

## **Field Evaluation of Arbuscular Mycorrhizal Fungi in the Management of *Orobanche*: A Parasitic Weed in Tobacco (*Nicotiana tabacum* L.)**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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### **ABSTRACT**

An investigation was carried out to evaluate the methods of application of AMF cultures in the management of *Orobanche* viz., planting of pre colonized tobacco seedling; soil application and the combination of both. The experiment was carried out in *Orobanche* infested soils of tobacco growing areas of Belagavi district. The results of the present investigations have revealed that the treatment received STD AMF had reduced the emergence of *Orobanche* (1.33 plot<sup>-1</sup>) compared to UASDAMFT (1.67 plot<sup>-1</sup>) and UASDAMFS (2.89 plot<sup>-1</sup>). The results with respect to different methods of applications of AMF on *Orobanche* numbers revealed that planting of pre colonized tobacco seedling plus soil application at the time of planting suppressed the *Orobanche* emergence (0.00 plot<sup>-1</sup>) compared to planting of pre colonized seedlings (1.67 plot<sup>-1</sup>) and direct soil application of AMF cultures at the time of planting (4.22 plot<sup>-1</sup>). The results pertaining to the interactive effect between mycorrhizal cultures in conjunction with the methods of application of AMF cultures significantly reduced the population of *Orobanche* with the treatment received planting of pre

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colonized seedling along with soil application of UASDAMFT, UASDAMFS and STD AMF recorded zero emergences of *Orobanche* at 60 DAP. However, the highest numbers of weeds were recorded in uninoculated control (68.67plot<sup>-1</sup>). Furthermore, mycorrhizal parameters like spore count and percent of root colonization were found to be the highest in the plots received mycorrhization in the form of pre-colonization and soil application with STD AMF at the time of transplanting in the main field compared to uninoculated control.

**Keywords:** AMF; *Orobanche*; tobacco; pre colonized; soil application.

## 1. INTRODUCTION

Tobacco (*Nicotiana tabacum* L.) belongs to the family of *Solanaceae* or the nightshade family and the genus *Nicotiana*. Tobacco is one of the commercial crops prepared products from the tobacco leaves by curing them. Among the different species of tobacco, *Nicotiana tabacum* and *Nicotiana rustica* are well-known tobacco species grown commercially across the world. Tobacco contains several phytochemicals like nicotine a principle alkaloid known for its insecticidal property in the form of nicotine sulphate (40%), solanesol a co-enzyme Q-9 used as a cardiac drug, malic acid (4.5%) and citric acid (0.5%) used in food beverages. India stands second in exports next to Brazil and the USA. During 2017-18 tobacco and tobacco products earned Rs.20, 000 crores as a national exchange, while 5000 crore excise duty in the form of foreign exchange [1]. In Karnataka Nipani of Belgavi district occupies 90 percent of the area under tobacco cultivation. The important bidi varieties like A-2, Bhagyshree, Anand-119 and NBD-209 grown in the Nipani region of northern Karnataka are considered to be the best quality as compared to the leaves produced from the rest of the country. Our field survey has revealed that the reduction in the yield of tobacco in the region of Nipani was due to *Orobanche* infestation. *Orobanche* a complete root parasitic weed robs all nutrients and water from the host, resulting in stunted growth of the plant leading to an extent of yield reduction from five to 75 percent depending on the intensity of infestation.

The management of *Orobanche* is very difficult to achieve because of its micro seed size, high fecundity, asynchronous germination of seed and most importantly their life cycle occurs below ground near the root zone of the host plant. Hence difficult to suppress these parasitic weeds

under field conditions. Thus an integrated approach is needed for the management of these parasitic weeds like cultural practices, crop rotation, host plant resistance, chemical and biological treatments [2,3,4,5]. Hence, there is a need to develop a comprehensive and eco-friendly management system for their control. The studies have revealed that Mycorrhizal colonization persuades resistance to plant parasitism by converting strigolactones and orobanchol into mycorradicin [6,4] which is assembled in the tobacco root zone and thereby reduced accessibility of strigolactones and orobanchol for germination of *Orobanche*.

## 2. MATERIALS AND METHODS

A field experiment was conducted during *Kharif* 2018-19 to study the "Field evaluation of Arbuscular mycorrhizal fungi in the management of *Orobanche*: A parasitic weed in tobacco (*Nicotiana tabacum* L.)." These experiments were carried out in *Orobanche* infested soils of tobacco growing areas of Nipani in the Belagavi district of northern Karnataka. An investigation was carried out to evaluate the methods of application of AM fungal cultures in the management of *Orobanche viz.*, planting of pre colonized tobacco seedling; soil application and the combination of both with the use of following AMF culture UASDAMFT (Isolated from *Orobanche* suppressive soils in tobacco), UASDAMFS (Isolated from *Striga* suppressive soils in sugarcane) and STD AMF Consortium (Department of Agricultural Microbiology, UAS Dharwad). The experiment was laid out in randomized complete design with the factorial concept. There were 3 main factors and 3 sub-factors consisting of a combination of AM fungi and different methods of application and UIC outside the experiment run with RCBD as given below:

**Chart 1. 3 main factors and 3 sub-factors**

|   |  |
|---|--|
| <b>I factor: AMF cultures</b>                       |  |
| M <sub>1</sub>                                      | UASDAMFT consortium (tobacco native)   |
| M <sub>2</sub>                                      | UASDAMFS consortium (sugarcane native)   |
| M <sub>3</sub>                                      | STD AMF consortium (UASD reference)  |
| <b>II factor: Methods of application (Three)</b>    |  |
| S <sub>1</sub>                                      | Pre colonization of the tobacco seedlings in the nursery beds @ 2 kg /m <sup>2</sup> |
| S <sub>2</sub>                                      | Soil application (@ 6-8 kg/acre mixed with 200 kg of vermicompost)                   |
| S <sub>3</sub>                                      | Pre colonization + Soil application  |
| <b>UIC</b> UIC outside the experiment run with RCBD |  |

## 2.1 Application of Mycorrhizal Cultures

### Pre colonization of the tobacco seedlings in the nursery beds with AMF @ 2 kg /m<sup>2</sup>

Nursery beds were prepared and subjected for solarization (4 to 5 weeks) in order to prevent the native AMF infective propagules. AMF culture along with vermicompost @ 2:25 was applied in the furrows prior to the sowing of tobacco seeds.

### 2.2 Soil Application

AMF culture @ 8 kg per acre was applied along with 200 kg of vermicompost at the time of transplanting of tobacco seedlings.

### 2.3 Parameters Measured

#### *Orobanche* parameters

The number of *Orobanche* weed emergence per plot was documented at 60, 90 and 120 DAP. The shoot and root portions of *Orobanche* plants were separated and oven-dried at 60°C to constant weight. The dry weights were then recorded separately for shoots and roots and the average of three were expressed in grams.

### 2.4 parameters of Tobacco

#### Plant height

The plant height of tobacco was recorded at 60, 90, 120 DAP. The tobacco plant height is defined as the average stem distance from the soil to the insertion of the top visible leaf on the stem and was expressed in centimetre (cm).

#### Relative chlorophyll content (SPAD Reading)

Relative chlorophyll content of the tobacco plants were recorded at the intervals 60, 90 and 120,

DAP. The single photoelectric analyzing diode (SPAD) meter was used for recording the relative chlorophyll content.

#### Mycorrhizal parameters

The chlamydospores in the rhizosphere of tobacco were determined by the wet sieving and decantation method as outlined by Gerdemann and Nicholson [7]. Spores counts were taken under a stereo zoom microscope. Mycorrhizal root colonization was determined as per the procedure proposed by Phillips and Hayman [8]. The percentage of roots colonized by mycorrhizae was calculated by the formula.

$$\text{Percent root colonization} = \frac{\text{Root bits positive for colonization}}{\text{Total number of root bits}} \times 100$$

### 2.5 Statistical Analysis and Data Interpretation

The data collected at different growth stages of crops were subjected to statistical analysis. Based on mean values obtained, analysis and interpretation of data were studied using the Fischer's method of analysis of variance technique as described by Gomez and Gomez [9]. The level of significance used in the 'F' and 't' test was p = 0.05. Critical difference values were calculated wherever the 'F' test was significant.

## 3. RESULTS AND DISCUSSION

### 3.1 The Number of *Orobanche* weed Emergence Per Plot was Documented at 60, 90 and 120 DAP

The results of the present investigations have revealed that the treatment received STD AMF had reduced the emergence of *Orobanche* (1.33 plot<sup>-1</sup>) compared to UASDAMFT (1.67 plot<sup>-1</sup>) and

UASDAMFS (2.89 plot<sup>-1</sup>). The results with respect to different methods of applications of AMF on *Orobanche* numbers revealed that, planting of pre colonized tobacco seedling plus soil application at the time of planting suppressed the *Orobanche* emergence (0.00 plot<sup>-1</sup>) compared to planting of pre colonized seedlings (1.67 plot<sup>-1</sup>) and direct soil application of AMF cultures at the time of planting (4.22 plot<sup>-1</sup>). The results pertaining to the interactive effect between mycorrhizal cultures in conjunction with the methods of application of AMF cultures significantly reduced the population of *Orobanche* with the treatment received planting of pre colonized seedling along with soil application of UASDAMFT, UASDAMFS and STD AMF recorded zero emergences of *Orobanche*. However, the highest numbers of weeds were recorded in uninoculated control (68.67plot<sup>-1</sup>). at 60 DAP (Table 1) Fig. 2. Similar trends were also recorded at 90 and 120 DAP.

The biomass of *Orobanche* per plot was documented At 120 DAP (Table 2), The results with respect to the interactive effect between mycorrhizal cultures and their methods of application significantly reduced the biomass of *Orobanche* with the treatment received planting of pre colonized seedling along with soil application at the time of planting with UASDAMFT, UASDAMFS and STD AMF recorded zero biomass of *Orobanche* at 120 DAP compared to uninoculated control (155.67g plot<sup>-1</sup>). This might be due to the reduced availability of signaling molecules in mycorrhized roots like strigolactones and orobanchol as reported by Walter et al. [6]. Furthermore, our research findings are in accordance with the finding of Yoneyama et al., [10] and Lopez-Raez et al., [11], wherein inoculation of mycorrhizal fungi significantly reduced the haustorial formation, thereby controlling the parasitic weeds. Caterina et al. [13] revealed that strigolactones are secondary metabolites produced in root zones of the host plant under the nutrient stress conditions which also act as stimulant for both AMF and parasitic weed. In presence of mycorrhizal fungi the strigolactones availability very less for parasitic weed germination, which helps to avoid weed emergence and enhance host plant physiological parameters.

### 3.2 Plant Height of Tobacco

The results from the interactive studies indicated that planting of pre colonized tobacco seedling

and soil application of STD AMF at the time of planting recorded significantly the highest plant height, (135.87 cm) followed by the treatment received pre colonized UASDAMFT (133.33 cm) which is statistically on par with each other. Pre colonized tobacco seedling with AMF STD (129.80 cm), pre colonized UASDAMFS plus soil application (128.87 cm) both were found to be statistically on par with each other. However, reduced plant height was noticed in uninoculated control (110.8 cm) at 120 DAP (Table 3). The reduction in plant height with nonmycorrhizal plots due to parasitization of the host with *Orobanche*, as initially reported by Olakojo et al. [12] in maize genotypes. The positive interactions between native isolates AMF and host plants indicated an increased plant height in sugarcane by several workers Madhura et al. [3]; Shubha et al. [4] and Waratadar et al. [14] reported that the tomato plants pre colonized with native AM fungal cultures enhanced the plant height compared to nonmycorrhizal tomato plants under *Orobanche* infested soils.

### 3.3 Relative Chlorophyll Content (SPAD Reading)

At 120 DAP (Table 4), Among the interactive studies, the highest relative chlorophyll content was recorded in the treatment received as both pre-colonization as well as soil applications with STD AMF (43.80), which is significantly superior to the treatment received pre colonized tobacco seedling with UASDAMFT plus soil application (43.19). However, the lowest relative chlorophyll content was recorded in the nonmycorrhized treatment (33.23). Abdel and Mohamedin [15] documented an increase in the physiological parameter like relative chlorophyll content due to the mycorrhizal inoculation in buffalo grass.

### 3.4 Mycorrhizal Spore and Root Colonization

At 90 DAP (Table 5), Spore counts were maximum in the treatment received both pre - colonization as well as soil applications of STD AMF (493.33/50 g of soil) and the second highest was recorded in the treatment received pre colonized tobacco seedling with UASDAMFT alone (413.67/50 g of soil) followed by pre colonized tobacco seedling UASDAMFT plus soil application (411.33/50 g of soil). However, the least number of mycorrhizal spore counts were observed in the rhizosphere of nonmycorrhized tobacco plants (104.33/ 50 g of soil).

**Table 1. Effect of AMF fungal cultures on *Orobanche* emergence in tobacco**

| Treatment      | Number of <i>Orobanche</i> per plot |                |                |                   |                       |                |                |                   |                       |                |                |                   |
|----------------|-------------------------------------|----------------|----------------|-------------------|-----------------------|----------------|----------------|-------------------|-----------------------|----------------|----------------|-------------------|
|                | 60 DAP                              |                |                |                   | 90 DAP                |                |                |                   | 120 DAP               |                |                |                   |
|                | Method of application               |                |                |                   | Method of application |                |                |                   | Method of application |                |                |                   |
| AM Fungi       | S <sub>1</sub>                      | S <sub>2</sub> | S <sub>3</sub> | Mean              | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              |
| M <sub>1</sub> | 0.00                                | 5.00           | 0.00           | 1.67              | 0.00                  | 6.00           | 0.00           | 2.00              | 0.00                  | 6.33           | 0.00           | 2.11              |
| M <sub>2</sub> | 3.33                                | 5.33           | 0.00           | 2.89              | 5.67                  | 5.67           | 0.00           | 3.78              | 5.33                  | 2.67           | 0.00           | 2.67              |
| M <sub>3</sub> | 1.67                                | 2.33           | 0.00           | 1.33              | 2.33                  | 3.00           | 0.00           | 1.78              | 2.67                  | 2.33           | 0.00           | 1.67              |
| Mean           | 1.67                                | 4.22           | 0.00           |                   | 2.67                  | 4.89           | 0.00           |                   | 2.67                  | 3.78           | 0.00           |                   |
| UIC            |                                     |                |                | 68.67             |                       |                |                | 22.33             |                       |                |                | 9.33              |
|                | <b>S.Em±</b>                        |                |                | <b>C D at 5 %</b> | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> |
| M              | 0.23                                |                |                | 0.90              | 0.25                  |                |                | 1.00              | 0.12                  |                |                | 0.50              |
| S              | 0.15                                |                |                | 0.46              | 0.24                  |                |                | 0.74              | 0.24                  |                |                | 0.75              |
| M at S         | 0.31                                |                |                | 1.11              | 0.42                  |                |                | 1.44              | 0.36                  |                |                | 1.17              |
| UIC            | 0.48                                |                |                | 1.43              | 0.64                  |                |                | 1.92              | 0.37                  |                |                | 1.11              |

| AM Fungi                                     | Method of application                             |
|--|---|
| M <sub>1</sub> : UASDAMFT (tobacco native)   | S <sub>1</sub> : pre-colonized                    |
| M <sub>2</sub> : UASDAMFS (sugarcane native) | S <sub>2</sub> : soil application                 |
| M <sub>3</sub> : STD AMF                     | S <sub>3</sub> : pre-colonized + soil application |
| UIC: Uninoculated control                    |   |

**Table 2. Effect of AMF fungal cultures on dry biomass of *Orobanche* at 120 DAP**

| Treatment      | Biomass (g/plot)      |                |                |                   |
|----------------|-----------------------|----------------|----------------|-------------------|
|                | Method of application |                |                |                   |
| AM Fungi       | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              |
| M <sub>1</sub> | 0                     | 69.33          | 0              | 23.11             |
| M <sub>2</sub> | 44                    | 41.33          | 0              | 28.44             |
| M <sub>3</sub> | 36.33                 | 37.00          | 0              | 24.44             |
| Mean           | 26.78                 | 49.22          | 0              |                   |
| UIC            |                       |                |                | 155.67            |
|                | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> |
| M              | 0.22                  |                |                | 0.87              |

| Treatment | Biomass (g/plot)      |      |
|-----------|-----------------------|------|
|           | Method of application |      |
| S         | 1.24                  | 3.84 |
| M at S    | 1.77                  | 5.50 |
| UIC       | 2.20                  | 6.54 |

| AM Fungi                                     | Method of application                             |
|--|---|
| M <sub>1</sub> : UASDAMFT (tobacco native)   | S <sub>1</sub> : pre-colonized                    |
| M <sub>2</sub> : UASDAMFS (sugarcane native) | S <sub>2</sub> : soil application                 |
| M <sub>3</sub> : STD AMF                     | S <sub>3</sub> : pre-colonized + soil application |
| UIC: Uninoculated control                    |   |

**Table 3. Plant height of tobacco as influenced by AMF fungal cultures in *Orobanche* infested soil**

| Treatment       | Plant height (cm)     |                      |                      |             |                       |                      |                      |             |                       |                      |                      |             |
|-----------------|-----------------------|----------------------|----------------------|-------------|-----------------------|----------------------|----------------------|-------------|-----------------------|----------------------|----------------------|-------------|
|                 | 60 DAP                |                      |                      |             | 90 DAP                |                      |                      |             | 120 DAP               |                      |                      |             |
|                 | Method of application |                      |                      |             | Method of application |                      |                      |             | Method of application |                      |                      |             |
| <b>AM Fungi</b> | <b>S<sub>1</sub></b>  | <b>S<sub>2</sub></b> | <b>S<sub>3</sub></b> | <b>Mean</b> | <b>S<sub>1</sub></b>  | <b>S<sub>2</sub></b> | <b>S<sub>3</sub></b> | <b>Mean</b> | <b>S<sub>1</sub></b>  | <b>S<sub>2</sub></b> | <b>S<sub>3</sub></b> | <b>Mean</b> |
| M <sub>1</sub>  | 80.27                 | 70.20                | 78.73                | 76.40       | 100.73                | 96.13                | 100.60               | 99.16       | 133.33                | 128.47               | 122.80               | 128.20      |
| M <sub>2</sub>  | 70.20                 | 69.60                | 68.33                | 69.38       | 97.47                 | 95.47                | 98.47                | 97.13       | 121.00                | 120.67               | 128.87               | 123.51      |
| M <sub>3</sub>  | 71.73                 | 70.20                | 80.73                | 74.22       | 99.27                 | 95.87                | 102.73               | 99.29       | 129.80                | 128.53               | 135.87               | 131.40      |
| Mean            | 74.07                 | 70.00                | 75.93                |             | 99.16                 | 95.82                | 100.60               |             | 128.04                | 125.89               | 129.18               |             |
| UIC             |                       |                      |                      | 61.47       |                       |                      |                      | 86.33       |                       |                      |                      | 110.8       |
|                 | <b>S.Em±</b>          |                      | <b>C D at 5 %</b>    |             | <b>S.Em±</b>          |                      | <b>C D at 5 %</b>    |             | <b>S.Em±</b>          |                      | <b>C D at 5 %</b>    |             |
| M               | 0.68                  |                      | 2.67                 |             | 1.34                  |                      | 5.28                 |             | 2.19                  |                      | 8.61                 |             |
| S               | 1.38                  |                      | 4.26                 |             | 1.55                  |                      | 4.79                 |             | 2.05                  |                      | 6.31                 |             |
| M at S          | 2.07                  |                      | 6.57                 |             | 2.57                  |                      | 8.53                 |             | 3.63                  |                      | 12.32                |             |
| UIC             | 2.10                  |                      | 6.24                 |             | 2.87                  |                      | 8.54                 |             | 3.89                  |                      | 11.5                 |             |

| AM Fungi                                     | Method of application                             |
|--|---|
| M <sub>1</sub> : UASDAMFT (tobacco native)   | S <sub>1</sub> : pre-colonized                    |
| M <sub>2</sub> : UASDAMFS (sugarcane native) | S <sub>2</sub> : soil application                 |
| M <sub>3</sub> : STD AMF                     | S <sub>3</sub> : pre-colonized + soil application |
| UIC: Uninoculated control                    |   |

**Table 4. Relative chlorophyll content as influenced by AM fungi in tobacco**

| Treatment      | Chlorophyll content (SPAD reading) |                |                |                   |                       |                |                |                   |                       |                |                |                   |
|----------------|------------------------------------|----------------|----------------|-------------------|-----------------------|----------------|----------------|-------------------|-----------------------|----------------|----------------|-------------------|
|                | 60 DAP                             |                |                |                   | 90 DAP                |                |                |                   | 120 DAP               |                |                |                   |
|                | Method of application              |                |                |                   | Method of application |                |                |                   | Method of application |                |                |                   |
| AM Fungi       | S <sub>1</sub>                     | S <sub>2</sub> | S <sub>3</sub> | Mean              | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              |
| M <sub>1</sub> | 45.18                              | 40.79          | 46.15          | 44.04             | 46.24                 | 41.22          | 45.55          | 44.34             | 43.14                 | 40.27          | 43.19          | 42.20             |
| M <sub>2</sub> | 40.21                              | 39.03          | 43.44          | 40.89             | 41.08                 | 39.61          | 44.50          | 41.73             | 38.90                 | 38.28          | 41.92          | 39.70             |
| M <sub>3</sub> | 45.60                              | 41.49          | 47.23          | 44.77             | 45.11                 | 41.71          | 46.93          | 44.58             | 42.48                 | 39.66          | 43.80          | 41.98             |
| Mean           | 43.66                              | 40.43          | 45.61          |                   | 44.14                 | 40.84          | 45.66          |                   | 41.51                 | 39.41          | 42.97          |                   |
| UIC            |                                    |                |                | 33.96             |                       |                |                | 35.47             |                       |                |                | 33.23             |
|                | <b>S.Em±</b>                       |                |                | <b>C D at 5 %</b> | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> |
| M              | 0.60                               |                |                | 2.38              | 0.89                  |                |                | 3.49              | 0.80                  |                |                | 3.14              |
| S              | 0.68                               |                |                | 2.10              | 0.52                  |                |                | 1.62              | 0.63                  |                |                | 1.96              |
| M at S         | 1.13                               |                |                | 3.78              | 1.16                  |                |                | 4.16              | 1.20                  |                |                | 4.16              |
| UIC            | 1.23                               |                |                | 3.67              | 1.28                  |                |                | 3.81              | 1.17                  |                |                | 3.50              |

| AM Fungi                                     | Method of application                             |
|--|---|
| M <sub>1</sub> : UASDAMFT (tobacco native)   | S <sub>1</sub> : pre-colonized                    |
| M <sub>2</sub> : UASDAMFS (sugarcane native) | S <sub>2</sub> : soil application                 |
| M <sub>3</sub> : STD AMF                     | S <sub>3</sub> : pre-colonized + soil application |
| UIC: Uninoculated control                    |   |

**Table 5. Mycorrhizal spore load as influenced by AM fungi in tobacco rhizosphere**

| Treatment      | Number of spores /50g of soil |                |                |                   |                       |                |                |                   |                       |                |                |                   |
|----------------|-------------------------------|----------------|----------------|-------------------|-----------------------|----------------|----------------|-------------------|-----------------------|----------------|----------------|-------------------|
|                | 45 DAP                        |                |                |                   | 60 DAP                |                |                |                   | 90 DAP                |                |                |                   |
|                | Method of application         |                |                |                   | Method of application |                |                |                   | Method of application |                |                |                   |
| AM Fungi       | S <sub>1</sub>                | S <sub>2</sub> | S <sub>3</sub> | Mean              | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              | S <sub>1</sub>        | S <sub>2</sub> | S <sub>3</sub> | Mean              |
| M <sub>1</sub> | 321.67                        | 206.67         | 326.50         | 284.94            | 365.33                | 200.33         | 353.67         | 306.44            | 413.67                | 367.33         | 411.33         | 397.44            |
| M <sub>2</sub> | 196.33                        | 184.00         | 289.67         | 223.33            | 218.67                | 326.33         | 223.33         | 256.11            | 236.00                | 215.33         | 378.00         | 276.44            |
| M <sub>3</sub> | 306.67                        | 268.89         | 380.11         | 318.56            | 342.67                | 310.33         | 426.00         | 359.67            | 404.33                | 246.67         | 493.33         | 381.44            |
| Mean           | 274.89                        | 219.85         | 332.09         |                   | 308.89                | 279.00         | 334.33         |                   | 351.33                | 276.44         | 427.56         |                   |
| UIC            |                               |                |                | 63.00             |                       |                |                | 85.00             |                       |                |                | 104.33            |
|                | <b>S.Em±</b>                  |                |                | <b>C D at 5 %</b> | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> | <b>S.Em±</b>          |                |                | <b>C D at 5 %</b> |

| Treatment | Number of spores /50g of soil |       |                       |       |                       |       |
|-----------|-------------------------------|-------|-----------------------|-------|-----------------------|-------|
|           | 45 DAP                        |       | 60 DAP                |       | 90 DAP                |       |
|           | Method of application         |       | Method of application |       | Method of application |       |
| M         | 4.03                          | 15.85 | 4.97                  | 19.53 | 9.63                  | 37.83 |
| S         | 4.44                          | 13.68 | 5.41                  | 16.68 | 7.94                  | 24.47 |
| M at S    | 7.46                          | 24.85 | 9.13                  | 30.42 | 14.79                 | 50.90 |
| UIC       | 7.39                          | 21.96 | 9.54                  | 28.35 | 13.86                 | 41.20 |

| AM Fungi                                     | Method of application                             |
|--|---|
| M <sub>1</sub> : UASDAMFT (tobacco native)   | S <sub>1</sub> : pre-colonized                    |
| M <sub>2</sub> : UASDAMFS (sugarcane native) | S <sub>2</sub> : soil application                 |
| M <sub>3</sub> : STD AMF                     | S <sub>3</sub> : pre-colonized + soil application |
| UIC: Uninoculated control                    |   |



**Table 6. AMF root colonization in tobacco as influenced by AM fungi**

| Treatment       | Percentage (%)        |                      |                      |             |                       |                      |                      |             |
|-----------------|-----------------------|----------------------|----------------------|-------------|-----------------------|----------------------|----------------------|-------------|
|                 | 60 DAP                |                      |                      |             | 90 DAP                |                      |                      |             |
|                 | Method of application |                      |                      |             | Method of application |                      |                      |             |
| <b>AM Fungi</b> | <b>S<sub>1</sub></b>  | <b>S<sub>2</sub></b> | <b>S<sub>3</sub></b> | <b>Mean</b> | <b>S<sub>1</sub></b>  | <b>S<sub>2</sub></b> | <b>S<sub>3</sub></b> | <b>Mean</b> |
| M <sub>1</sub>  | 70.50                 | 46.11                | 67.43                | 61.34       | 72.20                 | 54.73                | 75.27                | 67.40       |
| M <sub>2</sub>  | 48.90                 | 48.27                | 65.23                | 54.13       | 51.53                 | 49.27                | 71.40                | 57.40       |
| M <sub>3</sub>  | 67.93                 | 63.40                | 73.18                | 68.17       | 72.13                 | 70.97                | 77.90                | 73.67       |
| Mean            | 62.44                 | 52.59                | 68.61                |             | 65.29                 | 58.32                | 74.86                |             |
| UIC             |                       |                      |                      | 28.60       |                       |                      |                      | 36.07       |
|                 | <b>S.Em±</b>          |                      | <b>C D at 5 %</b>    |             | <b>S.Em±</b>          |                      | <b>C D at 5 %</b>    |             |
| M               | 0.77                  |                      | 3.03                 |             | 1.14                  |                      | 4.50                 |             |
| S               | 0.86                  |                      | 2.67                 |             | 0.94                  |                      | 2.90                 |             |
| M at S          | 1.45                  |                      | 4.82                 |             | 1.75                  |                      | 6.05                 |             |
| UIC             | 2.08                  |                      | 6.18                 |             | 1.92                  |                      | 5.72                 |             |

| AM Fungi                                     | Method of application                             |
|--|---|
| M <sub>1</sub> : UASDAMFT (tobacco native)   | S <sub>1</sub> : pre-colonized                    |
| M <sub>2</sub> : UASDAMFS (sugarcane native) | S <sub>2</sub> : soil application                 |
| M <sub>3</sub> : STD AMF                     | S <sub>3</sub> : pre-colonized + soil application |
| UIC: Uninoculated control                    |   |



**Fig. 1. Pre colonized tobacco seedlings with native UASDAMF cultures in the nursery bed at 45 DAS**

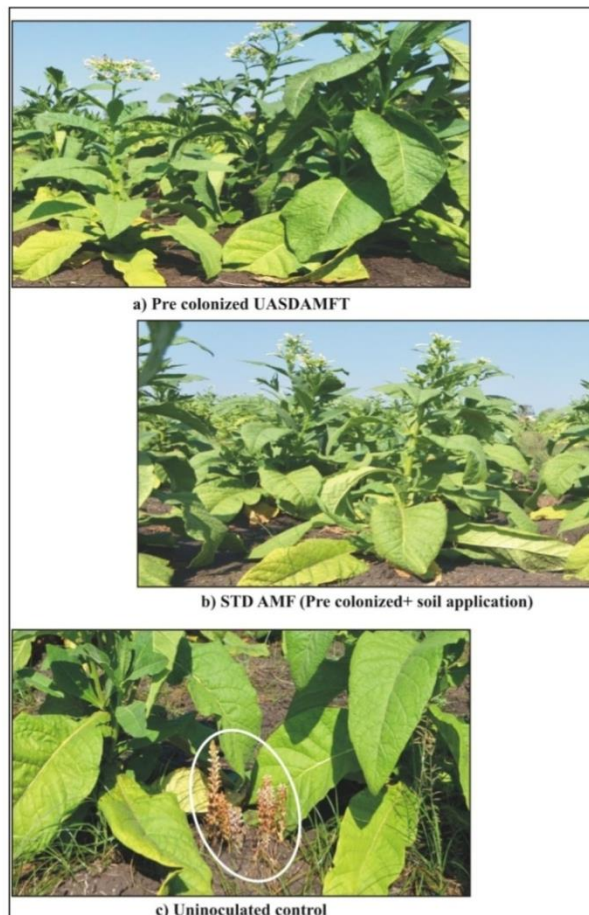


Fig. 2. Effect of mycorrhization on *Orobanche* emergence in tobacco at 60 DAP

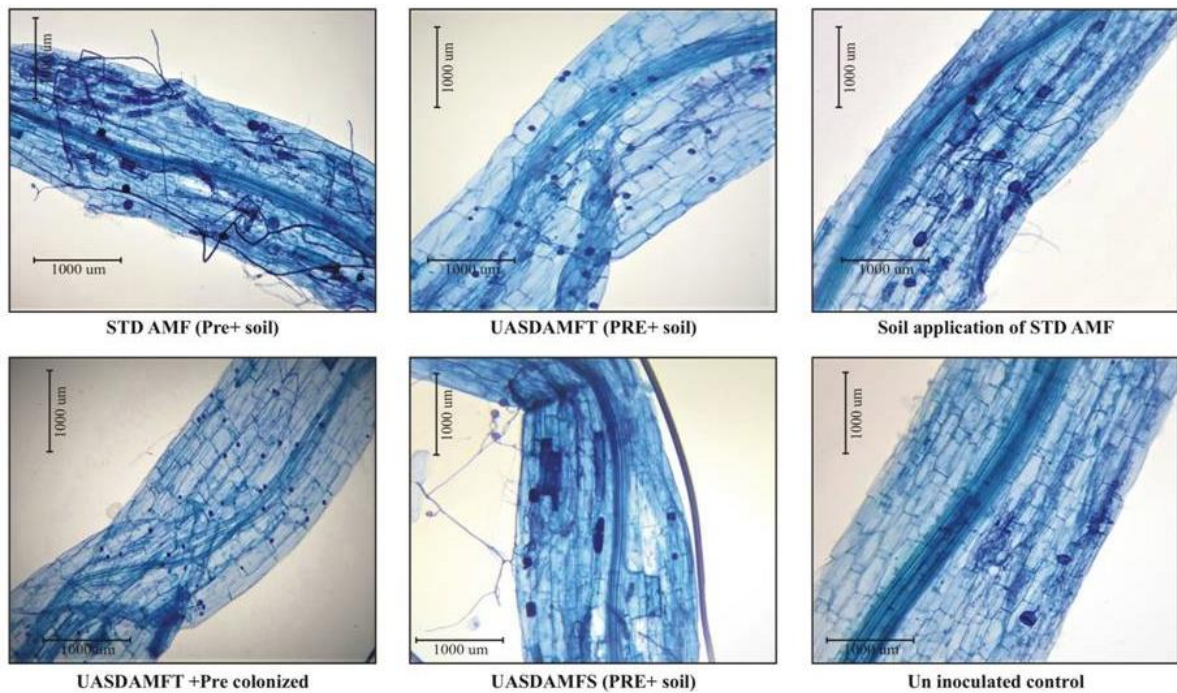


Fig. 3. AMF root colonization in tobacco

At 90 DAP (Fig. 3.), the percent of root colonization was a maximum in the treatment imposed, the highest mycorrhizal root colonization was recorded pre colonized tobacco seedling with STD AMF plus soil application at the time of planting (77.90%), followed by the treatment received pre colonized UASDAMFT plus soil applications (75.27%) and pre colonized UASDAMFT alone (72.20%). However, least percent of root colonization was observed in the roots of nonmycorrhized tobacco plants (36.07%) Table 6. The inoculation of sugarcane plants with AM fungal cultures increased the root colonization and spore number was recorded with mycorrhization compared to nonmycorrhizal plants Jones et al. [2] and Shubha et al. [4].

#### 4. CONCLUSION

The suppression of *Orobanche* by AM fungi is chiefly known to be due to depletion of strigolactones by them in the rhizosphere of the host plants. Strigolactones are signaling compound that play a vital role as germination stimulants of the parasitic *Orobanche*. Hence research of the present investigation is a promising strategy to develop a bio herbicide against parasitic weeds with a special reference to *Orobanche* in tobacco and other solanaceous crops like tomato brinjal and others.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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