

EVALUATION OF BIO-FERTILISER (BIO-INOCULANT) CONSORTIA AND THEIR EFFECT ON PLANT GROWTH PERFORMANCE OF SANDALWOOD (*SANTALUM ALBUM*) SEEDLINGS

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An attempt was made to evaluate the efficacy of bio-inoculant in single form and as consortium (two or more) involving various treatments (T1 to T12) for the growth performance of *Santalum album* seedlings. Analysis of growth parameters (at 90th and 180th day) included shoot length (20.4 cm and 52.50 cm), collar diameter (2.88 mm and 4.27 mm), root length (11.76 cm and 26.00 cm), shoot dry weight (8.90 g and 15.18 g), root dry weight (8.10 g and 15.21 g) and total dry weight (16.99 g and 30.39 g). The parameters revealed growth augmentation (shoot length, collar diameter, root length) at 180th day in T12 plants with bio-fertiliser consortia compared to control (T1) (19.80 cm, 3.42 mm, 8.31 cm) and single bio-inoculants (T2) (21.80 cm, 2.83 mm, 11.92 cm). Growth indices, viz., root shoot ratio (0.91 and 1.01), volume index (266.04 and 1462.82) and Dickson's quality index (2.08 and 2.28) at 90th and 180th day, with treatment (T12) involving arbuscular mycorrhizal fungi (AMF) + *Azospirillum* + *Azotobacter* + phosphobacteria + potash mobiliser, performed better than control and single bio-inoculant. This study highlights that the efficacy of two or more bio-inoculant in the form of consortium enhances the growth of *Santalum album* plants in nursery conditions.

Keywords: *Santalum album*, *Azotobacter*, phosphobacteria, potash mobiliser, microbial consortia, bio-inoculant

INTRODUCTION

Bio-fertilisers (bio-inoculant) are an effective, cheap and renewable supplement compared to chemical fertilisers. Considering the problem of chemical fertilisers, it has been globally recommended to incorporate bio-inoculants in integrated plant nutrition system (IPNS) to meet the nutritional demand of plants (Sharma & Chaubey 2015). Application of beneficial micro-biomes as bio-inoculants in sustainable agriculture practices has emerged as innovative and environment-friendly technology for improving soil fertility and plant growth (Kumar et al. 2022, Adesemoye et al. 2009, Bertola et al. 2019, Ullah et al. 2019, Murgese et al. 2020, Fasusi et al. 2021). Consortium of bio-inoculants showed positive influence on plant growth and yield in addition to soil organic carbon, organic matter and available phosphorous as well as higher concentration of nutrients in soil (Pellegrini et al. 2021). Intervention of bio-inoculants like arbuscular mycorrhizal fungi (AMF), *Rhizobium*, *Azotobacter* and phosphate solubilising bacteria

(PSB) were reported to be more effective in biomass production and AMF colonisation of nursery seedlings of *Albizia lebbek* (Pavan 2011). The influence of microbial inoculants had been proven to be the most efficient for better growth performance of seedlings at nursery stage of most of forestry tree species of tropical and sub-tropical regions (Asif et al. 2018).

In reference to *Santalum album*, previous study has reported that the root zone of *S. album* contains more nitrogen fixing bacteria and AMF, indicating the inseparable association of *S. album* with the host tree species and microorganisms (Subbarao et al. 1990). It is also reported that the spores extracted from the rhizosphere of *S. album* are predominantly of *Glomus* and *Gigaspora* species, where the association of *Glomus* species with *S. album* seedlings is more compatible in relation to water uptake and nutrient content (Subbarao et al. 1990, Thappar et al. 1992, Nagaveni et al. 1997). Bio-inoculant consortia, during the initial stage of growth (nursery stage)

increases plants vigour and enhance growth by improving root activity and robustness of plantlets for proper field establishment (Arade et al. 2020). *Santalum album* is a semi-parasitic plant depending on the host roots for initial survival and establishment. Application of bio-inoculants as growth promoting substance has not been experimented periodically in *S. album*. In addition, no report is available on the efficacy of the consortium of bio-inoculants on the growth performance of *S. album* seedlings in nursery. The present study was an attempt to understand the efficacy of various bio-fertilisers as single inoculants and in combination, contributing to the growth performance of *S. album* seedlings.

MATERIALS AND METHODS

The experiment was conducted in the nursery of Van Vignan Kendra (VVK), Institute of Wood Science and Technology (IWST), Gottipura. The bio-inoculants were supplied by the Institute of Forest Genetics and Tree Breeding (IFGTB), Coimbatore. The bio-inoculants were maintained at ambient room temperature, i.e., 28 ± 2 °C and used for further work at IWST, Bengaluru. A short compatibility study was carried out by streaking dual inoculants on a solidified nutrient agar medium, to exclude the antagonism among bio-inoculants. Primarily, each of the co-inoculated strains was grown in nutrient agar medium at 30 °C for at least 3–6 days. The first inoculant streak was allowed to grow at 30 °C for 3 days. The second strain was streaked at an angle of approximately 90 ° going outward from the emerged colonies of the first strain. The second colony was incubated at 30 °C for another 3 days (Belkar & Gade 2012).

The experiment consisted of 12 treatments (T1 to T12) where T1 represented control, while T2–T6 represented the single bio-inoculant treatment of *S. album* seedlings, viz., AMF, *Azospirillum*, *Azotobacter*, phosphobacteria and potash mobiliser. Treatments T7–T12 represented the bio-inoculant consortium AMF + *Azospirillum*, AMF + *Azotobacter*, AMF + phosphobacteria, AMF + potash mobiliser, AMF + *Azospirillum* + *Azotobacter* + phosphobacteria and AMF + *Azospirillum* + *Azotobacter* + phosphobacteria + potash mobiliser. Treatments T11 and T12 represented the microbial consortia of bio-inoculants of two and above.

Effectiveness of the bio-inoculant formulation was tested on the growth of *S. album* seedlings

maintained at a temperature of 22–26 °C and the light intensity of 2690.98–10,763.90 lux (full daylight) in nursery conditions. The plant growth parameters, i.e., shoot length, collar diameter, root length, shoot and root dry weight and total dry weight were recorded after the application of bio-inoculants.

Measurement of growth parameters

Shoot length, collar diameter and root length

The plant height was measured from the base of the plant to the terminal growing point of the main stem in centimeters (cm). The collar diameter was measured in millimeters with the help of a digital Vernier calliper. The seedlings were removed from poly bags gently without damaging the roots, and the root length was measured in centimeters (cm) from the collar region to the tip of the root. The observations and the data were collected on the 90th and 180th day of the plants.

Shoot/root dry weight, total dry weight

The root and shoot region of the seedlings were separated and the samples were dried at 85 °C for 48 hours and the dry weights were recorded until constant weights were obtained in grams (g) per seedling. Total dry weight was calculated by summing up the dry weight of the shoot as well as the root (shoot dry weight + root dry weight). The primary data were focussed on 180th day plants, however, data from 90th day plants (replicates maintained separately under similar conditions) were also collected.

Growth indices

Absolute growth rate (AGR)

The rate of increase in growth variable (dry weight) at time 't' is called absolute growth rate (AGR). It was measured by differential coefficient of 'w' with respect of time 't'. Absolute growth rate was calculated for two growth variables by the following formula and expressed in mg g⁻¹ day⁻¹ (Wareing & Philips 1981).

$$AGR = \frac{W_2 - W_1}{t_2 - t_1}$$

where W_1 and W_2 are the means of plant dry weights at times t_1 and t_2 .

Relative growth rate (RGR)

Relative growth rate (RGR) indicates rate of growth per unit dry matter. It is alike to compound interest wherein the incremental growth in any interval adds to the capital for succeeding growth. This rate of increment is known as RGR and was calculated by using the following formula and expressed in $\text{mg g}^{-1} \text{day}^{-1}$ (Fisher 1921, Hoffmann & Poorter 2002).

$$\text{RGR} = \frac{\ln W_2 - \ln W_1}{t_2 - t_1}$$

where $\ln W_1$ and $\ln W_2$ are the means of the natural logarithm transformed plant dry weights at times t_1 and t_2 .

Quality indices

Growth indices related to the relative growth of the photosynthetic parts of the plant to light, temperature, moisture and soil nutrients were calculated as below:

Root-shoot ratio

Root-shoot ratios are indicative of plant response to growing conditions, but ratios are not a definitive measure because values change as plants grow. In the current study 180th day plants were assessed to elucidate the difference in treatment. Root-shoot ratio was measured by dividing the root dry weight with shoot dry weight.

Sturdiness quotient

The data on the morphological features of the seedlings were further used to compute sturdiness quotient. The sturdiness quotient (SQ) refers to the ratio of the height of the seedling to the root collar diameter and expresses the vigour and robustness of the seedling. The ideal value for a seedling to be considered sturdy is less than six (Jaenicke 1999). The sturdiness quotient was calculated by dividing the shoot length with collar diameter (Ritchie 1985).

Volume index (VI)

Volume index (VI) was determined by multiplying diameter² (cm) × height (cm), where, height

includes root and shoot length (Kumaran & Surendran 1999).

Dickson's quality index (DQI)

Dickson's quality index (DQI) was determined by dividing the total dry weight of the seedlings with sum value of division of height (cm)/diameter (mm) and shoot dry weight (g)/root dry weight (g) (Dickson et al. 1960).

Microbial inoculation effect (MIE):

The microbial inoculation effect (MIE) was calculated based on the formula of Bagyaraj (1992), to understand the effect of introduced bio-inoculants compared with the inherent conditions. The MIE is calculated by dividing the difference in the dry weight of inoculated plants and the means of dry weight of uninoculated plants with the dry weight of inoculated plants and expressed in percentage.

Statistical analysis

The data were subjected to analysis of variance and the significant variances among the means were compared by Duncan's multiple range test (DMRT) using SPSS (Version 10.0) statistical software to determine the effects due to treatments (Little & Hills 1978).

RESULTS AND DISCUSSION

In a previous research evaluating microbial consortia versus single bio-inoculants, microbial consortia was found to increase the efficiency of crop production particularly under challenging environmental conditions, thus, compatibility of the bio-inoculants in the consortia is fundamental (Bradacova et al. 2019). Dual streak assay experiment to understand the compatibility between the microbial bio-inoculants revealed that these microorganisms were compatible with each other and they did not show antagonistic interaction during their growth (Figure 1). The dual streak test facilitated the compatibility of the appropriate inoculum, which is a key step towards the development of a successful bio-fertiliser, where bio-fertiliser consortia appear to have greater efficacy on the improvement of plant growth than single bio-inoculants (Stamenkovic 2018, Thomludi 2019).

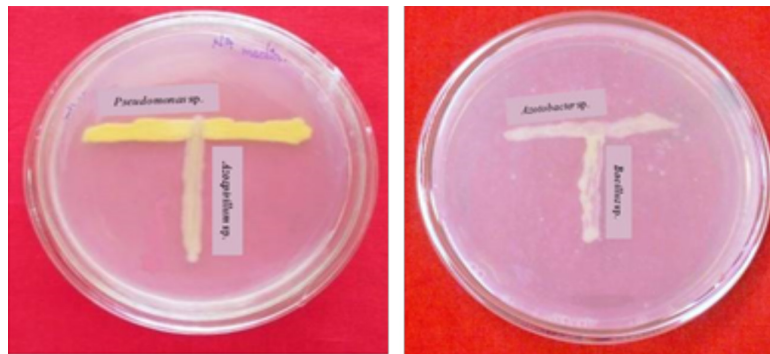


Figure 1 Dual assay test showed the absence of antagonism between the bio-inoculants

Growth parameters/biometric observation

Shoot length, collar diameter and root length

Most approaches for plant growth advancement involve the use of single bio-inoculant as biofertilisers, while only few contemplate microbial consortia products, i.e., the combination of two or more microbial species, as a valid strategy to increase community efficiency and promote plant growth (Bradacova et al. 2019, Vishwakarma et al. 2020). In the present study plant growth performance was shown through plant biometric observations, where plant height (shoot length) is an important morphological and developmental phenotype that directly indicates overall plant growth. The plants receiving bio-fertilisers in the form of consortia of T12 recorded maximum shoot length of 20.4 cm at 90th day and 52.50 cm at 180th day. The next maximum growth in shoot length/height, i.e., 17 cm at 90th day and 43.10 cm at 180th day, was observed in the treatment, T11 (Table 1). The primary growth parameters such as plant height and number of leaves increased by the combined application of microbial consortia when compared to control plants (Mohan et al. 2022). The least plant height of 13.70 cm and 19.80 cm at 90th and 180th day were observed in control plants, thereby the results obtained on shoot height response to T11 and T12 were in concurrence with the findings of Mohan et al. (2022) and Mounika et al. (2017). The lowest plant height of 19.80 cm was observed in the control treatment, T1 (without any bio-inoculants), which was at par with all other treatments except T9. The findings indicate that consortia of bio-inoculants associated with all the stages of plant growth for

better supply of nitrogen by biological means, efficient solubilisation of unavailable phosphorus to available phosphorus and mobilisation of potassium (Liu et al. 2012, Mahmud et al. 2020).

The collar diameter was 2.88 mm and 4.27 mm at 90th and 180th day for those plants received T12. Comparatively, the collar diameter was less with control (1.77 mm and 3.42 mm) and T2 (1.85 mm and 2.83 mm) at 90th and 180th day respectively, which was in resemblance to the outcomes of Mohan and Rajendran (2019) and Muthu Kumar et al. (2021, 2022). The collar diameter recorded for T12 plants was on par with the T11 plants and significantly the lowest collar diameter was recorded in the control treatment, T1, at 90th day. However, at 180th day, T12 plants with collar diameter of 4.27 mm was at par with treatments T7, T8 and T9, while the least collar diameter was 2.86 mm in T3 plants, specifying the relevance of two or more microbial inoculants or consortia.

The impact of microbial consortium was obvious in the roots of treated plants, where the 90th day observation showed highest root length (11.76 cm and 26.00 cm) in T12 plants associated with consortia bio-inoculants. The highest root length at 90th day was observed in T12 plants followed by T10 plants with 10.17 cm, while the lowest root length was 2.80 cm recorded in the control (T1). At 180 days, root length was significantly maximum (26.00 cm) in T12 plants followed by T1 and the minimum root length was observed in the control (T1, 8.31 cm). In the same way, microbial inoculants increase seedling height, number of leaves and leaf area in *S. album*, and there was a significant increase in shoot and root length recorded in *Azadirachta* seedlings inoculated with different biofertilisers (Binu et al. 2015, Gunasundari et al. 2022). A combination

Table 1 Growth parameters observation of treated plants (No. of replications = 5)

| No. | Treatments | Shoot length | | Collar diameter | | Root length | |
|-----|-----------------|--------------------|----------------------|----------------------|---------------------|--------------------|----------------------|
| | | 90 days | 180 days | 90 days | 180 days | 90 days | 180 days |
| 1 | T ₁ | 13.7 ^c | 19.80 ^d | 1.77 ^d | 3.42 ^{bcd} | 2.80 ^h | 8.31 ^g |
| 2 | T ₂ | 14.6 ^{bc} | 21.80 ^d | 1.85 ^{cd} | 2.83 ^{cd} | 3.60 ^{gh} | 11.92 ^{de} |
| 3 | T ₃ | 15.4 ^{bc} | 20.80 ^d | 1.92 ^{cd} | 2.86 ^d | 5.72 ^f | 15.00 ^c |
| 4 | T ₄ | 14.6 ^{bc} | 23.20 ^{cd} | 2.30 ^{abcd} | 2.94 ^{cd} | 7.13 ^e | 9.00 ^{fg} |
| 5 | T ₅ | 15.2 ^{bc} | 21.40 ^d | 2.21 ^{bcd} | 3.48 ^{bcd} | 4.28 ^g | 13.00 ^{cde} |
| 6 | T ₆ | 16.2 ^{bc} | 27.70 ^{bcd} | 2.12 ^{bcd} | 3.42 ^{bcd} | 7.61 ^{de} | 14.00 ^{cd} |
| 7 | T ₇ | 16 ^{bc} | 30.70 ^{bcd} | 1.89 ^{cd} | 3.81 ^{ab} | 5.81 ^f | 12.00 ^{de} |
| 8 | T ₈ | 15.2 ^{bc} | 35.48 ^{bcd} | 2.00 ^{bcd} | 4.26 ^a | 8.00 ^d | 10.97 ^{efg} |
| 9 | T ₉ | 16.2 ^{bc} | 38.90 ^{abc} | 2.45 ^{abc} | 3.91 ^{ab} | 9.16 ^c | 11.50 ^{def} |
| 10 | T ₁₀ | 14.4 ^{bc} | 27.50 ^{bcd} | 2.09 ^{bcd} | 3.53 ^{bc} | 10.17 ^b | 15.31 ^{bc} |
| 11 | T ₁₁ | 17 ^b | 43.10 ^{ab} | 2.55 ^{ab} | 3.36 ^{bcd} | 9.71 ^{bc} | 18.00 ^b |
| 12 | T ₁₂ | 20.4 ^a | 52.50 ^a | 2.88 ^a | 4.27 ^a | 11.76 ^a | 26.00 ^a |
| | CD @ 0.05 | 2.76 | 6.42 | 0.62 | 0.63 | 0.85 | 2.84 |
| | SD | 1.66 | 9.89 | 0.31 | 0.46 | 2.67 | 4.51 |

Superscript alphabets represent the comparison of means of treatment and level of significance (CD @ 0.05), CD = critical difference, SD = standard deviation

Table 2 Growth parameters observation of treated plants

| No. | Treatments | Shoot dry weight | | Root dry weight | | Total dry weight | |
|-----|-----------------|--------------------|---------------------|----------------------|----------------------|---------------------|----------------------|
| | | 90 days | 180 days | 90 days | 180 days | 90 days | 180 days |
| 1 | T ₁ | 0.31 ^g | 3.56 ^d | 0.09 ^f | 2.06 ^g | 0.41 ^h | 5.62 ^g |
| 2 | T ₂ | 0.50 ^{fg} | 4.05 ^d | 0.35 ^{ef} | 3.11 ^{fg} | 0.85 ^{gh} | 7.17 ^{fg} |
| 3 | T ₃ | 1.39 ^{ef} | 3.77 ^d | 0.84 ^{def} | 2.95 ^{fg} | 2.23 ^{fgh} | 6.72 ^g |
| 4 | T ₄ | 2.16 ^{de} | 6.31 ^{cd} | 1.83 ^{cdef} | 4.68 ^{efg} | 3.99 ^{def} | 10.99 ^{efg} |
| 5 | T ₅ | 2.11 ^{de} | 6.96 ^{cd} | 0.96 ^{def} | 5.59 ^{ddef} | 3.06 ^{efg} | 12.54 ^{ef} |
| 6 | T ₆ | 3.82 ^c | 8.80 ^c | 2.05 ^{bcde} | 7.64 ^{bcde} | 5.87 ^{cd} | 16.44 ^{de} |
| 7 | T ₇ | 2.72 ^d | 9.62 ^{bc} | 1.91 ^{cde} | 9.04 ^{bcd} | 4.63 ^{de} | 18.66 ^{cd} |
| 8 | T ₈ | 3.85 ^c | 8.55 ^c | 2.32 ^{bcd} | 6.24 ^{cdef} | 6.17 ^{cd} | 14.80 ^{de} |
| 9 | T ₉ | 5.37 ^b | 12.35 ^{ab} | 3.58 ^{bc} | 11.08 ^{ab} | 8.95 ^b | 23.43 ^{bc} |
| 10 | T ₁₀ | 4.98 ^b | 13.37 ^a | 3.03 ^{bc} | 9.81 ^{bc} | 8.01 ^{bc} | 23.18 ^{bc} |
| 11 | T ₁₁ | 5.95 ^b | 13.58 ^a | 3.70 ^b | 11.20 ^{ab} | 9.65 ^b | 24.78 ^{ab} |
| 12 | T ₁₂ | 8.90 ^a | 15.18 ^a | 8.10 ^a | 15.21 ^a | 16.99 ^a | 30.39 ^a |
| | CD @ 0.05 | 0.98 | 3.41 | 1.78 | 4.17 | 2.34 | 5.62 |
| | SD | 2.40 | 3.90 | 2.05 | 3.83 | 4.41 | 7.68 |

Superscript alphabets represent the comparison of means of treatment and level of significance (CD @ 0.05), CD = critical difference, SD = standard deviation

of *Azotobacter* + *Pseudomonas* + *Rhizobium* + vesicular-arbuscular mycorrhiza (VAM) (AMF) + PSB was proven to be effective on tissue culture raised planting material of *S. album* for survival

and growth (Arade et al. 2020). Overall, the results corresponded with the reports of Bose et al. (2022) and Mohan & Rajendran (2019), where the plants treated with microbial consortia

were recorded with maximum plant height, collar diameter and root length than control.

Shoot dry weight, root dry weight and total dry weight

Microbial consortia exerted a significant influence on *S. album* growth characteristics which were also assessed through shoot/root and total dry weight. Several studies revealed a relation between shoot dry weight and inoculation where biofertilisers increased shoot dry weight up to 28.8 to 45.2% (Gholami et al. 2009). In this study, an attempt was made to know the efficacy of bio-fertilisers in relation to shoot dry weight, root dry weight and total dry weight of *S. album* (Table 2). The T12 produced the maximum shoot and root dry weight of 15.18 g and 15.21 g, with total dry weight being 30.39 g on the 180th day plants. The control (T1) and single bio-inoculants (T2 and T3) produced minimum shoot and root dry weight (3.56 g and 2.06 g), summing up the total dry weight to 5.62 g. Santos et al. (2019), reported that bio-inoculants increase shoot and root dry weight of 180th day plants, and further, the co-inoculation of bio-inoculants had positive effects on the total shoot and root dry weight (Wang et al. 2019).

The additional experiment conducted on the 90th day plants of a separate lot similar conditions, also revealed the desired impact of bio-inoculants on the plants. The T12 plants showed maximum total dry weight of 16.99 g, when compared to T1 (0.41 g), T2 (0.85 g) and T3 (2.23 g) plants respectively. This supplementary experiment helped to interpret the enduring (in relation to the growth character) effect of bio-inoculants on the shoot and root dry weight of the plants, i.e., the total dry weight of T12 plants at 90th day was 16.99 g, which then increased to 30.39 g at 190th day.

The above findings were in accordance with the study of Kumaran & Surendran (1999) and Mohan & Rajendran (2019), apart from the investigations of Archana Sharma & Chaubey (2015). In general, the impact of the bio-inoculants on growth is because of the microbe's tremendous mobilising power of the nutrients to the root zone, especially by promoting the insoluble form of phosphate to soluble form by producing organic acids and fixing nitrogen (Somani 1987).

Growth indices

Absolute growth rate (AGR) and relative growth rate (RGR)

The weight of dry matter accumulated in plant is an index of the plant growth. Mean values of AGR and RGR based on dry matter (g/day) for the treatments (T1 to T12) at 180th day are presented in Table 3. The overall data indicated the dry matter was more in T9, T10, T11 and T12 plants treated with microbial consortia and less in the single bio-inoculant plants (T2, T3, T4) and control (T1). The increase in dry matter, due to the positive effects of multiple bio-inoculants on the AGR of plants, may be due to an elevated level of internal plant hormone, well-developed root structure and high dry matter accumulation (Abdelmoaty et al. 2022). Many researchers point out that RGR is size dependent, i.e., individuals with a smaller initial size (variables) have a larger RGR (Turnbull et al. 2008, Rose et al. 2009, Rees et al. 2010). Thereby, important relationships are hidden and it may be difficult to tell whether a tree grows slowly because it is

Table 3 Absolute growth rate and relative growth rate of treated and non-treated plants

| No. | Treatments | AGR | RGR |
|-----|-----------------|----------------------|---------------------|
| 1 | T ₁ | 0.058 ^e | 0.029 ^a |
| 2 | T ₂ | 0.070 ^{de} | 0.024 ^a |
| 3 | T ₃ | 0.050 ^e | 0.013 ^{bc} |
| 4 | T ₄ | 0.078 ^{de} | 0.011 ^{bc} |
| 5 | T ₅ | 0.105 ^{cd} | 0.016 ^b |
| 6 | T ₆ | 0.117 ^{bcd} | 0.011 ^{bc} |
| 7 | T ₇ | 0.096 ^{de} | 0.015 ^{bc} |
| 8 | T ₈ | 0.096 ^{de} | 0.010 ^{bc} |
| 9 | T ₉ | 0.161 ^{ab} | 0.011 ^{bc} |
| 10 | T ₁₀ | 0.169 ^a | 0.011 ^{bc} |
| 11 | T ₁₁ | 0.168 ^a | 0.011 ^{bc} |
| 12 | T ₁₂ | 0.149 ^{abc} | 0.006 ^c |
| | CD @ 0.05 | 0.049 | 0.006 |
| | SD | 0.04 | 0.01 |

Superscript alphabets represent the comparison of means of treatment and level of significance (CD @ 0.05), CD = critical difference, SD = standard deviation, AGR = absolute growth rate, RGR = relative growth rate

large or because it is pursuing a slow growth strategy (Pommerening 2015). Accordingly, the results were in coherence, where the RGR of T12 plants was 0.006, while other treatments T2 to T11 were in the range between 0.11 to 0.24, with control being the maximum of 0.29. Although the amount of dry matter of the plant increased with time, the general belief that a seedling with a higher RGR is inherently more efficient than one with a lower RGR has obscured understanding and has caused some confusion (South 1995, Parviz Rezvani-Moghaddam 2020).

Quality indices

Root-shoot ratio and sturdiness quotient

Nursery practices are favorable for maintaining physiological processes of seedlings as there are more probabilities for loss of small absorbing roots during lifting and handling of nursery stock, often leading to dehydration of transplanted trees (Kozłowski 1975, Kozłowski & Davies 1975). Hence, important requirements for survival of transplanted plants are a high root-shoot ratio and rapid growth of roots into a large volume of soil in order to maintain high rates of absorption of water and mineral nutrients (Theodore et al. 1997). The root-shoot ratio relates to the water absorbing area (roots) to the transpiring area (shoot) and a good ratio, one which indicates a healthy plant, is 1:1 to 2:1 root-shoot mass

(Jaenicke 1999). The maximum root-shoot ratio observed was 0.91 at 90th day and 1.01 at 180th day in the T12 plants receiving consortium bio-inoculants (Figure 2). The root-shoot response had a desirable output with bi-inoculants consortia treated plants, specifically in T7, T9 and T10 plants apart from T12 plants. The root-shoot response was less in T1 (control) plants and the lowest (< 0.1) was in T2 plants at 90th day, however at 180th day T2 plants root-shoot ratio was above 0.8, indicating the necessity to further understand AMF species suitability in haustorium mechanism in the semi-parasitic *S. album*.

Sturdiness quotient, although a good indicator of the ability to withstand physical damage in all stock-types, is of particular importance to polybag-grown seedlings where the sturdiness quotient can get very high on undesirable spindly stock (Durvea 1985). The sturdiness quotient was in the range 6 to 9 for the 90th day observation, where the quotient was 8.88 for T1 (control) plants and 6.61 for T4 plants (Figure 3). The sturdiness quotient at 180th day was also in the range of 6 to 9, but the quotient was above 10 for T9 (10.28), T11 (13.06) and T12 (12.24) plants, as reported by Durvea (1985). The findings, thereby, emphasize the poly-bag size and age/growth stage of the microbial consortia fed *S. album*. However, nursery seedlings with sturdiness quotients greater than six were seriously damaged when exposed to wind and drought (Roller 1977).

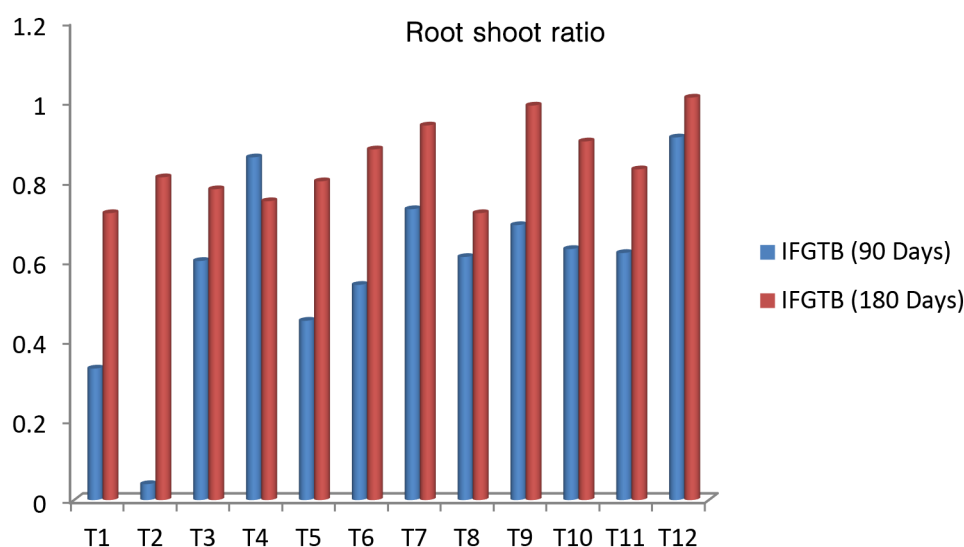


Figure 2 Root shoot ratio depicting different bio-inoculant treatments (T1 to T12) (SD: 0.22 for 90 days, SD: 0.10 for 180 days)

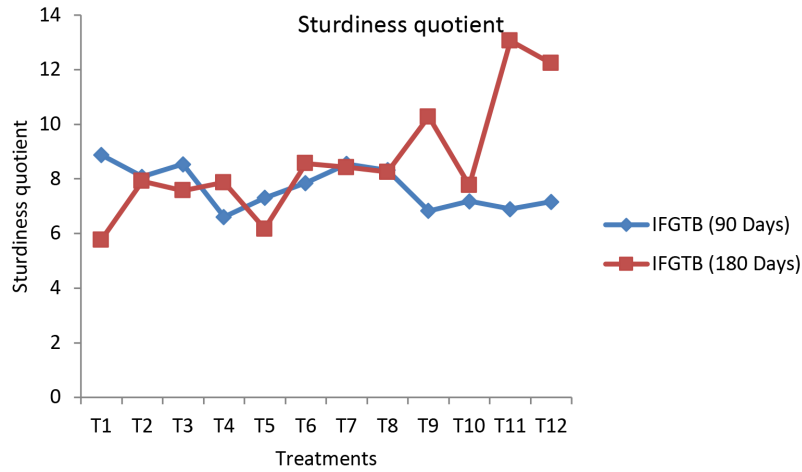


Figure 3 Sturdiness quotient depicting different bio-inoculant treatments (T1 to T12) (SD: 0.75 for 90 days, SD: 2.10 for 180 days)

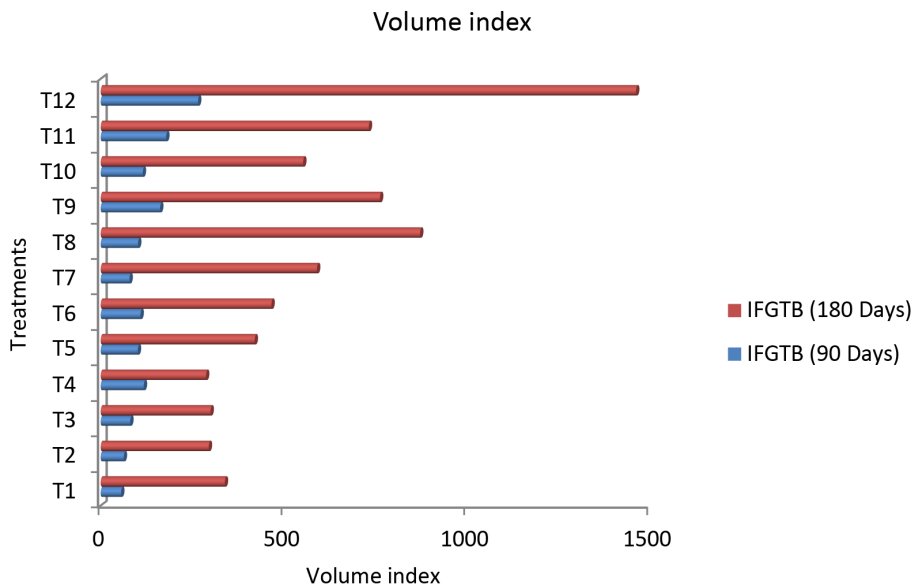


Figure 4 Volume indices depicting different bio-inoculant treatments (T1 to T12) (SD: 56.27 for 90 days, SD: 324.33 for 180 days)

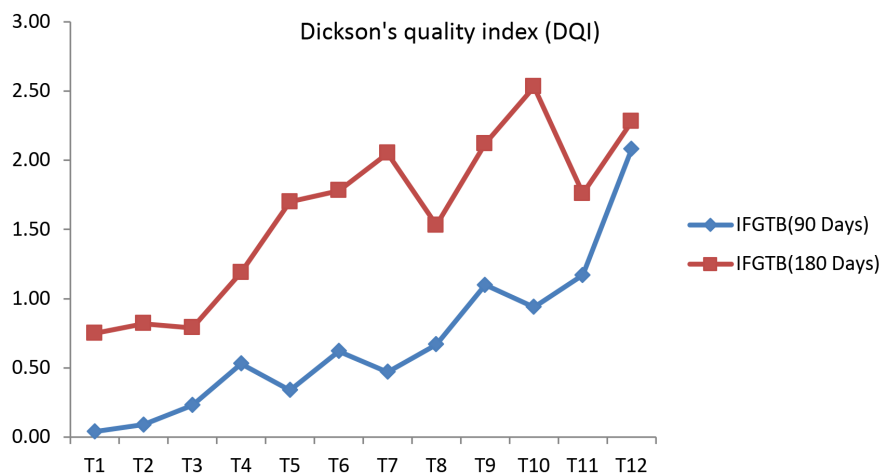


Figure 5 Dickson's quality index (DQI) depicting different bio-inoculant treatments (T1 to T12) (SD: 0.55 for 90 days, SD: 0.58 for 180 days)

Volume index and Dickson's quality index

The data on the VI and DQI on *S. album* showed that the plants receiving bio-fertilisers showed greater index, wherein the plants with multiple bio-inoculants, i.e., T12 plants, showed the maximum VI (266.04 and 1462.82) and DQI (2.08 and 2.28) at 90th and 180th day, respectively (Figure 5). On the other hand the VI of T1 (control) plant was 339.18, which was at par to the VI of T2 (295.35), T3 (300.13) and T4 (287.81) plants fed with single bio-inoculants during the observation at 180th day (Figure 4). Also, the DQI was less (0.04 and 0.75, 0.09 and 0.82, 0.23 and 0.79) in T1 (control), single inoculant T2 and T3 plants at 90th and 180th day. Comparatively, T8 recorded less (1.53) DQI among the dual bio-inoculants of T7, T8, T9 and T10, where T10 possessed the maximum (2.53). Overall, the VI and DQI of *S. album* revealed a substantial difference between the single, dual and multiple bio-inoculants treated plants. Thereby the present study supports the suggestions of Kumaran & Surendran (1999), i.e., volume index and quality index can very well be utilised for the selection of growing stock at nursery level. Similar studies were also reported by Chavan et al. (2013), Manavalan (1990) and Kumaran (1991, 1995).

Microbial inoculation effect (MIE)

The MIE is very useful for the assessment of the extent to which introduced, beneficial microbial inoculants compete with native endophytes (potting/polybag mixture, unsterilised soil mixture) to bring about plant growth response (Bhagyaraj 1992). In the present study, MIE was calculated for treatment involving multiple bio-inoculants, where MIE was 97.58% at 90th day and 81.50% at 180th day, respectively, for T12 plants.

CONCLUSION

The findings of the present study showed that plant growth responded better with microbial consortia containing two or more beneficial microorganisms, in association with forestry species under nursery conditions. The growth parameters of *S. album* increased with treatment involving two or more microbial

consortia (AMF + *Azospirillum* + *Azotobacter* + phosphobacteria + potash mobiliser), which was further validated through growth and quality indices. Though, several factors are involved in the growth of bio-inoculants treated seedlings, in the current study, the prime focus was to compare the efficacy of bio-inoculants in the form of single or two, and more. It is therefore very much essential to further understand the unexplored factors governing the growth of bio-inoculant treated semi-parasitic *S. album*, viz., authentic identification of species involved in bio-inoculants (as most of the bio-fertilisers lack species information, especially in Indian market), species compatibility test for multiple bio-inoculants, elucidation of precise dosage of microbial consortia, location specific adaptability (as bio-inoculants are always a foreign species) and mechanism involved in haustorium and bio-inoculant species region-specific suitability.

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