Assessment of effect of inoculation of efficient Azospirillum strains on growth and yield of foxtail millet [Setaria italica (L.) Beauv.]

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Abstract
A pot culture experiment was conducted under greenhouse condition to study the effect of efficient isolates of Azospirillum strains on plant growth and yield parameters in foxtail millet. From foxtail millet growing areas of Raichur and Koppal districts, 40 samples were collected for isolation and enumeration of Azospirillum. In the investigation 21 isolates were found to be negative for denitrification and thus selected for further study. Based on in vitro studies, six efficient strains of Azospirillum were selected for pot culture experiment on foxtail millet growth and yield parameters at 30 and 60 days after sowing and at harvest. All the isolates including reference strain showed significant increase in plant height, No. of leaves per hill, No. of tillers/hill, Plant biomass, No. of panicles/hill, No. of filled seeds/panicle, No. of chaffy seeds/panicle, test weight, seed yield/plant, straw yield/hill of foxtail millet was observed over control. The strain MARV-18 was found to be efficient strain for foxtail millet crop.

Keywords: Azospirillum, effect, foxtail millet, growth, inoculation and yield

Introduction
Millets have been neglected despite their nutritive value and therapeutic uses (Anju and Sarita, 2010) [1]. Being rich in protein and calcium, millet serves as an important staple food for rural populations in tropical and sub-tropical developing countries, where calcium deficiency and anemia are wide spread. Even though, the crop had such significant importance, work done on the study and development of this crop is negligible. Not much study and research has been done on the neglected crops, especially on foxtail millet. Characterization of the accessions of foxtail millet can provide pivotal information for crop breeding and management of genetic resources. Characterization and evaluation of indigenous foxtail millet landraces is necessary for the utilization in crop improvement. There has only been some limited research and handful of publication on the study of this underutilized crop in the country. Thus, this research can act as a pivotal study and initiation as well as a reference for further researches on millets in Hyderabad-Karnataka region.

Inoculation of plants with Azospirillum can result in a significant change in various plants growth parameters, which may affect crop yield. Use of microbial inoculants or biofertilizers is the cheapest or low cost and eco-friendly input for sustainable agriculture (Gadagi et al. 2002) [4]. This eco-friendly approach will not only reduce the dependency on chemical fertilizers but also reduces the cost of cultivation. Associative nitrogen fixing microorganisms (Azospirillum) developed elsewhere have not been very consistent in their performance everywhere, due to their poor adaptability to the changing soil and agro-climatic conditions. Thus there is a need to study the associative nitrogen fixing microorganism Azospirillum and develop region specific Azospirillum strains for the foxtail millet crop. Keeping in view of the above facts or information attempts were made to investigate the effect of efficient isolates of Azospirillum on growth and yield of foxtail millet.

Materials and Methods
A pot culture experiment was conducted under greenhouse condition at Department of Agricultural Microbiology UAS Raichur, to study the effect of efficient isolates of Azospirillum strains on plant growth and yield parameters in foxtail millet. The pot culture
The experiment was conducted by using Complete Randomized Block Design with three replications. The detailed characteristics of soil used are in Table 1.

| Location     | Type   | Texture    | pH | EC (ds/m) | Organic carbon (%) | Available nitrogen (kg/ha) | Available phosphorous (kg/ha) |
|--------------|--------|------------|----|-----------|--------------------|---------------------------|-------------------------------|
| UAS, Raichur | Black  | Sand (%)   | 8.14 | 0.38     | 0.82              | 276.30                    | 30.42                         |
|              |        | Silt (%)   |      | 15.12    | 16.90            |                           |                               |
|              |        | Clay (%)   |      | 75.23    |                   |                           |                               |

**Inoculum preparation**
Efficient *Azospirillum* (MARV-2, 17, 18, 25, 30 and 33) strains were grown separately, in a 250 ml flask containing 100 ml NFBTB for 2 days. The grown cultures were homogenized then used for inoculation.

**Soil preparation and layout**
The experimental soil was brought to fine tilth by crushing the clods and removing the stones. The experiment was laid out in CRD. The pots were washed in running water and dried separately in a chamber to avoid contamination of soil and water from the pots. Tap water was connected to green house to facilitate irrigation.

**Seed inoculation and sowing**
The healthy foxtail millet seeds were sprinkled with the sticker solution of jaggery slurry (30 gm of jaggery free from preservatives was dissolved in 200 ml water and boiled for 10 minutes and cooled at room temperature). Then seeds were mixed separately and uniformly with each efficient strains of *Azospirillum* inoculums which were prepared in NFBTB cultured broth and inoculated seeds were air-dried for 20-30 minutes under shade. Then 10-15 seeds in each pot were sown by hand. Biometric observations were recorded on three randomly selected and labeled plants from each pot. The growth and yield parameters were recorded at 30 and 60 DAS and at harvest.

**Results and Discussion**

**Effect of efficient isolates of *Azospirillum* on foxtail millet plant growth parameters**
The results pertaining to plant height, number of leaves per hill, number of tillers per plant at harvest and plant biomass at 30 DAS, 60 DAS and at harvest are presented in Table 1. All the isolates including reference strain showed significant increase in plant height over control at 30 DAS, 60 DAS at harvest. Among the isolates, the maximum plant height (35.67 cm), number of leaves per hill (7.67), and plant biomass (3.74 g/plant) was observed with MARV-18 followed by MARV-17 and reference strain at 30 DAS. Similarly at 60 DAS the efficient *Azospirillum* isolate MARV-18 significantly increased the plant height with 166.33 cm, number of leaves per hill (11.00), and plant biomass (14.40 g/plant) followed by MARV-17 with 163.33 cm and reference strain with 160.67 cm over all the other treatments. Control recorded the lowest plant height of 151.67 cm at 60 DAS. At harvest efficient strain MARV-18 recorded height of 175.00 cm, number of leaves per hill (8.67), number of tillers at harvest (2.70 per plant) and plant biomass (24.47 g/plant) followed by MARV-17 and reference strain. Control had lowest growth parameters among the treatments.

The performance of *Azospirillum* in pot experiments (Subba Rao, 1982) and field trials (Subba Rao, 1981) was found to be highly significant in increasing crop growth. Nitrogen fixation is the first major mechanism for the enhancement of plant growth by *Azospirillum* (Prasad and Govindarajan, 2001).

**Effect of efficient isolates of *Azospirillum* on foxtail millet plant yield parameters**
Likewise plant growth attributes, yield attributes were also computed. The results pertaining to number of panicles per hill, number of filled seeds per panicle, number of chaffy seeds per panicle, test weight of 1000 seed (g), seed yield per plant (g), straw yield per hill (g) at harvest are furnished in Table 2. Efficient *Azospirillum* strain MARV-18 was observed with maximum number of panicles per hill (3.67 per plant), number of filled seeds per panicle (1073.33 filled seeds per panicle), number of chaffy seeds per panicle (100.00 chaffy seeds per panicle), test weight of 1000 seed (5.94 g), seed yield per plant (15.58 g), straw yield per hill (7.06 g) followed by MARV-17 and reference strain. Santhosh et al., (2015) reported that 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 and 100% K in DSR (T13).

| Treatments | *Azospirillum* strains | Plant height (cm) | Number of leaves per hill | Plant biomass (g plant⁻¹) | Number of tillers/ plant |
|------------|-----------------------|-------------------|--------------------------|---------------------------|-------------------------|
|            | 30 DAS| 60 DAS | At harvest | 30 DAS| 60 DAS | At harvest | 30 DAS| 60 DAS | At harvest |
| T1         | Control            | 31.33           | 151.67            | 154.33           | 3.67       | 6.67      | 5.33      | 2.00   | 9.40   | 19.93   | 1.40 |
| T2         | MARV-2              | 32.66           | 156.33            | 160.67           | 6.00       | 8.33      | 6.67      | 2.43   | 11.99  | 21.67  | 1.93 |
| T3         | MARV-17             | 33.67           | 163.33            | 167.00           | 7.00       | 10.00     | 8.33      | 3.14   | 13.80  | 23.07  | 2.60 |
| T4         | MARV-18             | 35.67           | 166.33            | 175.00           | 7.67       | 11.00     | 8.67      | 3.74   | 14.40  | 24.47  | 2.70 |
| T5         | MARV-25             | 33.00           | 159.33            | 163.66           | 6.66       | 9.00      | 7.33      | 2.76   | 12.10  | 22.86  | 2.20 |
| T6         | MARV-30             | 31.67           | 156.33            | 157.33           | 5.67       | 8.33      | 6.33      | 2.43   | 10.01  | 20.30  | 1.80 |
| T7         | MARV-33             | 32.33           | 158.33            | 163.33           | 6.00       | 8.33      | 7.00      | 2.66   | 12.00  | 21.93  | 2.00 |
| T8         | Reference strain    | 32.67           | 160.67            | 166.00           | 6.67       | 9.33      | 7.66      | 2.93   | 12.27  | 23.00  | 2.50 |
| S.Em±      |                     | 0.42            | 1.03              | 2.37             | 0.44       | 0.73      | 0.31      | 0.19   | 0.59   | 0.82   | 0.13 |
| C.D at 1%  |                     | 1.76            | 4.27              | 9.78             | 1.82       | 3.00      | 1.29      | 0.77   | 2.42   | 3.38   | 0.40 |

DAS – Days after sowing. Values are average of three replications.

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Table 1: The detailed characteristics of soil used are in Table.
Table 2: Effect of inoculation of *Azospirillum* isolates on yield parameters of foxtail millet.

| Sl. No. | Azospirillum strains | No. of panicles /hill | No. of filled seeds/panicle | No. of chaffy seeds/panicle | Test weight of 1000 seeds (g) | Seed yield/plant (kg/ha) | Straw yield/hill (g) |
|---------|----------------------|-----------------------|-----------------------------|-----------------------------|-----------------------------|------------------------|---------------------|
| 1       | Control              | 1.00                  | 458.33                      | 533.33                      | 3.09                        | 9.88                   | 4.85                |
| 2       | MARV-2               | 1.67                  | 658.33                      | 409.00                      | 3.46                        | 10.81                  | 5.74                |
| 3       | MARV-17              | 3.00                  | 966.67                      | 116.67                      | 5.16                        | 12.91                  | 6.85                |
| 4       | MARV-18              | 3.67                  | 1073.33                     | 100.00                      | 5.94                        | 13.58                  | 7.06                |
| 5       | MARV-25              | 2.33                  | 766.67                      | 191.66                      | 4.57                        | 12.07                  | 6.09                |
| 6       | MARV-30              | 1.00                  | 598.67                      | 433.33                      | 3.87                        | 10.58                  | 5.58                |
| 7       | MARV-33              | 2.00                  | 733.33                      | 296.66                      | 4.06                        | 11.54                  | 6.01                |
|         | Reference strain     | 2.67                  | 833.33                      | 133.33                      | 4.70                        | 12.34                  | 6.26                |
|         | S.Em±                | 0.42                  | 31.03                       | 19.62                       | 0.18                        | 0.27                   | 0.82                |
|         | C.D.at 1%            | 1.76                  | 128.19                      | 18.04                       | 0.73                        | 1.11                   | 3.38                |

Values are average of three replications

**Conclusion**

The investigation clearly showed the efficiency of native isolate of *Azospirillum* in increasing the growth and other N parameters of foxtail millet. The strain MARV-18 was found to be efficient nitrogen fixer for foxtail millet crop grown in the region. Increased use of various biological processes in soil, of which some examples have been given in the present study (Biological nitrogen fixation and plant growth promoting substances), will decisively contribute to make agriculture more productive with less harm to the environment. It is hoped for substantial increase in foxtail millet production in Northern Karnataka region through the use of our efficient *Azospirillum* strain (MARV-18). Further, the results need to be tested under field conditions for both growth and yield attributing characteristics of foxtail millet.

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