed yields for the strains are averaged, very little difference is found between strains. However. strains NM 5 and NM 8 are considered to be the best strains of those which were put into the field four times or more. These strains consistently produced higher yields in the numerous locations, years, and experiments but not necessarily the highest yields. Both strains NM 5 and NM 8 have been given to CIAN upon their request. Strains NM 6 and NM 9 were considered to be the poorest strains because of their inconsistency in the field. Many of the strains with numbers NM 10 and above will probably be eliminated after further field testing.

In most experiments, inoculation increased yield over the uninoculated control; although in some cases, the increased yield was not statistically significant. These yield increases were most dramatic in the dryland area near Cuauhtemoc and in the irrigated experiment in Deming. The irrigated valleys (Las Cruces, Delicias, CENAMAR, Matamoros) probably had higher populations of indigenous R. phaseoli, thus limiting the response to inoculation. The increases in yield due to inoculation at Deming and Cuauhtemoc were considered to be due to the addition of R. phaseoli as well as the introduction of more efficient strains. However, the yield increases or trends in the irrigated valleys were thought to be due to the introduction of more efficient strains.

Thus, we concluded that inoculation will probably result in a yield increase over uninoculated and unfertilized soil in the dryland areas around Cuauhtemoc, and inoculation can take the place of normally recommended fertilizer rates in these areas. Inoculation will probably not result in a yield increase over uninoculated and unfertilized soil in the irrigated valleys unless the inoculation is added many times the

normal recommended rate. This lack of response is attributed to a high indigenous R. phaseoli population. However, more work is needed in both the dryland and irrigated areas to determine if normal application rates will increase yield and to confirm these conclusions.

The persistence experiment at Cuauhtemoc demonstrated that inoculation not only increased yield but that the bacteria were able to survive and able to increase yields over the uninoculated controls the following year. This result was the most encouraging to the Mexican researchers.

## FUTURE RESEARCH

Future research under a new USDA-SEA/AID BNF grant (#59-2351-1-5-007-0) will focus on the dryland areas where most of the beans are grown in northcentral Mexico. Additional strain and persistence experiments will be planted. These experiments will include new strains, different varieties, persistence of rhizobia in the normal rotation of crops, and screening of dryland bean genotypes for greater nodulation. Additional research will be conducted under irrigated conditions to screen bean genotypes for greater nodulation as in the dryland experiment, phosphorus and nodulation interactions, and irrigation timing influence on nodulation.

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