Effect of inoculation and N application on growth, yield, nodulation and N fixation of cowpea (*Vigna unguiculata*)

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ABSTRACT

A series of experiments were conducted to study the effect of inoculation and N application on growth, yield, nodulation and N fixation of cowpea, involving both local and introduced varieties. In experiment 1, cowpea varieties, MI 35, Bombay and Arlington were used with (i) control (No fertilization), (ii) inoculation and (iii) fertilizer N (80 kg N ha⁻¹). In experiment 2, N fixing capacity of above three cowpea varieties was studied. In experiment 3, ten genotypes of cowpea i. e. Arlington, Bombay, MI 35, Ieta, CP 230, CP 889, CP 789, IT - 82 D-789, IT - 82 E-32 were used under two N regimes, with 80 kg N ha⁻¹ and without fertilizer (-N). All experiments were arranged in a randomized complete block design with 4 replications.

Results revealed that inoculation had no effect in producing a significant yield improvement in all local cowpea varieties. In response to N fertilization, MI-35 and Arlington produced significantly higher yield (51% and 40% increase over control). Cultivar Bombay did not show a significant response to fertilizer N, but produced the highest yield compared to other cvs. without fertilizer application. Cultivar Bombay showed significantly a higher N fixing capacity than MI-35 and Arlington. Bombay and leta showed a greater nodulation capacity followed by Arlington and MI 35. Among the introduced genotypes, IT 82D 889, IT 82E 32 and IT 82D 787 showed higher nodulation potential over other genotypes at 60 days after planting (DAP). But at 75 DAP, CP 230, CP 889 and CP 789 showed superior nodulation capacity.

A considerable high genotypic variability was observed among cowpea cvs. in BNF potential, total dry matter production and seed yield. Locally recommended cowpea cvs., Bombay and Ieta showed better nodulation capacity over introduced genotypes. N fertilizer inhibited nodulation in almost all cvs. The total dry matter production and yield responded positively to N fertilization and the effect was prominent in most of the introduced genotypes.

Key Words: Inoculation, Vigna unguiculata, Nodulation, N fixation, % Ndfa

INTRODUCTION

Nitrogen is one of the major plant nutrients for plant growth. The economic and environmental cost of increasing N fertilizer use has seriously restricted its application in agriculture, thus affecting increase food production, especially in developing countries. On the other hand, legumes are able to derive a considerable proportion of their N requirement from the atmosphere *via* symbiotic N_2 fixation and a part of fixed N is added to the soil. Therefore, N_2 fixation by legumes is considered to be an important alternative to chemical N fertilizer (Ofori and Stern 1987).

Although legumes are capable of fixing atmospheric N, it largely depends on the effectiveness of available rhizobial bacteria strain in the soil, since certain bradyrhizobia strains nodulate only certain legumes (Ann *et al.* 2001). Therefore, studies on the effectiveness of locally available bradyrhizobium strains are very important. Some

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studies have revealed that exotic bradyrhizobia strains were more effective than indigenous strains in some countries. For example, Okereke et al. 2001 reported that, some exotic bradyrhizobia strains, were more effective than indigenous strains in soybean. On the other hand, cowpea is one of the most important food legumes in the tropics and subtropics, including Sri Lanka, due to its wide adaptability and ease of cultivation. Inoculation with effective and efficient Bradyrhizobium strains may have a positive role in increasing biological N fixation and yield of cowpea, if properly exploited. Unfortunately, in Sri Lanka, farmers are not exploiting the benefits of bradyrhizobia inoculation technology due to lack of research on the inoculation and non availability of inoculum to farmers.

The following series of experiments were carried out with these objective of determining the effectiveness of indigenous bradyrhizobial population in meeting the N requirement of cowpea; studying the impact of N fertilization on BNF,

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growth and yield of some local and introduced cvs. of cowpea, and estimation of BNF by employing the ¹³N methodology.

MATERIALS AND METHODS

A series of two field experiments and a pot experiment were conducted at the Seed Farm of the Department of Agriculture, Bata-atha, Hambantota in the dry zone of Sri Lanka, during the period November 1998 to August 1999. Experiment 1 (field experiment) was conducted to determine the effect of inoculation and N fertilizer application on growth and seed yield of three varieties of cowpea i.e. MI 35, Bombay and Arlington. A peat-based indigenous bradyrhizobium multi-strain inoculum was used to inoculate seeds. Three treatments;

- i. control (no fertilizer N, no inoculation)
- ii. inoculation (inoculated with above strains)
- iii. fertilizer N (80 Kg N ha^{-1})

were used in this experiment.

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In experiment 2 (field experiment), N fixing capacity of the above three cowpea varieties were studied. *Setaria italica* was used as the reference crop (Control). In micro-plots, for the determination of N fixation, 20 kg ha⁻¹ of 5% ¹⁵N labelled Ammonium Sulphate was used, while rest of the plot, received 20 kg ha⁻¹ of normal Ammonium Sulphate.

In experiment 3 (pot experiment), ten cowpea genotypes i.e.; Arlington (V1), Bombay (V2), MI 35 (V3), leta (V4), CP 230 (V5), CP 889 (V6), CP 789 (V7), IT-82D - 889 (V8), IT-82 E-32 (V9) and IT-82D-787 (V10) were grown at two N regimes; (80 kg N ha⁻¹ and without N). The experimental design used for all experiments was a randomized complete block design (RCB) with 4 replications. cvs. MI 35, Bombay, leta and Arlington were the recommended cvs. of cowpea by the Department of Agriculture, Sri Lanka and the rest were introduced genotypes from IITA, Nigeria. Each cultivar was planted along 2m long rows with 50cm x 12cm spacing between and within rows. Fertilizer was applied in split doses; 40 Kg N ha⁻¹ at planting and 40 Kg N ha⁻¹ (as ammonium sulphate) 2 weeks after planting.

In addition to N fertilizer, recommended doses of P as Concentrates super phosphate and K as Muriate of Phosphate by the Department of Agriculture were used for all experimental plots. All experiments were arranged in a randomized complete block design with 4 replications.

Fresh weight of nodules, % N by kjeldhal method (Bremner and Mulvaney 1982) and ¹⁵N atom

excess by mass spectrometry (Fiedler and Proksch 1975) were determined at 60 and 75 days after planting and seed yield and dry matter yield of stover were recorded at harvest.

RESULTS

It is evident from the results, that seed inoculation had no significant effect on yield improvement of cowpea cvs. In response to N fertilization, cvs. MI-35 and Arlington produced significantly higher yield (51% and 40% increase over control), while cv Bombay showed no significant response to fertilizer N but produced the highest yield, over other cvs, without fertilizer application (Table 1).

 Table 1 : Effect of seed inoculation and N fertilization on growth and yield of some cowpea cvs.

Cultivar	Treatment	Seed yield (kg ha ⁻¹)	Total Dry matter (kg ha ⁻¹)
MI 35	Control	1876 ^c	6078 ^{abc}
	Inoculated	1942 ^c	5260 ^c
	Fertilizer N	2836 ^{ab} (51)*	6500 ^{abc}
Arlington	Control	2209 ^{bc}	6993 ^{ab}
	Inoculated	2276 ^{bc}	5536 ^{cb}
	Fertilizer N	3098 ^a (40)*	7922 ^a
Bombay	Control	2462 ^{abc}	5818 ^{abc}
	Inoculated	2311 ^{bc}	5638 ^{abc}
	Fertilizer N	2662 ^{ab} (8)*	7587 ^a

* - % Increase over control

Means with the same letter are not significantly different ($P \le 0.05$)

This suggests that cv Bombay having originated in the Asian region, may have formed effective symbiosis with native strains present in soil in meeting its N requirement. Arlington and MI 35, being introduced cvs, the local bradyrhizobia compatibility and effectiveness may not be present adequately in the soil. Vincent et al (1979) reported that poor growth of legumes could be attributed to the absence of specific rhizobium strains. Salema and Chowdhury (1980) observed that local varieties of soybean did not benefit from rhizobia inoculation, whereas indigenous soybean rhizobia in the soil were abundant. The fact that the indigenous bradyrhizobia did not appreciably increase the nodule number in un-inoculated plants demonstrated that indigenous bradyrhizobia were not efficient in nodulating soybean cultivars.

In the next experiment, Nitrogen fixing capabilities of M1 35 and Arlington were estimated and the amount of % N derived from air (%Ndfa) and the comparable amounts of N_2 fixed in kg ha⁻¹ are given in Table 2.

The cv Bombay had significantly higher N

Table 2: Effect of variety on % Ndfa and N fixation of cowpea

Cowpea cultivars	% Ndfa	N ₂ fixed (kg ha ⁻¹)
MI 35	22 ^a	19 ^a
Bombay	48 ^b	54b
Arlington	18 ^a	18 ^a

Means with the same letter are not significantly different ($P \leq 0,\,05$)

fixing ability than MI 35 and Arlington. Probably, Bombay may have a higher affinity towards local strains and thus make effective symbiotic relationship, through which higher amounts of N could be fixed, resulting in a poor response to fertilizer-N. In the next experiment, prospects for yield improvement through enhanced BNF was studied and a considerable genotypic variability in nodulation was evident from the results (Fig 1).

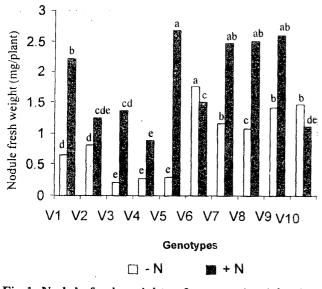


Fig 1. Nodule fresh weight of cowpea (mg/plant) as affected by genotype and N level Means with the same letter on the column are not significantly different ($P \le 0.05$)

Cultivars Bombay and leta showed a higher nodulation potential and Arlington, medium nodulation, while nodulation of cv MI 35 was the lowest. From the introduced genotypes, IT 82 D 889, IT 82 E 32 and IT 82 D 787 showed higher nodulation over other genotypes at 60 days after planting (DAP). But, at 75 DAP, CP 230, CP 889 and CP 789 showed superior nodulation among genotypes.

The total dry matter production and pod yield were significantly higher in response to N fertilization in many of the genotypes and at the sametime a high degree of variability was observed between genotypes; the total dry matter production was in the range of 2.59 - 4.82 g per plant without fertilizer N and 4.79 - 9.35 g per plant with fertilizer N (Table 3), while seed yield was in the range of 0.10 - 2.7 g per plant with fertilizer N (Fig.2).

 Table 3 : Total dry matter production of cowpea as affected by genotype and N levels

Cowpea genotype	Total Dry matter Production (g/plant)					
	-N at 42 DAP	+N at 42DAP	-N at PM	+N at PM		
VI	0.9 ^{ed}	1.35 ^{dc}	3.1 ^c	6.95 ^d		
V2	1.4 ^{cb}	2.25 ^b	4.0 ^b	6.12 ^{ed}		
V3	0.8^{e}	1.5 ^{dc}	2.6 ^c	4.45 ^{ed}		
V4	1.0 ^{ed}	3.65 ^a ∘	2.64c	5.95 ^{ba}		
V5	1.16 ^{cd}	1.2 ^{dc}	4.22 ^b	9.16 ^{ed}		
V6	1.2^{cd}	1.05 ^d	4.45 ^b	5.58 ^{bc}		
V7	1.0 ^{ed}	1.48 ^{dc}	5.8 ^a	8.22 ^{ed}		
V8	1.08 ^{ed}	1.58c	2.48 ^c	5.45 [°]		
V9	1.16 ^b	1.8 ^C	4.07 ^b	9.18 ^e		
V10	2.42 ^a	2.4 ^b	4.18 ^b	5.65 ^a		

PM - physiological maturity: DAP - days after planting Means with the same letter are not significantly different ($P \le 0.05$)

The cv Bombay cowpea showed some response to fertilizer-N in terms of dry matter production and seed yield, although there was no response in the previous experiment. This difference could be probably due to some seasonal effects since two experiments were conducted at two different seasons. The cvs MI - 35 and Arlington failed to establish a successful symbiotic relationship and this shows high response to fertilizer - N, as revealed from the results of previous experiment. Therefore it was clear that the response of nodulation in legumes to applied nitrogen varies with the species, thus no conclusions can be made on the optimum level of N for legume. The introduced genotypes seemed high late fixers, while the local cvs were early fixers (Fig. 2).

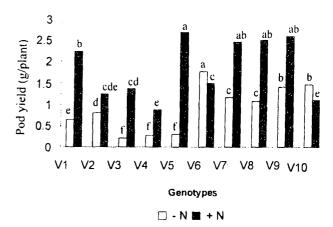


Fig 2. Pod yield of cowpea as affected by genotype and N application Means with the same letter on the column are not significantly different ($P \le 0.05$)

The highest and lowest response to fertilizer-N were observed in genotypes CP 230 and CP 889 respectively for seed yield.

CONCLUSIONS

High genotypic variability was found in nodulation, BNF, total dry matter production and seed yield among cowpea cvs. Locally recommended cowpea cvs. Bombay cowpea and leta showed better nodulation capacity over introduced genotypes. N fertilizer inhibited nodulation in almost all cvs. The total dry matter production and seed yield responded positively to N fertilization and the effect was particularly prominent among introduced genotypes. Most of the introduced genotypes were superior in dry matter production and yield performance compared to local cultivars, especially with fertilizer -N.

REFERENCES

Ann, M. Harsch, Michelle R. Lum, and J. Allan Downie (2001). What makes the Rhizobia legume symbiosis so special, Plant physiology, December 2001, Vol. 127 pp. 1484-1492.

- Bremner, J.M., Molvancy, C.S. (1982) Nitrogen total. In: Miller RH, Kenney DR (eds). Methods of Soil analysis. Am Soc Agron, Madison, Wisconsin, pp 595-622.
- Fiedler, R., Proksch, G. (1975). The determination of N-15 by emmission and mass sprectometry in biochemical analysis: A review. Anal Chim Acta 78: 1-62.
- Ofori, F., Stern, W.R. (1987) Cereal- legume intercropping systems. Adv Agron 41: 41-90.
- Okereke, G.U. Onochie, C. and Onunkwo, A. (2001). Effectiveness of foreign bradyrhizobia strains in enhancing nodulation, dry matter and seed yield of soybean (Glycine max L) cultivars in Nigeria. Biol. Fertil. Soils (2001) 33: 3-9.
- Salema, M.P. Chowdhury M.S. (1980) Rhizobia inoculation and fertilizer effect of soybean in the presence of native rhizobia. Beir Trop Landwirtseh Veterinaermed 18:245-250.
- Vincent, J..M. Nutman, P.S. Skinner, F.A. (1979). The identification and classification of rhizobium. In: Skinner FA lovelock DW (eds) Methods for microbiologists (society for applied bacteriology, Technical series No 14) Academic press. London.