INTERNATIONAL JOURNAL OF RESEARCHES IN BIOSCIENCES, AGRICULTURE AND TECHNOLOGY
© VISHWASHANTI MULTIPURPOSE SOCIETY (Global Peace Multipurpose Society) R. No. MH-659/13(N)
www.vmsindia.org

CO-INOCULATION OF *AZOSPIRILLUM* AND ARBUSCULAR MYCORRHIZAL FUNGI IN DIRECT SEEDED RICE

S. C. Patil and G. P. Santhosh
Department of Agricultural Microbiology,
University of Agricultural Sciences, Raichur, Karnataka, India
sgp2222@rediffmail.com

Abstract:
Interaction effects of *Azospirillum* and arbuscular mycorrhizal fungi (AMF) with reduced levels of Nitrogen and Phosphorous on growth and yield of Direct Seeded Rice (DSR) was determined. Highest growth and yield were obtained with combined inoculation of *Glomus* + *Azospirillum* along with reduced (75%) levels of N and P compared to control and individual inoculations. Among individual treatments, *Azospirillum* was significantly superior over AMF at different nutrient levels. Population of *Azospirillum* and AM fungal infection was significantly higher in dual inoculated plants with 75% N and P. Nitrogen concentration in the plants was significantly increased by *Azospirillum* even more in association with the efficient fungal strain. Further, dual inoculated plants showed a significant increase in P uptake.
Keywords: *Azospirillum*, Arbuscular Mycorrhizal Fungi, Co-inoculation, Direct Seeded Rice

Introduction:

Oryza sativa L. is the second most important cereal in the world after wheat, and the principal staple crop in Asia, serving as food for 50% of the world's population. India is the second largest producer of rice next to China where it is grown in an area of 42 m ha annually with a production of 151 m t and accounts for about 45% of food grain production in the country.

Direct seeding rice, a common practice before green revolution in India, is becoming popular once again because of its potential to save water and labour. Currently, DSR in Asia occupies about 29 m ha which is approximately 21% of the total rice area in the region. In India, estimated DSR area is 11,900 ha and occupies 28% of total area of rice cultivation (Pandey and Velasco, 2002).

Rice requires high quantity of N and P for maximization of seed yield and the cost of N and P fertilizers is increasing day by day, hence it becomes imperative to substitute N and P by some other cheap sources which can partially meet the crop requirement. In such a situation soil microbes play a central role in maintaining soil quality by decreasing the dose of chemical fertilizers and increasing the crop production.

The present investigation was aimed at studying the co-inoculation of *Azospirillum* and arbuscular mycorrhizal fungi (AMF) in reducing the dose of chemical nitrogen and phosphorous on growth and yield of DSR.

Material and Methods:

Triplicate pot experiment was conducted to study the effect of inoculation of efficient *Azospirillum* {ADSR-9 (*A. brasilense*) and ADSR-23 (*A. lipoferum*)} and AMF {MDSR-3 (*Glomus* sp.)} strains (isolated from DSR root and soil samles) on growth and yield of DSR (var. BPT-5204) in Glass house at Dept. of Agricultural Microbiology, College of Agriculture, Raichur, Karnataka during 2014.

Recommended dose of fertilizer for DSR was 150: 75: 75 NPK (kg/ha), applied using urea, DAP and MOP respectively. Nitrogen was applied in split doses with 50% applied at the time of sowing, 25% at tillering stage and 25% at panicle emergence. Nitrogen and phosphorous in plant was determined by Micro Kjeldal and Vanadomolybdate yellow colour method respectively. The microbial count was done and expressed as MPN/g of root bits (Weaver *et al.*, 1994). Per cent root colonization of AMF was determined according to the method followed by Phillips and Hayman (1970). Number of spores in the soil surrounding the roots was determined by the wet sieving and decanting technique.

The experimental data collected on various characters taken at harvest were subjected to Fisher's method of "Analysis of variance" and interpretation of data as given by Gomez and Gomez (1984). The means were compared by Duncan's Multiple Range Test (DMRT). The level of significance used in 'F' test and't' test was P= 0.01 in pot culture experiments.

Result and Discussion:

Two isolates of *Azospirillum* which fared best under *in vitro* conditions *viz.*, ADSR-9, ADSR-23 and one isolate of AMF *viz.*, MDSR-3 were used individually and in combination with different levels of recommended N and P *viz.*, RDF, 75%N, 100%P, 100%K and 75%N, 75%P, 100%K.

Plant growth parameters

The pooled data indicated that combined inoculation of *Azospirillum* and AMF with 75%N, 75%P and 100%K significantly recorded maximum plant height and number of tillers/hill at harvest of crop. The seeds treated with ADSR-9 + MDSR-3 + 75%N, 75%P, 100%K (T_{13}) recorded highest plant height (78.13 cm) and highest number of tillers/hill (23.0) followed by T_{14} (ADSR-23 + MDSR-3 + 75%N, 75%P, 100%K) which recorded 77.63 cm plant height and 22.9 number of tillers/hill at harvest. Treatment T_{13} was significantly superior over all other treatments. All the individual and combined treatments were superior when imposed with fertilizer dose of 75%N, 75% P, 100%K compared to 75%N, 100% P, 100%K. *Azospirillum* is known to fix atmospheric nitrogen and is also known to have a role in phosphorus solubilisation while, AM fungi is a well-known phosphorus mobilizer and in addition it is also known to supplement nitrogen to crop plants. In addition to this, the higher plant height and number of tillers/hill might be due to production of plant growth promoting substances *viz.,* IAA and GA which play a central role in root elongation and shoot growth. *Azospirillum* enhances the IAA synthesis which in turn enables the plants to grow better.

Dry matter production

A perusal of data revealed that higher total dry matter of 56.10 g/plant was noticed in T_{13} compared to all other treatments. Higher dry matter production in the above treatments might be due to higher plant height and more number of tillers/hill in *Azospirillum* and AM fungi inoculated treatments which further might have contributed for accumulation of higher dry matter in the rice crop.

Microbiological attributes

Microbiological attributes like most probable number (MPN) of *Azospirillum*, spore count and per cent colonization of AM fungi significantly increased in combined inoculations rather than their individual inoculations.

The MPN count of *Azospirillum* significantly was highest in combined treatment (T_{13}) showing microbial count of 4.10×10^4 MPN/gram of roots. Similarly higher spore count and root colonization of AM fungi was highest in combined inoculation of T_{13} recording 205 spores/50g of soil and 74.03% of root colonization. In pot culture, the root growth stops or slows down as a result of root saturation due to limited space. This could be attributed to high spore production as observed in the study. According to the earlier findings, Finger millet treated with dual inoculation of AM fungi and *Azospirillum* showed higher plant height, dry weight of root and shoot, per cent root colonization and spore count (Ramakrishna and Bhuvaneswari, 2014).

Yield parameters and nutrient uptake

Yield parameters like number of productive tillers/hill, panicle length, number of filled and unfilled grains/panicle and grain yield significantly increased due to combined inoculation with *Azospirillum* and AM fungi with 75%N, 75%P 100%K in DSR (T_{13}).

Significantly more number (21.80) of productive tillers/hill was read at the harvest of crop in treatment T_{13}. Data pertaining to the panicle length showed that application of *Azospirillum* + AMF +75% N and P fertilizers significantly increased the length of panicle at harvest of crop. Simultaneously, combined inoculation of both the endophytes in T_{13}, with only 75 per cent N and P produced significantly higher grain yield compared to the plants supplied with 100 per cent N and P in T_7. The nitrogen and phosphorous uptake at harvest of plant increased significantly with *Azospirillum* + AM fungi + 75%N, 75%P, 100% (T_{13}). Increase in grain yield was associated with increase in growth parameter (plant height) and yield attributing characters which was evident from the significant positive correlation between grain yield and growth parameter and, grain yield and yield attributing characters. The supporting reviews for the above parameters cited by Naidu *et al.* (2003) reported that *Azospirillum* increased the number of tillers, dry matter, and number of panicles, number of filled grains and 1000 grain weight in rice under field experiment. The improved growth in mycorrhizal plants in the pot culture may be due to improved nitrogen and phosphorus nutrition. Yeasmin *et al.* (2007) stated that Mycorrhizal fungi significantly improved rice plants growth by increasing soil nutrients such as nitrogen and phosphorus.

Nitrogen and Phosphorous are essential elements in improving crop productivity throughout the world. Nitrogen and P availability to plants can be increased using combined inoculation of N-fixing and P-mobilising microorganisms. At the same time, their application is a highly efficient way to resolve the environmental problems such as deteriorating soil quality.

Conclusions:

The overall results indicated that the treatment with combined inoculations of *Azospirillum* and AM fungi was superior to their individual

inoculations. Among the combined inoculations, combination of ADSR-9 + MDSR-3 (T_{13}) was superior to ADSR-23 + MDSR-3 (T_{14}). The results thus indicate the compatibility and synergistic effect of both the endosymbionts on the growth and yield parameters of DSR. Further, the present work has evidently proved the advantage of combining *Azospirillum* and AM fungi with reduction of atleast 25% of N and P for DSR.

References:

Gomez, K.A. and Gomez, A.A. (1984): Statistical Procedure for Agricultural Research, **2nd** Edition. A Wiley Inter-Sciences Publications, New York. (USA).

Naidu, V.S.G.R., Panwar, J.D.S. and Annapurna, K. (2003): Yield response in rice to auxin application and inoculation with *Azospirillum brasilense*. Indian J. Plant Physiol., **8**: Pp. 96-98.

Pandey, S. and Velasco, L. (2002): Economics of direct seeding in Asia: Patterns of adoption and research priorities. In "Direct Seeding: Research Strategies and Opportunities" (Pandey, S., Mortimer, M., Wade, L., Tuong, T. P., Lopez, K. and Hardy, B., Eds.), pp. 3–14. International Rice Research Institute, Los Ban˜os, Philippines.

Phillips, J.M. and Hayman, D.S. (1970): Improved procedures for clearing roots and staining parasitic and vesicular arbuscular mycorrhizal fungi for rapid assessment of infection. Trans. Br. Mycol. Soc., **55**: Pp. 158-161.

Ramakrishna, K. and Bhuvaneswari G. (2014): Effect of inoculation of AM fungi and beneficial microorganisms on growth and nutrient uptake of *Eleusine coracana* (L.) Gaertn (Finger millet). ILNS, **8**(2): Pp. 59-69.

Weaver, R. W., Angla, S., Bottomley, P., Bezdicek, D., Smith, S., Tabatabai, A. and Wollum, A. (eds), 1994, Methods of soil analysis part-2, Microbiological and Biochemical Properties., Soil Sci. Soc. AM. Book Series, Madison, Wisconsin.

Yeasmin, T., Zaman, P., Rahman, A., Absar, N. and Khanum, N.S. (2007): Arbuscular mycorrhizal fungus inoculum production in rice plants. Afr. J. Agric. Res., **2**: Pp. 463-467.