Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

Inoculation Effect of Arbuscular Mycorrhizal Fungi on the Growth and Yield of Cotton

G. Kumaresan[1], J. Jayachitra[1] and R. Shanthi[2]
1. Assistant professors,Department of Agricultural Microbiology,
Faculty of Agriculture, Annamalai University, Annamalai Nagar, Tamil Nadu, India-608002

2. Assistant professor, Department of Mathematics, Faculty Science, Annamalai University,
(Deputed to Department of Mathematics, PSPT MGR Govt. Arts and Science College,Puthur-Sirkazhi, Tamil Nadu – 609108.)
Mail ID: microsen1974@gmail.com

ABSTRACT

A pot culture experiment was conducted in the Department of Microbiology, Faculty of Agriculture at Annamalainagar to study the effect of*Glomusfasciculatum* on the growth and yield of MCU-7 cotton as influenced by different levels of N and P *viz*., 25,50,75 and 100 per cent recommended dose (RD) with Arbuscular – mycorrhizal fungi (AMF) and 100 per cent N and P without AM fungi (control) were maintained. Among the different treatments, 75 per cent P with 100 per cent N+ AM fungi was observed with better yield and P up take in cotton and maximum mycorrhizal colonization and spore number were recorded at 50 per cent P and 100 per cent N RD + AM fungi followed by application of 50 per cent P with 100 per cent N RD+AM fungi. Hence, this study revealed that 25 per cent of phosphatic fertilizer could be saved by the inoculation of *G.fasciculatum* in cotton.

Keywords: *Glomus fasciculatum*, AM fungi, cotton, recommended dose (RD)

INTRODUTION

Cotton is the king of fibers, which provides the basic material for the clothing of man. It is a vital and basic input for the textile industry and one of the world's most important industrial and commercial crops. India holds the maximum area (9. 16 m ha with a production of 17. 17 million bales) under cotton. To meet the future requirement the current production has to be increased in largely and this increase has to come from increased productivity alone by the way of sustainable agriculture. As the importance of sustainable agricultural practices and increased role of arbuscular mycorrhizal symbiosis in contributing to sustainability has also been recognized.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

The large use of chemical fertilizers has a serious impact on the environment (Tilman *et al.*, 2002) and the agricultural practices influence soil microorganisms greatly, decreasing soil fertility and organic matter turnover (Altieri, 1999). However, the more crucial issue for modern agriculture is that the natural reservoir of some nutrients as phosphorus (P) is decreasing in the world, leading to increase in fertilizer prices in the last decade (Cordell *et al*., 2009). The challenge for crop production is change to sustainable practices, by finding alternatives for increasing nutrient availability for plant nutrition as organic fertilization. Some these alternatives for organic fertilization include the use de soil microorganisms (Miransari, 2011). Soil microorganisms play an important role by contributing significantly to nutrient availability through biochemical transformations. Some of these microorganisms act directly on plant nutrition by establishing symbiotic associations with plant roots (Bardgett, 2005). The symbiosis between arbuscular mycorrhizal fungi (AMF) and plant roots is one of the most known beneficial interactions occurring in soil (Smith and Smith, 2011), playing an important role in crop production and nutrient turnover (Andrade, 2004).

The symbiotic association established between plant roots and fungi belonging tothe Endogonaceae , known as Arbuscular mycorrhizae fungi (AM fungi) can significantly improve the plant growth and development due to enhanced uptake of soil phosphorus and certain other nutrients.AM fungi are non-specialized host range, yet are apparently obligate endosymbionts. They depend on their host plants for by the energy source (carbohydrates). They could be recognized by the irregular coenocytic hyphae, which ramify within the cells of root cortex and form vesicles (storage pouches) and arbuscules (absorptive structures). The later are finely branched to help in the absorption of nutrients. The various mechanisms proposed to account for this increased nutrient uptake includes physical exploration of soil, increased translocation of P into mycorrhizal hyphae and modification of root environment.

The objective of this work was to determine the effectiveness of AMF (*Glomus fasciculatum*) inoculation in cotton (*Gossypium hirsutum* L.), assessing its effect on plant growth, nutrient uptake and yield when compared with conventional chemical fertilization under pot culture conditions. Our hypothesis is that AMF inoculation can be an alternative for total nutrient supply or more effective nutrient absorption, when combined with chemical fertilization.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

MATERIALS AND METHODS

AMF Inoculum Production

In this experiment the Tata Energy Research Institute in India (Gaur, 1997) developed technique was followed for inoculum production of AM fungi. In this method, starter inoculum of AM fungi was mixed into furrows in raised beds of fumigated soil. The sorghum crop was grown successively in the bed for three months. The soil and roots were chopped and remixed prior to planting the next nurse host. This method produced an average of 22.7 infectious propagules cm-3 after three one-year cycles. A modification of this method using sorghum crop as a host plant produced approximately one propagule cm-3 in soil amended 1:1 or 1:2 (v/v) with compost (Gaur *et al.*, 2000). Petri dishes with massive growth (mycelia and spores) of *Glomus fasciculatum* and colonized roots were used as crude inoculum.

Pot culture experiment

Cement pots (50 x45x30 cm sizes) were used to conduct the pot culture experiments. In the cement pots, clay loam soil of pH 7.3, available N 145Kg ha^{-1}, $P_2O_5$16.5Kg ha^{-1} and K_2O 215 Kg ha^{-1} was filled @15Kg soil per pot. The dose of manures and fertilizers per pot were worked out on the soil weight basis keeping the base of their recommended doses *viz*., 60kg N, 30kg P_2O_5,30kg K_2O and 12.5 t of FYM ha^{-1} for MCU-7 cotton. The half dose of N and full dose P_2O_5 and K_2O were applied as basal application. Remaining N was top – dressed on 45^{th} days after sowing (DAS). The N, P_2O_5 and K_2O through urea, super phosphate and muriate of potash were used respectively. The method of inoculation of arbuscular - mycorrhizal fungal culture was by placementmethod @ 25 gram of soil inoculum per plant. The cotton (MCU -7) seeds were sown @ 4 seeds per pot and suitable control was also maintained. The experiment was laid out in completerandomized block design (CRBD) with three replications.

Estimation of AM fungi root colonization

Fresh samples of soil were taken to the laboratory. Fine roots were fixed in the formalin acetic acid alcohol solution (90:5:5) after washing thoroughly to determine the root infection. Soil samples were dried air in the shade for additional spores at laboratory temperature. Roots were autoclaved in KOH solution for 15 to 20 minutes (10 per cent), cleaned in distilled water

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

and neutralized with HCl (2 per cent) and stained in lacto phenol in trypan blue (0.05 per cent). Phillips and Hayman's (1970) method measured the percentage of the root infection.

Estimation of AM fungi spore population

100 g of air - dried soil mixture with 1000 ml of tap water has been placed in a beaker. The mixture of the root soil has been strongly mixed with glass rod for 30 seconds. The remaining soil-root-hyphae-spore suspension was slowly poured through 240, 170, 150,100 and 72 μm sieves after the soil particles and organic debris were settled. The extracts were washed off the sieves to what man filter paper. Spores, aggregates and sporocarps were picked by needle using trinocular research microscopes (Gerdemann and Nicolson, 1963). To each PVLG drop, 5 - 10 spores have been added. The mounting system was allowed to set for 3 – 5 minutes before a cover slip was added. The identification of isolated spores was carried out by Schenck and Perez (1990) using the key proposed

Statistical analysis

The experimental results were statistically analyzed as suggested by Gomez and Gomez (1984). For the significant results, critical difference was worked out at 5 per cent probability level.

RESULTS AND DISCUSSION

Application of P significantly increased the plant height and number of bolls, boll weight and seed yield up to 75 per cent recommended dose (RD) with Arbuscular – mycorrhizal fungi (AMF). Application of ‘p’ significantly increased the plant height and number of bolls up to 75 percent recommended dose (RD) with AM-fungi (Table1). Thereafter, RD reduced proportionately to the application. The maximum plant height (106.37cm), number of bolls per plant (22.3),20 boll weight (106.42 g) and seed yield (3.56tonnes ha^{-1}) were observed at 75 percent P with 100 percent N RD + AM-fungi followed by the application of N and P 100 percent RD + AMF at 120 DAS. This observation supports the earlier findings of Bagyaraj (1980). Torelli *et al*. (2000) and Boldt *et al*. (2011) reported that the enhanced photosynthates and phytohormones, which modulated by arbuscular mycorrhizal fungi, led to higher flower and fruit numbers in AMF-inoculated plants.AMF colonization promoted plant growth and reproductive growth. The maturity of the

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

cotton boll was accelerated, and the time at maturity was prolonged. Subsequently, the fiber maturity grade was high. The principal function of mycorrhizal is to increase the soil volume explored for uptake of phosphorus, nitrogen and potassium and other minor nutrients*viz*.., Zn, Fe, Cu and Mn. This phenomenon would have increased the growth and yield of cotton.

The effect of different levels of N and P application on the root colonization by AM- fungi and spore population in the rhizosphere of cotton are presented in the Fig. 1 and Fig. 2. The percent root colonization and AM fungal spore population increased with increased in the application of P from 25 to 50 percent RD. On the other hand, the highest root colonization (91.81 percent) and AM fungal spore population (188.6 per 100g of soil) were recorded at 120 DAS at 50 percent P with 100 percent N + AM fungi followed by 25 percent with 100 percent N+ AM fungi and minimum in the control (100 percent N and P RD without AM fungi). This is probably because the soil p concentration was still below the critical concentration of P to inhibit the mycorrhizal colonization and AM fungal spore population. The study revealed that the ‘p’ application highly reduced the root colonization percent and AM fungal spore number than N application. The same findings from Champawat (1990) and Ryan(1994) in peanut. Phosphorus also inhibits AMF colonization in roots by inhibiting the expression of plant symbiotic genes, especially genes encoding carotenoids and lactones biosynthesis enzymes, as well as symbiotic related phosphorus transporters (Breuillin*et al*.,2010). Therefore, lower levels of P may regulate the expression of P transporter family genes and then increase the trading ratio and the AMF inoculation speed. Therefore, this study is consistent with our research results.

CONCLUSION

The inoculum of *Glomus fasciculatum* evaluated was very competitive against endogenous AMF and also increased plant growth and yield of cotton. *G. fasciculatum* obtained in vitro and tested in the pot culture was efficient in starting early AMF infection in seedlings, improving AM colonization in cotton. The AM fungi inoculum of *G. fasciculatum* helped plants to take up P from fertilizer and showed high potential for use in combination with conventional fertilization, for intensive agriculture system in large areas in tropical soils, increasing P absorption and more efficient fertilization use, this is fundamental for the actual challenge of crops production. This study revealed that 25 per cent of phosphatic fertilizer could be saved by the inoculation of *G. fasciculatum* in cotton and prevent the root knot nematode problem.

ACKNOWLEDGMENTS:

Author is thankful to Head of the Department of Microbiology, Faculty of Agriculture, Annamalai University, Annamlainagar, Chidambaram for providing lab facilities.

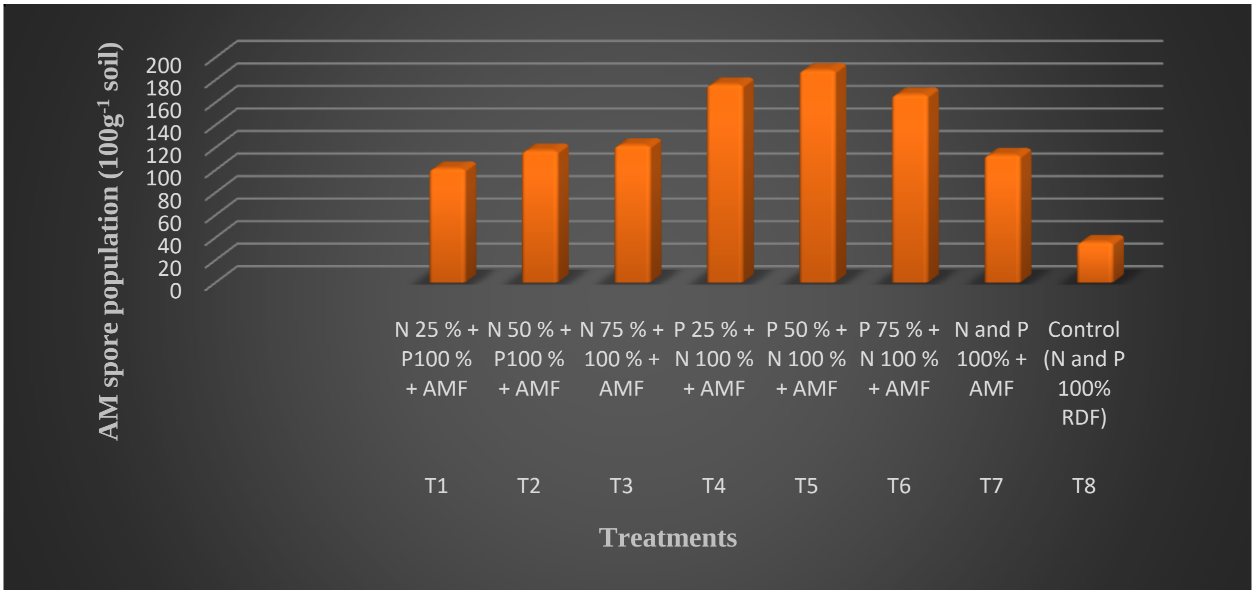

Fig. 1. Effect of *G. fasciculatum* inoculation on spore populationin cotton as influenced by the application of various levels of N and P

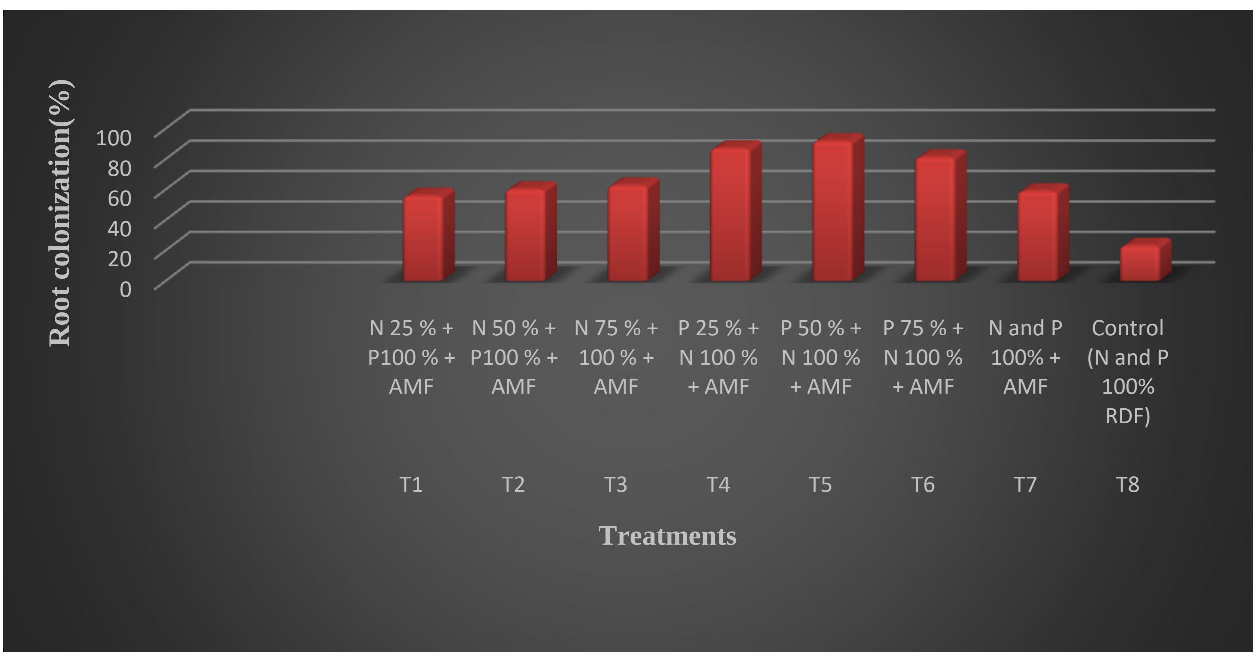

Fig. 2. Effect of G. fasciculatum inoculation on spore population in cotton as influenced by the application of various levels of N and P

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

Table1. Effect of *G. fasciculatum* inoculation on plant parameters and seed yield of cotton as influenced by the application of various levels of N and P

T. No.	Treatment	Plant height (cm)	No. of bolls per plant	Weight of 20 bolls (g)	Seed yield (tonnes ha^{-1})
		120 DAS			
T_1	N 25 % + P100 % + AMF	86.80	12.33	84.10	2.91
T_2	N 50 % + P100 % + AMF	95.10	15.00	95.37	3.13
T_3	N 75 % + 100 % + AMF	102.40	17.33	101.42	3.41
T_4	P 25 % + N 100 % + AMF	95.57	15.33	93.67	3.01
T_5	P 50 % + N 100 % + AMF	99.10	18.00	99.73	3.36
T_6	P 75 % + N 100 % + AMF	106.37	22.37	106.42	3.56
T_7	N and P 100% + AMF	105.57	20.33	103.10	3.48
T_8	Control (N and P 100% RDF)	103.20	18.67	101.47	3.33
	S. Ed.	**0.2448**	**0.3591**	**1.4501**	**0.0321**
	C.D.(P=0.05)	**0.6960**	**1.0208**	**3.2360**	**0.1120**

REFERENCES

1. Altieri, M. A. (1999). The ecological role of biodiversity in agroecosystems. Agric. Ecosyst. Environ. 74, 19–31.
2. Andrade, G. (2004). “Role of functional groups of microorganisms on the rhizosphere microcosm dynamics,” in Plant Surface Microbiology, eds A. Varma, L. Abbott, D. Werne, and R. Hampp (New York, NY: Springer), k51–68.
3. Bagyaraj, D.J. and Manjunath, A. 1980. Response of crop plants to VA-Mycorrhizal inoculation in an unsterile Indian soil. New Phytol., 85: 33-36.
4. Bardgett, R. D. (2005). “Organism interactions and soil processes,” in The Biology of Soil a Community and Ecosystem Approach, ed. R. D. Bardgett (New York: Oxford University Press), 57–85.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

5. Boldt, K. Y. Pors and B. Haupt. Photochemical processes, carbon assimilation and RNA accumulation of sucrose transporter genes in tomato arbuscular mycorrhiza. *J. Plant Physiol*. 168, 1256–1263 (2011).
6. Breuillin, F., Schramm, J., Hajirezaei, M., Ahkami, A. & Favre, P. Phosphate systemically inhibits development of arbuscular mycorrhiza in Petunia hybrida and represses genes involved in mycorrhizal functioning. Plant J. 64, 1002–1017 (2010).
7. Champawat, R.S. 1990. Effect of mycorrhizal inoculation and phosphorus on growth and nutrient of groundnut. Indian J. Agric, Sci., 60:425- 427.
8. Cordell, D., Drangert, J., and White, S. (2009). The story of phosphorus: global food security and food for thought. Global Environ. Change 19, 292–305
9. Gaur, A., A. Adholeya, and K. G. Mukerji. 2000. On-farm production of VAM inoculum and vegetable crops in marginal soil amended with organic matter. Tropical Agriculture 77: 21–26.
10. Gerdemann, J.W. and T.H. Nicolson, 1963. Spores of mycorrhizal *Endongone* species extracted fromsoil by wet sieving and decanting. *Trans. Br. Mycol. Sol.*, 235-244.
11. Gomez, K.A. and A.A. Gomez, (1984). Statistical procedures for agricultural research (2 ed.). John wiley and sons, NewYork, 680p.
12. Hayman, O.S. 1993. The physiology of VA - mycorrhizal symbiosis. Can. J. Bot., 61: 994- 999.
13. Jacksson, M.L. 1973. Soil chemical analysis. Prentice Hall of India Ltd., New Delhi.
14. Miransari, M. (2011). Soil microbes and plant fertilization. Appl. Microbiol. Biotechnol. 92, 875–885.
15. Olsen, S. R., C.V.Cole, F.S Watanabe and Dean L.A. 1954. Estimation of available P in soil by extraction with Na bicarbonate. USDA., Cir., 939.
16. Philips, J.M. and Haymann, D.S. 1970. Improved procedures for cleaning roots and staining parasiticand VA-mycorrhizal fungi for rapid assessment of infection. *Trans. Br. Myclo. Soc.*, **55:** 158-161.
17. Prathibha, C. K., A. R. Alagawadi and Sreenivasa. M. N.1994. Establishment of inoculated organisms in rhizosphere and their influence on nutrient uptake andyield of cotton. *Karnataka J. Agric.Sci*. **8(1)**: 22 – 27.
18. Ryan, M.H., G. A. Chilveres and Dumaresq, D.C. 1994. Colonization of wheat by VA-mycorrhizal fungi was found to be higher on a farm managed in an organic matter than on conventional neighbor. *Plant and Soil*,**160**: 33 – 40.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 24, Issue 2, 2020, Pages. 1180 - 1188
Received 24 October 2020; Accepted 15 December 2020.

19. Schenck and Perez 1990: Manual for the identification of V A Mycorrhizal fungi 3rd edition.
20. Smith, S. E., and Smith, F. A. (2011). Mycorrhizas in plant nutrition and growth: new paradigms from cellular to ecosystem scales. Annu. Rev. Plant. Biol. 62, 227–250.
21. Subbiah, B. V. and Asija, A.G.L.1956. A rapid procedure for estimation of available N in soil. *Curr. Sci.*,259 – 260.
22. Tilman, D., Cassman, K. G., Matson, P. A., Naylor, R., and Polasky, S. (2002). Agricultural sustainability and intensive production practices. Nature 418, 671–677.
23. Torelli, A. A. Trotta, L. Acerbi, G. Arcidiacono, G. Berta and C. Branca. IAA and ZR content in leek (Allium porrum L.) as influenced by P nutrition and arbuscular mycorrhizae, in relation to plant development. Plant Soil. 226, 29–35 (2000).