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# Growth of Mulberry Saplings as Influenced by the Application of Microbial Consortium

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Abstract: Studies on co-inoculation of phosphate solubilizing micro-organisms, nitrogen fixing bacteria and VA-mycorrhiza at different levels and sources of nitrogen and phosphorus on mulberry saplings under nursery conditions indicated significant influence on sub-sub plot effect of two varieties on the survivability of saplings. Highest survivability of saplings (90.47%) was recorded with the variety $V_1$ after 60 days of planting in nursery followed by $S_{36}$ which showed 47.71 % of survivability. Application of two sources of phosphorus as main plot effect also significantly influenced the height of the saplings in 60 days itself. Among the two sources of phosphorus, maximum height was recorded with $S_2$ (27.25 cm / sapling) followed by $S_1$ (26.05 cm). Significant influence of the application of different levels and sources of fertilizer ($F_1$-$F_{10}$) as sub plot effect on height of saplings was also observed. Maximum height (32.28 cm / sapling) was recorded in the treatment $F_9$ (recommended dose of phosphorus and nitrogen with co-inoculation). This was significantly higher as compared to other fertilizer levels including control ($F_{10}$) but was at par with $F_8$ and $F_7$. Significant difference in the height of the saplings was also observed due to different varieties as sub-sub plot effect. Maximum height (34.25 cm / sapling) was recorded with the saplings of $V_1$ variety after 60 days of plantation in nursery followed by $S_{36}$ (19.06cm / sapling). The co-inoculation of saplings with PSM, *Azotobacter* and VA-mycorrhiza and application of ¾ th of the recommended dose of phosphorus as rock phosphate (RP) as well as full dose of nitrogen ($F_8I_1S_2V_1$) has resulted in maximum height (45.55 cm / sapling). This was significantly higher over most of the treatments including control. No significant difference between the two sources of phosphorus ( $S_1$ and $S_2$) as main plot effect on the length of primary roots of saplings was observed. However, the sub plot effect was found to be significant. Among the sub plot effect ($F_1$-$F_{10}$), maximum length of primary roots (28.83 cm / sapling) was recorded due to treatment $F_5$. This was significantly higher as compared to all other fertilizer levels but was at par with $F_1$, $F_2$, $F_9$ and $F_{10}$. Significant difference in the primary root length of saplings was also observed due to different varieties as sub-sub plot effect. Maximum primary root length (27.49 cm / sapling) was recorded with the saplings of $V_1$ followed by $S_{36}$ (18.93 cm / sapling). Significantly higher leaf yield of 28.88 g / sapling was recorded in $S_1$ as compared to $S_2$ (25.45 g/ sapling) due to main plot effect. Sub plot effect on the leaf yield per sapling was also found to be significant. Among the sub plot effect ($F_1$-$F_{10}$), maximum leaf yield (38.03 g / sapling) was recorded in the treatment $F_7$. Significant difference in leaf yield per sapling was also observed due to different varieties as sub-sub plot effect. Maximum leaf yield (30.32 g / sapling) was recorded with the variety $V_1$ followed by $S_{36}$ (24.01 g / sapling). The interaction effect was also found to be significant . Maximum leaf yield (45.15 g / sapling) was recorded in $V_1$ due to inoculation with PSM, *Azotobacter* and VA-mycorrhiza with application of ½ of the recommended dose of phosphorus as single super phosphate (SSP) as well as 3/4th of nitrogen ($F_4I_1S_1V_1$). This was significantly higher over majority of the treatments including control.
**Keywords:** Microbial consortium, mulberry saplings, nursery Biofertilisers

## INTRODUCTION

Plant nutrient management in various agriculture crops has been a matter of concern and more awareness has been there owing to health hazards posed by various chemical fertilizers. Mulberry is one such agriculture crop whose leaf is exclusively used for silkworm *Bombyx mori* L. rearing. The quality production of mulberry leaf with reduced input cost is of utmost concern to all sericulturists. In order to generate more quality leaf major thrust is laid on use of various components contributing to growth of plants at nursery as well as field level. These components have to be ecologically safe, economically viable and environmentally safe. The agricultural land is the most complex microbiological medium [10, 11] which has come under threat due to excessive cropping and frequent use of chemical fertilizers. About 60 % of the sericultural farmers in India do not apply chemical fertilizers in required quantity because of its high cost and non availability in time and as such they harvest poor cocoon crop due to reduction in the nutrient value of leaf. In extreme cases, the crop even fails which creates an unprecedented economic burden on farmers. In order to avoid all these problems, the balanced and judicious use of chemical fertilisers, organic manures and beneficial micro-organisms as bio-fertilizers assumes significant importance. Bio-fertilizers are one of the most important components of the integrated plant nutrient management system. These are carrier based formulations of live or latent cells of beneficial micro-organisms like nitrogen fixing bacteria (*Azotobacter*, *Azospirillum* and *Rhizobium* etc.), phosphate solubilizing bacteria (*Bacillus megaterium*) and fungi (*Aspergillus awamori*) and VA-mycorrhiza (*Glomus fasciculatum* or *Glomus mosseae*) used for inoculation of seeds, soils and plant roots to encourage growth and yield of plants besides minimizing the use of chemical fertilizers. These bio-inoculants when used to inoculate seeds or soil can augment the supply of nutrients (nitrogen and phosphorus) to plant either by harnessing atmospheric nitrogen through symbiotic / non-symbiotic biological nitrogen fixation (BNF) or by solubilizing or mobilizing phosphorus from unavailable to available forms. The cocoon parameters due to co-inoculation with microbial consortium have indicated improved trend [3]. Association of VAM fungi in the rhizosphere of mulberry under temperate climatic conditions was also observed [4]. Further, inoculation of soil with these organisms enriches rhizosphere microflora which can have a vital influence on plant growth through mineral uptake [12]. The beneficial effect of inoculation of mulberry plants with *Azotobacter* bio-fertilizer and VA-mycorrhiza have been well documented [6, 9] respectively. Among the fertiliser elements, nitrogen and phosphorus play key role in plant growth and development. Added soil nitrogen increases protein content in wheat seeds [8], in rice grains [1] and in cotton seeds [7]. More-over phosphorus gets fixed in tropical soils of India and only 20-25% of it becomes available to the plants during the year of application which creates a sink effect. In an effort to avoid this sink effect of fixed phosphorus, use of (VAM) vesicular arbuscular mycorrhiza (*Glomus mosseae* and *Glomus fasciculatum*), phosphate solubilizing bacteria *Bacillus megaterium* (Var-Phosphaticum) and fungus (*Aspergillus awamori*) have been proved to be beneficial and economical as well as ecofriendly too.

## MATERIAL AND METHODS

The experiment was laid in a split- split plot design with 40 treatments and 2 replications. The treatments comprised of 2 sources of phosphorus (single super phosphate $S_1$ and Rock phosphate $S_2$ ) as main plot effect , 10 levels of fertilizers including inoculation with phosphate solubilizing micro-organisms (*Bacillus megaterium* var-*phosphaticum*, *Aspergillus awamori*, *Aspergillus niger*, nitrogen fixing bacteria (*Azotobacter*) and VA-mycorrhiza as bio-fertilizers and no-inoculation ($F_1$ to $F_{10}$ ) as sub-plot effect and 2 mulberry varieties (Victory-1: $V_1$ and $S_{36}$ : $V_2$ ) as sub-sub plot effect. The experimental nursery was divided in to two blocks of 40 plots each. Thus a total of eighty plots were made keeping sufficient space between each plot and block. The gross size of each plot was 1.35 m x 0.50 m. The cuttings of two selected varieties $S_{36}$ and $V_1$ (60 each) were planted in respective plots at a distance of 10 cm between cutting to cutting and 15 cm between row to row. The initial VAM spore load was found to be 2-3 spores / 20 g soil. Before planting cuttings in the nursery, furrows of shallow depth (15 cm) and 3-4 cm width were made and green house culture of VA-mycorrhizal inoculum @ 10 g / cutting (@ 20 spores/g of soil based mixed inoculum containing *Glomus fasciculatum* and *Glomus mosseae* (obtained from the Agronomy laboratory of CSR&TI, Mysore) was applied in each furrow in the form of thin layer. This was done only in respect of the treatments where VA-mycorrhizal inoculation was required. The cuttings were then planted on thin layer of VA-mycorrhizal inoculum in such a way that a proper contact between planted cuttings and the inoculum was established and the furrows were covered with the soil leaving one bud exposed. Irrigation was then followed immediately. After 15 days of planting the cuttings, the chemical fertilizers as per the treatments were applied. The irrigation was followed immediately after application of fertilizers. After 10 days of fertilizer application, the bio-fertilizer comprising of *Azotobacter Chroococcum* (as nitrogen fixer) , *Aspergillus awamori, Aspergillus niger* and *Bacillus magaterium* ( as phosphate solubilizers ) were applied to the respective plots. The inoculum of *Azotobacter chroococum* grown in the laboratory as broth culture (@ $10^7$ cfu / ml of broth) was mixed with equal quantity of broth culture (1:1 ) of phosphate solubilizers (@ $10^5$ to $10^6$ cfu / ml of broth) in a conical flask of 1000 ml capacity and was stirred thoroughly to obtain a composite broth culture of all the required micro-organisms. Finally this composite broth culture was mixed with dry powdered farm yard manure and applied @ 100 g / plot in open furrows between the rows of mulberry in the nursery and the furrows were closed. This was followed by irrigation to avoid the desiccation of live microbial cells in the soil. The data on survival percentage of saplings, average height of saplings (cm) after 60 days, average length of primary roots (cm) and leaf yield per sapling were recorded.

## RESULTS AND DISCUSSION

### Effect on survivability of saplings

The survivability of saplings was not influenced by the effect of two sources of phosphorus as main plot effect (Table-1). The similar result was obtained with the sub plot effect on the survivability of the saplings. However, sub-sub plot effect of two varieties on the survivability of saplings was found to be significant. Highest survivability of saplings (90.47%) was recorded with the variety $V_1$ after 60 days of planting in nursery followed by $S_{36}$ which showed 47.71 % of survivability. The interaction effect was also found to be non significant.

**Table-1: Effect of microbial consortium on survival % of saplings**

**A.** Main plot effect (Sources of phosphorus)

| | |
|---|---|
| $S_1$ | 69.31 |
| $S_2$ | 68.87 |

**B.** Sub plot effect (Fertilizer levels)

| | |
|---|---|
| $F_1$ | 68.62 |
| $F_2$ | 69.02 |
| $F_3$ | 70.66 |
| $F_4$ | 68.98 |
| $F_5$ | 68.56 |
| $F_6$ | 70.21 |
| $F_7$ | 67.77 |
| $F_8$ | 67.67 |
| $F_9$ | 69.53 |
| $F_{10}$ | 69.86 |

**INTERACTION EFFECT**

| **Fertilizer levels** | **$V_1$ $S_1$** | **$V_1$ $S_2$** | **$V_2$ $S_1$** | **$V_2$ $S_2$** |
|---|---|---|---|---|
| $F_1I_1$ | 92.95 | 90.25 | 44.20 | 47.10 |
| $F_2I_1$ | 90.85 | 91.20 | 42.10 | 51.95 |
| $F_3I_1$ | 93.20 | 89.35 | 50.60 | 49.50 |
| $F_4I_1$ | 90.30 | 91.05 | 47.05 | 47.55 |
| $F_5I_1$ | 88.65 | 90.00 | 48.05 | 47.55 |
| $F_6I_1$ | 91.35 | 89.65 | 50.35 | 49.50 |
| $F_7I_1$ | 91.00 | 82.20 | 48.00 | 49.90 |
| $F_8I_1$ | 88.10 | 90.50 | 46.55 | 45.55 |
| $F_9I_1$ | 94.40 | 90.35 | 48.80 | 44.60 |
| $F_{10}I_0$ * | 91.90 | 92.25 | 47.80 | 47.50 |

**C.** Sub sub plot effect (Varieties)

| | |
|---|---|
| $V_1$ | 90.47 |
| $V_2$ | 47.71 |

**Cd at 5% for**

| | |
|---|---|
| A: | NS |
| B: | NS |
| C: | 1.38 |
| AxBxC: | NS |

$S_1$ = Single super phosphate
$S_2$ = Rock phosphate
$V_1$= Victory-1
$V_2$= S36
$F_1$= ½ of recommended dose of N and P
$F_2$= 3/4th of P and ½ of N.
$F_3$= Recommended dose of P and ½ of N.
$F_4$= ½ P and 3/4th of N.
$F_5$= 3/4th N and P.
$F_6$ = Recommended dose of P and 3/4th N.
$F_7$ = ½ P and recommended dose of N.
$F_8$ = 3/4th of P and recommended dose of N.
$F_9$ = Recommended dose of N and P.
$F_{10}$= Recommended dose of N and P($I_0$)
$I_1$ =inoculation
$I_0$= No inoculation
NS= Non significant
* control

**Effect on height of the saplings after 60 days after planting**

The application of two sources of phosphorus as main plot effect have significantly influenced the height of the saplings in 60 days itself (Table-2). Among the two sources of phosphorus, maximum height was recorded with $S_2$ (27.25 cm / sapling) followed by $S_1$ (26.05 cm). Significant influence of the application of different levels and sources of fertilizer ($F_1$-$F_{10}$) as sub plot effect on height of saplings was also observed. Maximum height (32.28 cm / sapling) was recorded in the treatment $F_9$ (recommended dose of phosphorus and nitrogen with co-inoculation). This was significantly higher as compared to other fertilizer levels including control ($F_{10}$) but was at par with $F_8$ and $F_7$. Significant difference in the height of the saplings was also observed due to different varieties as sub-sub plot effect. Maximum height (34.25 cm / sapling) was recorded with the saplings of $V_1$ variety after 60 days of plantation in nursery followed by $S_{36}$ (19.06cm / sapling). Interaction effect was also significant. The co-inoculation of saplings with PSM, *Azotobacter* and VA-mycorrhiza and application of ¾ th of the recommended dose of phosphorus as RP as well as full dose of nitrogen ($F_8I_1S_2V_1$) has resulted in maximum height (45.55 cm / sapling). This was significantly higher over most of the treatments including control.

**Table-2:Effect of microbial consortium on height per sapling (cm) 60 DAP**

**A.** Main plot effect (Sources of phosphorus)

| | |
|---|---|
| $S_1$ | 26.05 |
| $S_2$ | 27.25 |

**B.** Sub plot effect (Fertilizer levels)

| | |
|---|---|
| $F_1$ | 25.45 |
| $F_2$ | 24.08 |
| $F_3$ | 26.92 |
| $F_4$ | 26.41 |
| $F_5$ | 26.33 |
| $F_6$ | 25.82 |
| $F_7$ | 31.56 |
| $F_8$ | 31.63 |
| $F_9$ | 32.28 |
| $F_{10}$ | 16.07 |

**INTERACTION EFFECT**

| **Fertilizer levels** | **$V_1$** | | **$V_2$** | |
|---|---|---|---|---|
| | **$S_1$** | **$S_2$** | **$S_1$** | **$S_2$** |
| $F_1I_1$ | 24.75 | 44.95 | 15.30 | 16.80 |
| $F_2I_1$ | 29.05 | 29.30 | 21.25 | 16.75 |
| $F_3I_1$ | 36.60 | 37.55 | 16.30 | 17.25 |
| $F_4I_1$ | 34.75 | 38.25 | 16.85 | 15.80 |
| $F_5I_1$ | 32.35 | 36.30 | 19.10 | 17.60 |
| $F_6I_1$ | 37.80 | 27.05 | 20.70 | 17.75 |
| $F_7I_1$ | 43.95 | 41.10 | 19.90 | 22.10 |
| $F_8I_1$ | 32.35 | 45.55 | 21.85 | 26.80 |
| $F_9I_1$ | 45.30 | 34.75 | 19.95 | 29.05 |
| $F_{10}I_0$ | * 17.30 | 16.05 | 16.55 | 14.40 |

**C.** Sub sub plot effect (Varieties)

| | |
|---|---|
| $V_1$ | 34.25 |
| $V_2$ | 19.06 |

**Cd at 5% for**

| | |
|---|---|
| A: | 1.12 |
| : | 2.27 |
| C: | 1.02 |
| AxBxC: | 4.57 |

$S_1$ = Single super phosphate
$S_2$ = Rock phosphate
$V_1$= Victory-1
$V_2$= S36
$F_1$= ½ of recommended dose of N and P
$F_2$= 3/4th of P and ½ of N.
$F_3$= Recommended dose of P and ½ of N.
$F_4$= ½ P and 3/4th of N.
$F_5$= 3/4th N and P.
$F_6$ = Recommended dose of P and 3/4th N.
$F_7$ = ½ P and recommended doze of N.
$F_8$ = 3/4th of P and recommended doze of N.
$F_9$ = Recommended dose of N and P.
$F_{10}$= Recommended dose of N and P($I_0$)
$I_1$ =inoculation
$I_0$= No inoculation
* control
DAP = Days after planting

**Effect on primary root length of saplings**

No significant difference between the two sources of phosphorus ( $S_1$ and $S_2$) as main plot effect on the length of primary roots of saplings was observed (Table-3). However, the sub plot effect was found to be significant. Among the sub plot effect ($F_1$-$F_{10}$), maximum length of primary roots (28.83 cm / sapling) was recorded due to treatment $F_5$. This was significantly higher as compared to all other fertilizer levels but was at par with $F_1$, $F_2$, $F_9$ and $F_{10}$. Significant difference in the primary root length of saplings was also observed due to different varieties as sub-sub plot effect. Maximum primary root length (27.49 cm / sapling) was recorded with the saplings of $V_1$ followed by $S_{36}$ (18.93 cm / sapling). The interaction effect due to different levels and sources of fertilizer with co- inoculation and varieties was found to be non- significant.

**Table-3:Effect of microbial consortium on primary root length of saplings (cm).**

**A.** Main plot effect (Sources of phosphorus)

| | |
|---|---|
| $S_1$ | 23.22 |
| $S_2$ | 23.20 |

**B.** Sub plot effect (Fertilizer levels)

| | |
|---|---|
| $F_1$ | 27.73 |
| $F_2$ | 23.90 |
| $F_3$ | 20.36 |
| $F_4$ | 18.91 |
| $F_5$ | 28.83 |
| $F_6$ | 22.37 |
| $F_7$ | 18.32 |
| $F_8$ | 18.68 |
| $F_9$ | 28.36 |
| $F_{10}$ | 24.65 |

**INTERACTION EFFECT**

| **Fertilizer levels** | **$V_1$** | | **$V_2$** | |
|---|---|---|---|---|
| | **$S_1$** | **$S_2$** | **$S_1$** | **$S_2$** |
| $F_1I_1$ | 27.80 | 39.70 | 25.05 | 18.40 |
| $F_2I_1$ | 25.10 | 32.70 | 20.50 | 17.30 |
| $F_3I_1$ | 24.40 | 18.75 | 21.05 | 17.25 |
| $F_4I_1$ | 20.10 | 20.45 | 16.80 | 18.30 |
| $F_5I_1$ | 29.35 | 42.15 | 25.60 | 18.25 |
| $F_6I_1$ | 25.50 | 21.90 | 22.80 | 19.30 |
| $F_7I_1$ | 20.25 | 20.35 | 16.80 | 15.90 |
| $F_8I_1$ | 18.90 | 22.15 | 17.85 | 15.85 |
| $F_9I_1$ | 41.65 | 39.10 | 16.00 | 16.70 |
| $F_{10}I_0$ | *27.60 | 31.95 | 21.40 | 17.65 |

**C.** Sub sub plot effect (Varieties)

| | |
|---|---|
| $V_1$ | 27.49 |
| $V_2$ | 18.93 |

**Cd at 5% for**

| | |
|---|---|
| A: | NS |
| B: | 4.97 |
| C: | 2.32 |
| AxBxC: | NS |

$S_1$ = Single super phosphate
$S_2$ = Rock phosphate
$V_1$= Victory-1
$V_2$= S36
$F_1$= ½ of recommended dose of N and P
$F_2$= 3/4th of P and ½ of N.
$F_3$= Recommended dose of P and ½ of N.
$F_4$= ½ P and 3/4th of N.
$F_5$= 3/4th N and P.
$F_6$ = Recommended dose of P and 3/4th N.
$F_7$ = ½ P and recommended dose of N.
$F_8$ = 3/4th of P and recommended doze of N.
$F_9$ = Recommended dose of N and P.
$F_{10}$= Recommended dose of N and P($I_0$)
$I_1$ =inoculation
$I_0$= No inoculation
NS= Non significant
* control

**Effect on leaf yield per sapling**

Significantly higher leaf yield of 28.88 g / sapling was recorded in $S_1$ as compared to $S_2$ (25.45 g/ sapling) due to main plot effect. Sub plot effect on the leaf yield per sapling was also found to be significant. Among the sub plot effect ($F_1$-$F_{10}$), maximum leaf yield (38.03 g / sapling) was recorded in the treatment $F_7$. Significant difference in leaf yield per sapling was also observed due to different varieties as sub-sub plot effect. Maximum leaf yield (30.32 g / sapling ) was recorded with the variety $V_1$ followed by $S_{36}$ (24.01 g / sapling). The interaction effect was also found to be significant ( Table- 4). Maximum leaf yield (45.15 g / sapling) was recorded in $V_1$ due to inoculation with PSM, *Azotobacter* and VA-mycorrhiza with application of ½ of the recommended dose of phosphorus as SSP as well as 3/4th of nitrogen ($F_4I_1S_1V_1$). This was significantly higher over majority of the treatments including control.

**Table-4: Effect of microbial consortium on leaf yield per sapling (g)**

**A.** Main plot effect (Sources of phosphorus)

| | |
|---|---|
| $S_1$ | 28.88 |
| $S_2$ | 25.45 |

**B.** Sub plot effect (Fertilizer levels)

| | |
|---|---|
| $F_1$ | 20.32 |
| $F_2$ | 24.73 |
| $F_3$ | 30.03 |
| $F_4$ | 27.33 |
| $F_5$ | 25.68 |
| $F_6$ | 21.40 |
| $F_7$ | 38.03 |
| $F_8$ | 33.48 |
| $F_9$ | 26.05 |
| $F_{10}$ | 24.83 |

**INTERACTION EFFECT**

| **Fertilizer levels** | **$V_1$** | | **$V_2$** | |
|---|---|---|---|---|
| | **$S_1$** | **$S_2$** | **$S_1$** | **$S_2$** |
| $F_1I_1$ | 22.10 | 21.65 | 19.65 | 17.90 |
| $F_2I_1$ | 24.15 | 32.25 | 22.05 | 20.50 |
| $F_3I_1$ | 39.85 | 27.25 | 35.25 | 17.80 |
| $F_4I_1$ | 45.15 | 15.80 | 33.00 | 15.40 |
| $F_5I_1$ | 37.05 | 18.95 | 30.60 | 16.05 |
| $F_6I_1$ | 25.70 | 23.25 | 23.35 | 13.30 |
| $F_7I_1$ | 42.05 | 43.40 | 38.65 | 28.05 |
| $F_8I_1$ | 30.88 | 43.00 | 26.50 | 33.65 |
| $F_9I_1$ | 19.80 | 33.95 | 17.05 | 33.40 |
| $F_{10}I_0$ | * 23.05 | 37.35 | 21.95 | 16.20 |

**C.** Sub sub plot effect (Varieties)

| | |
|---|---|
| $V_1$ | 30.32 |
| $V_2$ | 24.01 |

**Cd at 5% for**

| | |
|---|---|
| A: | 3.04 |
| B: | 1.93 |
| C: | 0.88 |
| AxBxC: | 3.93 |

$S_1$ = Single super phosphate
$S_2$ = Rock phosphate
$V_1$= Victory-1
$V_2$= S36
$F_1$= ½ of recommended dose of N and P
$F_2$= 3/4th of P and ½ of N.
$F_3$= Recommended dose of P and ½ of N.
$F_4$= ½ P and 3/4th of N.
$F_5$= 3/4th N and P.
$F_6$ = Recommended dose of P and 3/4th N.
$F_7$ = ½ P and recommended dose of N.
$F_8$ = 3/4th of P and recommended dose of N.
$F_9$ = Recommended dose of N and P.
$F_{10}$= Recommended dose of N and P($I_0$)
$I_1$ =inoculation
$I_0$= No inoculation
* control

The co-inoculation studies in general indicated highly synergistic effect on the survivability of $V_1$ mulberry saplings in the nursery signifying importance of the use of microbial consortium in mulberry cultivation. Right from the development of primary root to the production of vigorously growing saplings in the nursery, the synergistic effect of co-inoculation was highly evident especially on the variety $V_1$. The interaction of various fertilizer levels and sources also revealed better performance in inoculated treatments. This is attributed to the enhanced microbial interaction due to inoculation of different beneficial micro-organisms in the rhizosphere of mulberry where they play a pivotal role in various physiological processes including solubilization and transportation of minerals to the plants. This accentuates the availability of plant nutrients in the rhizosphere thereby improving the growth and yield of plants. The work of [2] on the combined inoculation of sorghum with *Azospirrillum* and *phosphobacteria* in increasing grain yield also supports the present findings. The non significant difference in the leaf yield of sapling in the nursery between inoculated and uninoculated treatments justifies the significant beneficial effect of the micro-organisms used in the present study as supplement to the chemical fertilizers nitrogen and phosphorus. This also indicates that the use of phosphate solubilizing micro-organisms, nitrogen fixing bacteria and VA-mycorrhiza not only supplements nitrogen and phosphorus, it can also improve the growth and leaf production in mulberry even after curtailing chemical fertilizers to a great extent. The results of [13] also demonstrated similarly significant beneficial effect of co-inoculation in increasing growth of *Leucaena* *leucocephala* using phosphate solubilizing bacteria and VA-mycorrhiza. Similar results were also obtained with *Acacia confusa* and *Acacia mangium* [13] and in mulberry [5].

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