

Field Response of L-Amino acid Micro-Mineral Complex (AAMMC) on Soybean *Rhizobium* Symbiosis

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A field experiment was conducted to evaluate the response of L-amino acid micro-mineral complex (AAMMC) on soybean *Rhizobium* symbiosis at instructional cum research farm, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.) with six treatments of different combinations (T1-T6) including control. Data revealed that treatment with recommended dose of fertilizers showed highest value among all the parameters followed by T3 i.e. foliar application of AAMMC @1L/ha 1st spray at 20-25 DAS and 2nd spray at 3-4 days before flower initiation. This treatment has highest number of effective nodules (54.5/plant), total N uptake (192.27 kg/ha) and grain yield (25.68 q/ha). However, maximum bacterial population (5×10^4 per g soil) and rhizobial population (4.59×10^4 per g soil) were observed in case of T4 i.e. foliar application of L-amino acid micro-mineral complex @1L/ha 1st spray at 20-25 DAS and 2nd spray at 3-4 days before flower initiation. The treatment T4 also showed maximum dehydrogenase activity (61 μ g TPF/h/g soil) at 50% flowering stage which might be due to the highest microbial population.

Ke ywords: Soybean, Amino acid, Micro-mineral complex, foliar spray.

Soybean (*Glycine max*) also known as Golden Bean is the largest oilseed crop in world accounting for more than 50% of the world oilseeds production. Above 80% of the global soybean output is crushed worldwide to obtain oil and meal. Its oil is widely used as edible oil whereas its meal is mainly used in animal feed industry. Further, biological nitrogen fixation in soybean is highly sensitive to soil drying¹. The soil moisture and temperature highly affect the legume-*Rhizobium* symbiosis².

Furthermore, the application of amino acids is based on its requirement by plants at critical stages of growth in particular. Plants absorb amino acids and it is proportional to environment

temperature. Amino Acids are basic ingredients in the process of protein synthesis. Importance of amino acids and micro-minerals as foliar spray to improve plant growth and crop output is recognized worldwide^{3,4,5}. An interesting research paper published by Fard *et al.*⁶ states the reduction of drought damage in plants by using of bio fertilizers as plant growth promoting bacteria, silicic acid and amino acids and improving physiological parameters and thus raising the level of plant yield in arid and semi arid areas are emergency management for drought control agriculture and also in wheat agronomy.

MATERIALS AND METHODS

For the of study effects of foliar spraying of amino acid on yield of soybean-*Rhizobium* symbiosis, an experiment was carried out in 2013-14 on the base of randomized complete block

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design with six treatments and five replications (Table 1) at Instructional cum Research Farm (Bharri land), College of Agriculture, Raipur (Chhattisgarh) located in 21° 16' N latitude and 81° 36' E longitude with an altitude of 298.56 M above the mean sea level.

Five plants were sampled were randomly from the field of each plot at 50% flowering stage. The whole plants were carefully uprooted using a spade so as to obtain intact roots and nodules for nodulation parameter and dry weight of plants. Uprooting was done by the exposing the whole root system to avoid loss of nodules. The adhering soil was removed by washing the roots with intact nodules gently with water over metal sieve. The same five plants from each plot were used to rate nodulation and to record the number of nodules per plant, and nodule per plant.

Define proportion of deferent L-amino acids along with micro mineral Complex (Table 2) was used for foliar application as per the treatments. In maturity time, studied traits were seed yield. The nitrogen content in the plant samples was estimated by mico-kjeldahl method as described by Jackson⁷ using auto digestion and distillation system. Microbial analysis of soil was done by serial dilution followed by pour plate method⁸. The procedure to evaluate the dehydrogenase activity of soil described by Lenhard. All the pre and post harvest observations were recorded and tabulated in a systemic manner and the data were analyzed using OPSTAT software from Hisar Agriculture University.

RESULTS AND DISCUSSION

Effect of AAMMC foliar spraying with attention to results of different parameters is varies treatment to treatment. The results obtained in present investigation are as follows:-

Plant biomass

At 50% flowering stage variation in fresh weight of shoot was observed from 18.732 to 28.033g different treatments (Table 3). In T1 lowest fresh weight of shoot was recorded i.e. 18.732g, while 28.033g was considered as highest which was associated with T5. Among the treatments associated with AAMMC the highest fresh weight was observed in T3 (22.533g). Similarly dry weight of shoot varies from 3.625 to 5.10 g different

treatments. The dry weight of shoot T1 (3.625g) was recorded as lowest while 5.10g was considered as highest. Among the treatments associated with amino acid micro-mineral complex the highest dry weight was observed in T3 (4.393g).

Sawan *et al.*,¹⁰ and Chellaiah *et al.*¹¹, has also reported that foliar spray of micro nutrient increased the plant height and dry matter production. These observations are in line with that of Tomar¹², Khutate *et al.*¹³, Azeez and Adetunji¹⁴, Ghose *et al.*¹⁵ and Wasule *et al.*¹⁶, who mentioned that legume plants raised from different levels of fertilizer showed significantly higher plant fresh and dry weight over control.

Nodulation study

Data of nodulation study (Table 3) indicated that highest number of nodules (50% flowering stage) was associated with plants raised with treatment T5 followed by T6. No. of nodules was vary from 50.2 to 73.9 per plant. Number of nodules 73.9 per plant was recorded as highest among all the nodulated plants, under field study. There was lowest number of nodule in T1. Among the treatment associated with amino acid micro-mineral complex the highest nodulation found in T3 (54.5/ plant).

Data revealed that nodule N-content vary from 2.10 to 2.85% due to different treatment. Nodule N-content 2.85% per plant was recorded as highest among all the nodulated plants, under field study. There was lowest N content of nodule in T1. Among the treatment associated with amino acid micro-mineral the highest N content was found in T4 (2.76%).

Nodule N-uptake at 50% flowering stage (Table 4) significantly increased. The N-uptake values vary from 3.99 to 6.70 mg per plant due to different treatments. Lowest nodule N-uptake value 3.99 mg/plant were associated with T1 while highest nodule N-uptake value 6.70 mg/plant were associated with inoculated and 100% RDF plots (T5). Among the treatment associated with amino acid micro-mineral complex the highest N-uptake found in T3 (5.94 mg/ plant).

Findings of the present investigation are close to observations of Zhang *et al.*¹⁷, who reported that significantly increased N accumulation due to treatment with different fertilizers levels. Similar findings were also reported by Prasad and Ram¹⁸, Alagawadi *et al.*¹⁹ and Quasim

et al²⁰. They mentioned that number of nodules and biomass can be increased due to different levels of fertilizer and rhizobial inoculation.

Dehydrogenase activity of soil

At 50% flowering stage dehydrogenase activity (Figure 1) of soybean rhizosphere soil shows variation from 48 to 65 µg TPF/hour/g soil due to different treatments. Lowest DHA 48 µg TPF/hour/g soil were associated with T6 while highest 65 µg TPF/hour/g soil was associated with inoculated and 100% RDF plots (T5). Among the

treatment associated with amino acid micro-mineral complex the highest DHA found in T4 (61 µg TPF/h/g soil). Chendrayan et al.²¹ gave the opinion that the increase in dehydrogenase activity was mainly due to the higher microbial population. Result shows that with increase in AAMMC the microbial activities increases.

Population dynamics study of *Rhizobium*

At 50% flowering stage, rhizobial population density was recorded (Table 4, figure 1) in between 5.02×10^4 to 7.65×10^4 per gram of

Table 1. Treatment details of field experiment conducted in the present study

T1	Foliar application of L-amino acid micro-mineral complex @ 1L/ha 20-25 DAS
T2	Foliar application of L-amino acid micro-mineral complex @ 2L/ha 20-25 DAS
T3	Foliar application of L-amino acid micro-mineral complex @ 1L/ha 1 st spray at 20-25 DAS and 2 nd spray at 3-4 days before flower initiation
T4	Foliar application of L-amino acid micro-mineral complex @ 2L/ha 1 st spray at 20-25 DAS and 2 nd spray at 3-4 days before flower initiation.
T5	RDF (recommended dose of fertilizer) with same amount of water.
T6	Control.

Table 2. Composition of L-amino acid micro mineral complex (AAMMC) used in the study

Components of Amino acid micro mineral complex (AAMMC)	Percent
free amino acids (p/p): L-tryptophan, L-aspartic acid, L-glutamic, L-acid serine, L-histidine, L-glycine, L-threonine, L-alanine, L-proline, L-tyrosine, L-agrinine, L-valine, L-methionine, L-isoleucine, L-leucine, L-phenylalanine, L-lysine	12%
Ultra soluble minerals (mainly) Ca and Mg (p/p):	6%
Chitin and its derivatives (p/p): Chitosan (poly-D-glucosamine)	1%
Glucosamine (D-glucosamine)	1%
Minerals	
Nitrogen %N	0.95%
Phosphorus %P	0.14%
Potash %K	0.75%

Table 3. Effect of amino acid micro-mineral complex on different parameters of field grown soybean

Treatment	Fresh weight of shoot (g/plant)	Dry weight of shoot(g/plant)	Grain yield q/ha	No. Of nodule /plant
T1	18.732	3.625	23.24	50.2
T2	20.194	4.032	22.76	52.3
T3	22.533	4.393	25.68	54.5
T4	19.051	3.665	23.48	50.7
T5	28.033	5.10	28.08	73.9
T6	20.975	3.645	21.28	55.6
C.D.	3.173	0.468	2.04	7.9

soil. Similarly at harvest it was recorded in between 3.36×10^4 to 4.87×10^4 per gram of soil due to different treatments. Keeping in view of estimated rhizobial population per gram of soil by dilution planting method, T5 showed the highest population density followed by T4. However, in case of un-inoculated control the population recorded per g soil at the above stage.

Similar type of results were also reported by Chowdhury²², reported that rhizobial population increased considerably up to flowering stage of crop growth due to higher degree of rhizosphere effect. These observations are also in close agreement with Gupta et al²³.

Grain yield

The grain yield ranges from 21.28 to 28.08 q/ha due to different treatments. Where control plot gave lowest grain yield (21.28 q/ha) while

highest grain yield (28.08 q/ha) was associated with T5 (100% RDF with rhizobial inoculation) Followed by T3 (25.68 q/ha). (Table 3)

Grain N- content (Table 5, figure 2) vary from 5.65 to 6.31, followed by 6.29 % similarly, straw % N content also Increased significantly due to application amino acid with effective homologous native Rhizobium. The straw N content increased significantly from 1.036 to 1.347 followed by 1.288 %. In most case, the application of amino acids led to decreased nitrate content and increased total nitrogen content in several crops^{24,25,26,27}. Castro and Boaretto²⁸, using nutrients in the treatment of seeds and via foliar obtained similar results to those found in this study. A similar result was obtained by Lima et al²⁹ when testing the foliar application of micronutrients on common bean. Findings of the present investigation are also close to

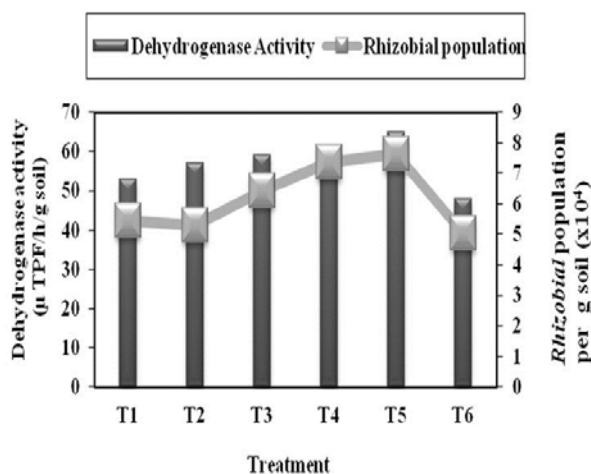


Fig. 1. Effect of amino acid micro-mineral complex on dehydrogenase activity and Rhizobial population

Table 4. Effect of amino acid micro-mineral complex on N content of field grown soybean (at 50% flowering)

Treatment	N uptake by nodule (mg/plant)	Straw N content (%)	N content of nodule (%)	Grain N content (%)
T1	4.24	1.064	2.10	6.12
T2	5.32	1.176	2.69	6.26
T3	5.94	1.134	2.57	6.29
T4	4.44	1.288	2.76	6.31
T5	6.70	1.347	2.85	6.23
T6	3.99	1.036	2.51	5.65
C.D. (0.5)	1.03	0.127	0.366	NS

observations of Tomar et al.¹², Tripathi et al.³⁰, and Tomer and Khajanji³¹. They clearly mentioned that grain and straw yield of soybean significantly increased by use of different levels of fertilizer.

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REFERENCES

- Ladrera, R., Marino, D., Larrainzar, E., Gonzalez, E.M. and Arrese, Igor, C. Reduced carbon availability to bacteroids and elevated ureides in nodules, but not in shoots, are involved in the nitrogen fixation response to early drought in soybean. *Plant-Physiology*. 2007; **145**(2): 539-546.
- Pareek, R.P., Kumar, N. and Chandra, R. Performance of chickpea Rhizobium under various hydrothermal regimes in soil. *J. Indian Soc. Soil Sci.* 1990; **38**: 168-188.
- Patil, S.N., Ulemale, R.B., Lande, S.S. and Mahajan, A.M. Phosphorus management in soybean (*Glycine max* L Merrill)- sunflower (*Helianthus anus* L.) cropping system. *Oilseed Research*. 2004; **21**:62-64.
- Abdel-Mawgoud, A.M.R.; A. M. El-Bassiouny; A. Ghoname, S.D. Abou-Hussein Foliar Application of Amino Acids and Micronutrients Enhance Performance of Green Bean Crop under Newly Reclaimed Land Conditions. *Australian Journal of Basic and Applied Sciences*, 2011; **5**(6): 51-55.
- Kumar, A. and Omae., H. Estimation of seed yield and yield attributes by cumulative temperature in common bean (*Phaseolus vulgaris*). *Indian Journal of Agril. Sciences*. 2008; **78**(2): 127-130.
- Fard, M. D., Habibi D., and Fard F.D. Effect of Plant Growth Promoting Rhizobacteria and Foliar Application of Amino Acids and Silicic Acid on Antioxidant Enzyme Activity of Wheat under Drought Stress International Conference on Chemical Engineering and Applications IPCBEE IACSIT Press, Singapore 2011; **23**.
- Jackson, M.L. Soil Chemical Analysis. Pub. Prentice Hall of india (Pvt.) Ltd. New Delhi 1973.
- Subba Rao, N.S. Biological Nitrogen fixation. Oxford and I.B.H. Pub. Co., New Delhi. 1988.
- Lenhard, G. Die Dehydrogenase activital des Bodens als mass fur die mikroorganismen-tatigkeit im Boden. *Z.P flanzene nahr Dueng. Bodenkd.* 1956; **73**: 1-11.
- Sawan, Z.M., Gregg, B.R., Yousef, S.F. Effect of phosphorus, chelated zinc and calcium on cotton seed yield, viability and seedling vigour. *Seed Science and Technology*, 1999; **27**(1): 329-337.
- Chellaiah, N., Solaiappan, U. and Senthivel, S. Studies on foliar nutrition on productivity of summer irrigated cotton. *Madras Agriculture Journal*, 2001; **88**(1-3): 180 -181.
- Tomar, S.S., Singh, R. and Singh, S.P. Response of phosphorus, sulphur and Rhizobium inoculation on growth, yield and quality of soybean (*Glycine max* L.). *Progressive Agriculture*. 2004; (1): 72-73.
- Khutate, N.G., Mendhe, S.N., Dongarkar, K.P., Gudadhe, N.N. and Gavande, V.H. Effect of nutrient management treatments on growth and yield of soybean. *Journal of Soil and Crops*. 2005; **15**(2): 411-414.
- Azeez, J.O. and Adetunji, M.T. 2008. Interactive effects of nitrogen and phosphorus on nodulation, yield and nutrient uptake of soybean (*Glycine max* L. Merrill). *International Journal of Tropical Agriculture*. 2008; **26**(3-4): 457-461.
- Ghose, P.K., Bandyopadhyay, K.K. and Singh, A.B. Effect of integrated nutrient management on dry matter production, water use efficiency and productivity of soybean (*Glycine max* L. Merrill) in Vertisols of Central India. *Journal of Oilseed Research*. 2005; **22**: 289-292.
- Wasule, D.L., Wadyalkar, S.R., Burgoni, E.B. and Buldeo, A.N. Effect of phosphate solubilizing bacteria on role of Rhizobium on nodulation of soybean. *Annals of Plant Physiology*. 2002; **16**(1): 69- 72.
- Zhang, F., Dashti, N., Hynes, R.K. and Smith, D.L. Plant growth promoting Rhizobacteria and soybean (*Glycine max* (L.) Merr.) nodulation and nitrogen fixation at suboptimal root zone temperatures. *Annal of Botany*. 1996; **77**: 453-459.
- Prasad, J. and Ram, H. Effect of zinc and copper and Rhizobium inoculation on nodulation and yield of green gram. *Indian J. Agril. Sci.* 1986; **58**: 230-233.
- Alagawadi, A.R., Siddaramegowda, T.K. and Habib, A.F. Groundnut crop response to Rhizobium inoculation. *Madras Agril. J.* 1993; **70**(10): 635-637.
- Quasim, M., Nawab Fazal, Himaytulla and Rahim Din. Response of inoculation and fertilizers on nodulation on economic yield of soybean cultivars. *Journal of Biological Science*. 2001; **1**(2):76-77.

21. Chendrayan, K., Adhya, J.K. And Sethunathan, N. Dehydrogenase and invertase activities of flooded soils. *Soil Biol. Biochem.* 1980; **12**: 271-273.
22. Chowdhury, T. & Gupta, S.B. Selection of effective Rhizobium isolates for kharif groundnut (*Arachis hypogaea* L.) under climatic conditions of chhatisgarh region. *J. Soils & Crops.* 2003; **13**(2):225-230.
23. Gupta, S.B., Rawat, A.K. and Khare, A.K. Effect of pre and post -inoculation seed treatment with fungicide on nodulation and grain yield of soybean. *Legume Res.* 1988; **11**(4):167-172.
24. Prasad, J. and Ram, H. Effect of zinc and copper and Rhizobium inoculation on nodulation and yield of green gram. *Indian J. Agril. Sci.* 1986; **58**: 230-233.
25. Gunes, A., A. Inal, and M. Aktas. Reducing nitrate content of NFT grown winter onion plants (*Allium cepa* L.) by partial replacement of NO₃ with amino acid in nutrient solution. *Sci. Hortic.* 1996; **65**: 203–208.
26. Gunes, A., W. H. K. Post, E. A. Kirkby, and M. Akas. Influence of partial replacement of nitrate by amino acid nitrogen or urea in the nutrient medium on nitrate accumulation in NFT grown winter lettuce. *J. Plant Nutr.* 1994; **17**: 1929–1938.
27. Chen, G., and X .Gao. Effect of partial replacement of nitrate by amino acid and urea on nitrate content of nonheading Chinese cabbage and lettuce in hydroponics (Chinese). *Sci. Agr. Sinica.* 2002; **35**: 187–191.
28. Wang, H., L. Wu, and Q. Tao. Influence of partial replacement of nitrate by amino acids on nitrate accumulation of pakchoi (*Brassica chinensis* L.) (Chinese). *China Environ. Sci.* 2004; **24**: 19–23.
29. Castro, A. M. C. and Boaretto, A. E. Adubação foliar do feijoeiro com nutrientes, vitamina B1 e metionina. *Scientia Agraria.* 2001; **2**: 117-121.
30. Lima, S. F; Andrade, M. J. B; Carvalho, J. G. de. Resposta de feijoeiro à adubação foliar de boro, molibdênio e zinco. *Ciência e Agrotecnologia.* 1999; **23**: 462-467.
31. Tripathi, R.K., Tomar, I.S., Mishra, S. and Vani, D.K. Effect of integrated nutrient management on growth and yield of soybean in Jhabua Hills Zone of Madhya Pradesh. *Soybean Research.* 2008; **6**: 44-48.
32. Tomer, G.S. and Khajanji, S.N. Effect of organic manuring and mineral fertilizer on the growth, yield and economics of soybean [*Glycine max* (L.) Merrill]. *International Journal of Agricultural Sciences.* 2009; **5**(2):590-594.