***Original Research Article***

**Effect of Pelletized Biofertilizer Consortia on Rooting, Growth, and Economic Viability in Mulberry (*Morus indica* L.)**

**ABSTRACT**

**Aims:** To evaluate the effect of pelletized biofertilizer formulations on quantitative traits and economic feasibility of mulberry (*Morus indica* L. cv. V1) cuttings under pot culture conditions.

**Study design:** Factorial randomized complete block design (RCBD) with ten treatment combinations.

**Place and Duration of Study:** Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam, India, between February and May 2024.

**Methodology:** The study involved ten treatments consisting of combinations of Orgafol, NAA, and microbial inoculants (*Azospirillum*, Phosphobacteria, and VAM), incorporated into pellet form and applied at planting time. Data were collected at intervals up to 75 days after planting. Observations recorded included rooting percentage, survival rate, root length, shoot length, number of leaves, and leaf area. Economic evaluation of pellet production was also conducted.

**Results:** Significant variations were observed among treatments. Treatment T9 recorded the highest rooting percentage (79.75%) and survival rate (91.60%), followed by T10. Root and shoot lengths were maximum in T9 (38.99 cm and 50.08 cm, respectively), along with the highest number of leaves (13.42) and leaf area (160.82 cm²) at 75 days. The control (T1) consistently showed the lowest performance across all traits. Economic analysis indicated that each pellet costs ₹0.128 to produce and sells at ₹0.60, yielding a net daily profit of ₹516.84 at a production scale of 2000 pellets/day. The Benefit-Cost (B:C) ratio was 1.8, reflecting high profitability and feasibility of the technology.

**Conclusion:** Pelletized biofertilizer formulations combining microbial inoculants and growth regulators significantly improve rooting, growth, and economic returns in mulberry propagation, making them a viable alternative for commercial nursery practices.

*Keywords*:Morus indica*; Biofertilizer pellets;* Azospirillum*; Phosphobacteria; Rooting and survival percentage; Leaf area; Benefit-cost ratio (B:C ratio); Sustainable sericulture*

1. **INTRODUCTION**

Mulberry (*Morus* spp.), a member of the Moraceae family, is a fast-growing deciduous tree widely cultivated for its leaves, which serve as the primary food source for the silkworm, Bombyx mori L., in sericulture. India ranks as the second-largest silk producer globally, where mulberry cultivation forms the cornerstone of sericultural activities, particularly involving the high-yielding Morus indica cv. V1 variety (Gunashekhar *et al.,* 2024; Hawramee *et al.,* 2019).

Despite the biological potential of mulberry to thrive in diverse agro-climatic conditions, its commercial cultivation is challenged by declining soil fertility, rising fertilizer costs, and environmental degradation caused by excessive chemical inputs (Baqual & Das, 2006; Baciu *et al.,* 2023). Studies have shown that conventional reliance on synthetic fertilizers—particularly nitrogen and phosphorus—poses serious risks to soil health, water quality, and microbial biodiversity (Devi & Sakthivel, 2018; Nazar *et al.,* 2019).

To address these concerns, integrated nutrient management strategies involving the application of biofertilizers have gained prominence. Biofertilizers, comprising beneficial microorganisms such as Azospirillum, phosphorus solubilizing bacteria (PSB), and arbuscular mycorrhizal fungi (AMF), are known to promote plant growth by enhancing nutrient uptake, producing phytohormones, and improving rhizosphere interactions (Lucy *et al.,* 2004; Glick, 2012; Pavankumar *et al.,* 2020). These microbial consortia have demonstrated significant positive effects on mulberry physiology, leading to improved shoot length, root development, biomass accumulation, and leaf yield (Baqual, 2013; Moorthi *et al.,* 2016; Diniță *et al.,* 2023). Specifically, vesicular-arbuscular mycorrhizal fungi (VAM) such as Glomus mosseae and G. fasciculatum have been reported to enhance the absorption of both macro and micro-nutrients while reducing dependence on phosphorus fertilizers (Begum *et al.,* 2019; Chakraborty *et al.,* 2015). Similarly, co-inoculation with PSB and nitrogen-fixing bacteria like Azotobacter or Azospirillum has significantly improved the shoot and root traits of mulberry (Baqual *et al.,* 2005; Baqual & Das, 2006; Rao *et al.*, 2007;Vikram, 2010).

Pelletization of biofertilizers, incorporating organic carriers such as Orgafol with microbial inoculants, represents an innovative approach to deliver nutrients more efficiently. Such formulations offer improved microbial viability, controlled nutrient release, ease of application, and enhanced root-soil contact, leading to better plant performance (Pathirana & Yapa, 2020; Pavankumar *et al.,* 2024). Economically, the use of biofertilizers can significantly reduce input costs by partially or completely replacing chemical fertilizers, while maintaining or even enhancing yield (Baqual, 2013; Bharathi *et al.,* 2022). Research indicates that mulberry cultivation with microbial consortia can reduce nitrogen and phosphorus application by up to 50% without compromising leaf productivity, thereby offering a more sustainable and cost-effective model for sericulture (Baqual, 2013; Diniță *et al.,* 2023).

Given this context, the present study was undertaken to develop and assess a pelletized biofertilizer formulation composed of Orgafol and a selected microbial consortium (Azospirillum, PSB, and AMF), and to evaluate its effect on key growth parameters of Morus indica cv. V1 under controlled greenhouse conditions. This study aims to provide insights into the economic viability and agronomic effectiveness of biofertilizer pellets, contributing to sustainable mulberry cultivation and enhanced sericultural productivity.

1. **MATERIALS AND METHODS**

**2.1 Experimental setup**

The present study was conducted through a factorial pot experiment in a naturally lit greenhouse at the Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam, India (11.20°N latitude and 76.56°E longitude; elevation 320 m above mean sea level). The greenhouse temperature ranged from 31°C to 42°C with a relative humidity of 68%. The soil used for the experiment was analyzed before planting, showing a pH of 6.97, EC of 0.38 dS m⁻¹, available nitrogen at 188.06 kg ha⁻¹, phosphorus at 10.27 kg ha⁻¹, and potassium at 215 kg ha⁻¹. V1 variety mulberry (*Morus indica* L.) cuttings served as the planting material.

**2.2 Preparation of biofertilizer pellets**

*Azospirillum* spp., Phosphobacteria, and vesicular arbuscular mycorrhizae (VAM) were employed as microbial biofertilizers. Azospirillum was isolated from surface-sterilized root segments of mulberry and cultured on nitrogen-free bromothymol blue (Nfb) medium at 33°C for 2–8 days. Colonies appeared as subsurface haloes and were further purified and preserved at 4°C (Dobereiner *et al.,* 1976). Phosphobacteria were isolated from undisturbed soil samples of the Forest College and Research Institute using serial dilution and cultured on Pikovskaya’s medium using both pour and streak plate methods, forming clear halo zones indicative of phosphate solubilization (Sundaro Rao & Sinha. 1963). VAM spores were extracted from mulberry rhizosphere soil using the wet sieving and decanting technique (Gerdemann & Nicolson, 1963), and propagated using the funnel method with onion as the host.

The microbial cultures were mass-multiplied in a broth medium composed of yeast extract, beef extract, peptone, and finely ground bone meal, each at 20 g L⁻¹, along with agar at 1 g L⁻¹. For emulsification, 250 mL of boiling water was added to 50 g molten beeswax and 2 g borax, dissolved thoroughly, and added at 100 mL L⁻¹ to the medium. For pellet formation, this enriched broth was combined with lignite (carrier) and guar gum (binder) and processed through a pelletizer to produce uniform pellets embedded with microbial inoculants and the organic growth promoter Orgafol.

The experiment followed a factorial randomized complete block design (RCBD) with four replications. Treatments involved ten types of biofertilizer combinations, each applied at the rate of 5 g pellet per plant (Table 1). The pellets were applied at planting time in individual pots containing the prepared soil.

**Table 1. Summary of the experimental treatments**

|  |  |
| --- | --- |
| **Treatment No.** | **Treatment Compositions** |
|
| **T1 (Control)** | Orgafol |
| **T2** | Orgafol + NAA |
| **T3** | Orgafol + *Azospirillum* |
| **T4** | Orgafol + Phosphobacteria |
| **T5** | Orgafol + VAM |
| **T6** | Orgafol + NAA + *Azospirillum* |
| **T7** | Orgafol + NAA + Phosphobacteria |
| **T8** | Orgafol + NAA + VAM |
| **T9** | Orgafol + NAA + *Azospirillum* + Phosphobacteria |
| **T10** | Orgafol + *Azospirillum* + VAM |

**\*NAA**- Naphthalene acetic acid. **VAM**- Vesicular Arbuscular Mycorrhizae

**2.3 Analytical and statistical methods**

After 75 days of planting, parameters such as rooting percentage and survivability were recorded. Additionally, root length, shoot length, number of leaves, and leaf area were measured at 30 days after planting and subsequently at 15-day intervals up to the 75th day across all treatments. The collected data were subjected to statistical analysis using OPSTAT and SPSS software (version 23) at a 5% level of significance. Mean comparisons were performed using Duncan’s Multiple Range Test (DMRT).

**2.4 Economic metrics used for evaluating treatment effectiveness**

To assess the economic viability of using biofertilizer pellets in mulberry cultivation, several standard financial indicators were used. These included net income, net profit margin, benefit-cost ratio (B:C ratio). These measures provide a comprehensive evaluation of the profitability and return on investment associated with different treatment combinations.

**Net income:** It represents the total profit generated from the treatments after subtracting all incurred costs. It is a key measure of profitability and reflects the effectiveness of biofertilizer application in improving the economic return from mulberry cultivation.

**Net profit margin**: It is a crucial indicator of financial efficiency, reflecting the percentage of revenue that remains as profit after all costs are accounted for. A higher net profit margin indicates better financial health and efficient cost management of the input-output system.

The **Benefit-Cost Ratio (B:C ratio)** was calculated to evaluate the profitability or viability of the biofertilizer pellet application. It is defined as:

Interpretation of B:C ratio values is as follows:

* **BCR < 1**: The investment is not viable and results in a loss.
* **BCR = 1**: The investment breaks even, with no loss or profit.
* **BCR > 1**: The investment is considered profitable and economically viable.

These economic metrics were computed for each treatment to identify the most cost-effective and profitable biofertilizer pellet formulations under the experimental conditions.

1. **RESULTS AND DISCUSSION**

**3.1 Rooting percentage (%) and Survival rate (%)**

The application of different biofertilizer combinations significantly influenced both rooting percentage and survival rate of the cuttings (Table 2). Among the treatments, the highest rooting percentage was recorded in T9 with 79.75%, which was significantly superior to all other treatments, followed closely by T10 with 77.08%. The lowest rooting percentage was observed in the control (T1), with only 53.00%. Similarly, T9 also recorded the highest survival rate (91.60%), followed by T10 (88.00%), whereas the lowest was again observed in T1 (47.78%). These results clearly demonstrate the beneficial effect of combining growth regulators and microbial inoculants in enhancing both rooting and survival. Statistical analysis confirmed that differences among treatments were significant at the 5% level.

Treatments involving NAA in combination with specific microbes, such as T7 and T6, also exhibited enhanced rooting (73.05% and 69.08%) and survival rates (77.80% and 72.05%), suggesting a synergistic effect between the synthetic auxin and microbial inoculants. The effectiveness of auxin hormones, particularly IBA and NAA, in maximizing rooting has been previously documented by Kiruthika *et al.* (2020). Supporting this, Koyuncu & Șenel (2003) observed improved rooting in black mulberry cuttings treated with auxins using the bunch planting method.

Phosphobacteria also played a significant role in root induction. Zenginbal & Demir (2018) reported a 53.63% rooting rate following treatment with Bacillus megaterium (M3 strain), while Kambl *et al.* (1999) noted increased rooting percentage and number of roots per cutting in cinnamon when phosphobacteria were applied at planting. Similar findings by Wange & Ranawade (1997) in grape cuttings also emphasized the positive influence of biofertilizers on rooting. The role of Azospirillum in improving survival was further supported by Bartolini *et al.* (2017), who found increased survival in Vitis vinifera L. plants treated with Azospirillum, likely due to its nitrogen-fixing ability and phytohormonal influence. Subbiah *et al.* (2021) similarly reported the highest survival in Vitis champini cuttings treated with Azospirillum, Phosphobacteria, and CSR-BIO. Additionally, Bharadwaj & Sharma (2006) recorded a 100% survival rate in Morus alba cuttings treated with mycorrhizae and supplemented with phosphorus, highlighting the importance of mycorrhizal associations and adequate phosphorus nutrition in plant establishment.

**Table 2. Effect of different biofertilizer treatments on the rooting percentage (%) and survivality rate (%) of mulberry saplings**

|  |  |  |
| --- | --- | --- |
| **Treatments** | **Rooting percentage (%)** | **Survival rate (%)** |
|
| **T1 (Orgafol) (Control)** | 53.00±0.041 | 47.78±0.024 |
| **T2 (Orgafol + NAA)** | 57.03±0.075 | 56.03±0.047 |
| **T3 (Orgafol + *Azospirillum*)** | 62.00±0.071 | 68.05±0.063 |
| **T4 (Orgafol + Phosphobacteria)** | 69.63±0.047 | 74.58±0.018 |
| **T5 (Orgafol + VAM)** | 58.90±0.040 | 65.00±0.041 |
| **T6 (Orgafol + NAA + *Azospirillum*)** | 69.08±0.102 | 72.05±0.063 |
| **T7 (Orgafol + NAA + Phosphobacteria)** | 73.05±0.066 | 77.80±0.040 |
| **T8 (Orgafol + NAA + VAM)** | 65.10±0.082 | 70.10±0.092 |
| **T9 (Orgafol + NAA + *Azospirillum* + Phosphobacteria)** | 79.75±0.029 | 91.60±0.041 |
| **T10 (Orgafol + *Azospirillum* + VAM)** | 77.08±0.064 | 88.00±0.074 |
| **Mean** | **66.40** | **71.10** |
| **CD @ 0.05%** | **0.184** | **0.131** |

**3.2 Shoot length (cm)**

The effect of different biofertilizer treatments on shoot length at 30, 45, 60, and 75 days after planting is summarized in Table 3. All biofertilizer combinations significantly outperformed the control (T1), with the highest shoot length recorded in T9, reaching 50.08 cm at 75 days. This was followed by T10 (49.55 cm) and T7 (49.28 cm), while the control (T1) recorded the lowest shoot length (48.58 cm). Statistical analysis confirmed that differences among treatments were significant at the 5% level. The superior performance of T9 may be attributed to the synergistic action of microbial inoculants and growth regulators, particularly *Azospirillum*, which enhances nitrogen fixation and phytohormone production, thereby accelerating vegetative growth (Avinash *et al.*, 2019). These findings align with Bartolini *et al.* (2017), who reported enhanced shoot growth in plants inoculated with Azospirillum brasilense Sp245.

Several studies have similarly documented the growth-promoting effects of bacterial inoculants on shoot development (Afzal & Bano, 2008; Farzana *et al.,* 2009; Gholami *et al.*, 2009; Sabir *et al.*, 2012). Subbiah *et al.* (2021) also observed increased shoot length in mulberry cuttings treated with a combination of *Azospirillum*, Phosphobacteria, and CSR-BIO. Pavankumar *et al.* (2020) demonstrated that integrating organic manures with Azospirillum significantly improves shoot growth.The present results further reinforce the importance of combining organic amendments with biofertilizers, which not only enhance shoot elongation but also contribute to improved nutrient use efficiency, soil fertility, and mulberry productivity (Senapati *et al.,* 2005; Moradi *et al*., 2014). Comparable results were reported by Pavankumar *et al*. (2020), Singh *et al.* (2012), and Dhanalakshmi *et al.* (2014).

**Table 3. Effect of different biofertilizer treatments on the shoot length (cm) of the mulberry saplings**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatments** | **Shoot length (cm)** | | | |
| **30th day** | **45th day** | **60th day** | **75th day** |
| **T1 (Orgafol) (Control)** | 12.85±0.104 | 20.68±0.210 | 31.65±0.155 | 48.58±0.062 |
| **T2 (Orgafol + NAA)** | 12.85±0.104 | 20.93±0.085 | 31.68±0.165 | 48.70±0.092 |
| **T3 (Orgafol + *Azospirillum*)** | 13.18±0.149 | 21.13±0.131 | 32.03±0.103 | 49.00±0.071 |
| **T4 (Orgafol + Phosphobacteria)** | 13.20±0.108 | 21.15±0.064 | 32.20±0.071 | 49.25±0.119 |
| **T5 (Orgafol + VAM)** | 13.33±0.131 | 21.43±0.075 | 32.30±0.041 | 49.28±0.062 |
| **T6 (Orgafol + NAA + *Azospirillum*)** | 13.28±0.075 | 21.35±0.065 | 32.30±0.108 | 49.33±0.118 |
| **T7 (Orgafol + NAA + Phosphobacteria)** | 13.38±0.132 | 21.43±0.165 | 32.38±0.125 | 49.28±0.111 |
| **T8 (Orgafol + NAA + VAM)** | 13.23±0.103 | 21.25±0.119 | 32.05±0.323 | 49.13±0.085 |
| **T9 (Orgafol + NAA + *Azospirillum* + Phosphobacteria)** | 14.00±0.091 | 22.28±0.149 | 33.18±0.111 | 50.08±0.085 |
| **T10 (Orgafol + *Azospirillum* + VAM)** | 13.60±0.108 | 21.58±0.075 | 32.58±0.048 | 49.55±0.065 |
| **Mean** | **13.29** | **21.32** | **32.23** | **49.22** |
| **CD @ 0.05%** | **0.274** | **0.304** | **0.427** | **0.264** |

**3.3 Root length (cm)**

The effect of various biofertilizer treatments on root length at different growth intervals (30th, 45th, 60th, and 75th day) is summarized in Table 4. Root length was significantly influenced by the treatments, with all biofertilizer combinations performing better than the control (T1). Among the treatments, **T9** recorded the highest root length throughout the observation period, reaching **38.99 cm** on the 75th day. This was followed by **T10** and **T7**, with root lengths of **38.13 cm** and **37.78 cm**, respectively. The control treatment (T1) consistently recorded the lowest root length across all time points, reaching **33.76 cm** at the end of the experiment. Statistical analysis confirmed that differences among treatments were significant at the 5% level. The enhanced root growth observed in T9 can be attributed to improved soil porosity and the synergistic effects of microbial inoculants, which likely facilitated better root penetration and elongation. Wani *et al.* (2017) reported that longer root systems are often associated with increased organic matter and micronutrient availability, which enhance meristematic activity. Similarly, Gangwar & Thangavelu (1992) demonstrated increased root length in treatments involving Azotobacter with FYM and Azospirillum with gum, supporting the present findings.

Auxin application, particularly NAA, played a crucial role in root development. Singh *et al.* (2014) observed significantly higher root length in Morus alba L. treated with auxins, while Zhang *et al.* (2017) reported a 1.2- to 3-fold increase in root length in cuttings treated with 300 mg/L NAA compared to the control. Kiruthika *et al.* (2020) also demonstrated maximum root elongation with auxin treatments, attributing this to enhanced hydrolysis and translocation of carbohydrates to the base of the cuttings, thereby promoting cell division and elongation. These results are further corroborated by Singh (2018) and Vijay *et al.* (2023), who noted improvements in root length and rooting success in mulberry cuttings treated with auxins. In addition, the role of plant growth-promoting rhizobacteria (PGPR) was evident in the present study. Zenginbal & Demir (2018) found that the highest root length in cuttings (6.44 cm) was obtained following treatment with Bacillus megaterium (M3 strain), a phosphobacteria species. This aligns with the present observations, where phosphobacteria in combination treatments significantly enhanced root growth. Further evidence from Ercisli *et al.* (2004), Erturk *et al.* (2008), and Erturk *et al.* (2010) demonstrated the positive effects of PGPR on root development in rosehip, tea, and kiwifruit, respectively.

**Table 4. Effect of different biofertilizer treatments on the root length (cm) of the mulberry saplings**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatments** | **Root length (cm)** | | | |
| **30th day** | **45th day** | **60th day** | **75th day** |
| **T1 (Orgafol) (Control)** | 3.78±0.004 | 6.76±0.005 | 18.74±0.007 | 33.76±0.006 |
| **T2 (Orgafol + NAA)** | 4.12±0.050 | 7.43±0.004 | 19.86±0.004 | 35.32±0.006 |
| **T3 (Orgafol + *Azospirillum*)** | 3.97±0.005 | 7.97±0.005 | 20.21±0.010 | 34.97±0.006 |
| **T4 (Orgafol + Phosphobacteria)** | 4.87±0.004 | 8.54±0.006 | 21.79±0.003 | 36.24±0.004 |
| **T5 (Orgafol + VAM)** | 3.92±0.004 | 7.76±0.004 | 20.12±0.003 | 34.91±0.004 |
| **T6 (Orgafol + NAA + *Azospirillum*)** | 4.34±0.005 | 8.12±0.008 | 21.32±0.008 | 35.96±0.006 |
| **T7 (Orgafol + NAA + Phosphobacteria)** | 5.78±0.004 | 8.97±0.004 | 21.99±0.004 | 37.78±0.009 |
| **T8 (Orgafol + NAA + VAM)** | 4.78±0.007 | 7.89±0.004 | 20.34±0.004 | 36.12±0.006 |
| **T9 (Orgafol + NAA + *Azospirillum* + Phosphobacteria)** | 5.89±0.007 | 9.13±0.007 | 22.54±0.006 | 38.99±0.004 |
| **T10 (Orgafol + *Azospirillum* + VAM)** | 5.19±0.004 | 8.26±0.004 | 21.46±0.005 | 38.13±0.004 |
| **Mean** | **4.66** | **8.04** | **20.84** | **36.22** |
| **CD @ 0.05%** | **0.047** | **0.017** | **0.013** | **0.008** |

**3.4 Number of leaves**

The number of leaves per plant increased progressively from the 30th to the 75th day across all treatments, with significant differences observed between treatments (Table 5). The maximum leaf count at 75 days was recorded in T9 with 17.20 leaves, followed by T6 (16.85) and T10 (16.50), while the control (T1) recorded the lowest value (13.90). Statistical analysis confirmed that differences among treatments were significant at the 5% level.

These results indicate that the combined application of biofertilizers and plant growth regulators significantly enhances foliar development. Treatments involving Azospirillum and Phosphobacteria either alone or in combination with NAA showed marked improvement in leaf number, supporting the observations of Bartolini *et al.* (2017), who reported that Azospirillum brasilense Sp245 treatments enhanced vegetative traits in moderately vigorous rootstocks. Azospirillum likely promotes leaf formation through its production of phytohormones such as IAA, gibberellin-like substances, and vitamins including B12, thiamine, and riboflavin (Avinash *et al.,* 2019). These bioactive compounds improve overall plant vigor, contributing to increased photosynthetic surface area. Consistent findings were reported by Mohanty *et al.* (2002) in tuberose and Singh & Singh (2005) in rose.

Yadav & Kumar (1993) also recorded enhanced leaf yield in mulberry when Azospirillum brasilense (strain SL-33) was combined with nitrogen fertilizer, suggesting a synergistic effect on nitrogen uptake. Similarly, Vijay *et al.* (2023) observed that auxin treatments improved root systems, which in turn supported increased leaf production due to better nutrient and water absorption. This root-to-shoot stimulation mechanism is well-documented (Siddiqui & Hussain, 2007). Supporting this, Sourati *et al.* (2022) reported a significant rise in leaf number in Morus alba cuttings treated with auxin and zinc sulfate.

**Table 5. Effect of different biofertilizer treatments on number of leaves of the mulberry saplings**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatments** | **Number of leaves** | | | |
| **30th day** | **45th day** | **60th day** | **75th day** |
| **T1 (Orgafol) (Control)** | 2.40±0.041 | 6.13±0.085 | 9.45±0.029 | 13.90±0.041 |
| **T2 (Orgafol + NAA)** | 3.20±0.091 | 6.30±0.041 | 9.90±0.041 | 14.30±0.071 |
| **T3 (Orgafol + *Azospirillum*)** | 3.70±0.041 | 6.90±0.041 | 10.50±0.041 | 15.80±0.041 |
| **T4 (Orgafol + Phosphobacteria)** | 3.40±0.041 | 6.50±0.091 | 10.30±0.041 | 14.80±0.071 |
| **T5 (Orgafol + VAM)** | 2.90±0.071 | 5.90±0.041 | 9.70±0.041 | 14.10±0.041 |
| **T6 (Orgafol + NAA + *Azospirillum*)** | 4.10±0.071 | 7.38±0.048 | 11.50±0.041 | 16.85±0.065 |
| **T7 (Orgafol + NAA + Phosphobacteria)** | 3.80±0.041 | 7.20±0.071 | 11.35±0.029 | 16.30±0.041 |
| **T8 (Orgafol + NAA + VAM)** | 3.10±0.041 | 6.20±0.041 | 10.20±0.041 | 14.50±0.071 |
| **T9 (Orgafol + NAA + *Azospirillum* + Phosphobacteria)** | 4.38±0.085 | 7.50±0.041 | 11.83±0.048 | 17.20±0.041 |
| **T10 (Orgafol + *Azospirillum* + VAM)** | 4.25±0.065 | 7.40±0.041 | 11.50±0.041 | 16.50±0.041 |
| **Mean** | **3.52** | **6.74** | **10.62** | **15.43** |
| **CD @ 0.05%** | **0.169** | **0.175** | **0.110** | **0.148** |

**3.5 Leaf area (cm2)**

Leaf area showed a steady increase across all treatments from the 30th to the 75th day, with significant variations among treatments (Table 6). The largest leaf area at 75 days was recorded in T9 with 160.82 cm², followed by T7 and T10, which recorded 147.66 cm² and 154.69 cm² respectively. The lowest value was observed in the control (T1), with 124.54 cm². Statistical analysis confirmed that differences among treatments were significant at the 5% level. This variation in leaf area can be attributed to the synergistic effect of plant growth regulators (PGRs) and biofertilizers, which stimulate cell division and elongation in meristematic tissues. These results align with Asghar *et al.* (2002), who reported that increased leaf area is associated with enhanced photosynthetic efficiency and better translocation of photosynthates. The involvement of PGPRs like Azospirillum and Phosphobacteria in improving vegetative growth through phytohormone production also supports the findings of Prud'homme *et al.* (1992).

Moreover, El-Yazied & Mady (2012) observed that bio-extracts such as yeast enhance enzyme activity and pigment concentration, promoting vigorous vegetative development, a mechanism likely paralleled in the current study through microbial and hormonal stimulation. Similar trends of increased leaf expansion due to biostimulant application were also documented by Sajid *et al.* (2009) in lilies, Rawgol *et al.* (2011) in mulberry, and Azizi & Mahmoudabadi (2013) in sesame.

**Table 6. Effect of different biofertilizer treatments on leaf area (cm2) of the mulberry saplings**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatments** | **Leaf area (cm2)** | | | |
| **30th day** | **45th day** | **60th day** | **75th day** |
| **T1 (Orgafol) (Control)** | 22.46±0.024 | 51.64±0.015 | 85.98±0.019 | 124.54±0.003 |
| **T2 (Orgafol + NAA)** | 26.32±0.004 | 59.67±0.013 | 92.45±0.012 | 131.33±0.075 |
| **T3 (Orgafol + *Azospirillum*)** | 29.69±0.004 | 67.88±0.010 | 100.32±0.004 | 147.32±0.008 |
| **T4 (Orgafol + Phosphobacteria)** | 24.87±0.003 | 62.79±0.004 | 95.67±0.012 | 139.55±0.033 |
| **T5 (Orgafol + VAM)** | 22.89±0.004 | 54.89±0.007 | 87.98±0.004 | 128.67±0.004 |
| **T6 (Orgafol + NAA + *Azospirillum*)** | 31.57±0.007 | 75.44±0.008 | 109.97±0.004 | 155.98±0.008 |
| **T7 (Orgafol + NAA + Phosphobacteria)** | 28.96±0.008 | 65.98±0.004 | 98.77±0.013 | 147.66±0.007 |
| **T8 (Orgafol + NAA + VAM)** | 27.99±0.004 | 63.79±0.004 | 96.58±0.019 | 140.98±0.003 |
| **T9 (Orgafol + NAA + *Azospirillum* + Phosphobacteria)** | 34.22±0.004 | 81.55±0.008 | 114.98±0.012 | 160.82±0.007 |
| **T10 (Orgafol + *Azospirillum* + VAM)** | 29.82±0.009 | 74.79±0.004 | 106.79±0.024 | 154.69±0.003 |
| **Mean** | **27.88** | **65.83** | **98.95** | **143.15** |
| **CD @ 0.05%** | **0.030** | **0.072** | **0.093** | **0.112** |

**3.6 Benefit-Cost Evaluation of Pelletized Biofertilizers in Sustainable Mulberry Production**

The economic analysis revealed that the application of biofertilizer pellets was economically viable, as indicated by the calculated **B:C ratio of 1.76**. This suggests that for every rupee invested in the preparation and application of the biofertilizer pellets, there was a return of 1.76 rupees, signifying a **profitable outcome**. The cost of preparing one pellet was calculated to be ₹0.128, while the selling price was ₹0.60 per pellet, yielding a favorable profit margin. Labor charges, carrier materials, and organic growth promoter ingredients were factored into the total cost computation. Notably, **2000 pellets could be prepared per day**, indicating the feasibility of large-scale pellet production with relatively low labor input (Table 7).

The favorable B:C ratio underscores the **cost-effectiveness and economic sustainability** of using pelletized biofertilizer formulations, especially when integrated with microbial consortia such as Azospirillum, Phosphobacteria, and VAM. The findings align with earlier studies by **Baqual (2013)**, who reported that the use of microbial inoculants such as Azotobacter, phosphate-solubilizing bacteria (PSB), and vesicular-arbuscular mycorrhizae (VAM) could reduce nitrogen and phosphorus fertilizer requirements by **25% to 50%,** without compromising yield. This partial replacement of synthetic fertilizers significantly reduces input costs, thereby enhancing net returns. Moreover, **Brindha *et al.* (2022)** emphasized that integrated nutrient management using biofertilizers improves nutrient use efficiency, sustains soil fertility, and supports economically viable mulberry production.

The high B:C ratio observed in this study supports the broader concept of **sustainable intensification** in sericulture. By improving root-soil interactions and promoting plant growth through microbial activity, biofertilizer pellets enhance biomass production while minimizing chemical dependency. This not only reduces environmental impact but also improves farmers’ income through reduced input costs and improved productivity.

**Table 7. Cost Structure and Benefit-Cost Analysis of Biofertilizer Pellet Production**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Sl. No.** | **Input** | **Commercial price (Rs/kg)** | **Unit price (Rs/g)** | **Required quantity (g)** | **Unit price x Quantity (Rs)** |
| **A** | **Preparation of the Organic Growth Promoter in Liquid form (1 litre):** | | | | |
| **1** | **Yeast Extract** | 290.00 | 0.29 | 20 | 5.8 |
| **2** | **Beef Extract** | 300.00 | 0.30 | 20 | 6.0 |
| **3** | **Peptone** | 160.00 | 0.16 | 20 | 3.2 |
| **4** | **Bone meal powder** | 15.00 | 0.015 | 20 | 0.3 |
| **5** | **Agar** | 1600.00 | 1.6 | 1 | 1.6 |
| **6** | **Citric acid** | 40.00 | 0.04 | 30 | 1.2 |
| **7** | **Bee wax** | 140.00 | 0.14 | 50 | 7.0 |
| **8** | **Borax** | 36.00 | 0.036 | 2 | 0.072 |
| **Total** | | | | | **25.172** |
| **B.** | **Preparation of the Carrier material** | | | | |
| **1** | **Lignite** | 17.00 | 0.017 | 3000 | 51.00 |
| **2** | **Guar Gum** | 70.00 | 0.07 | 10 | 0.70 |
| **3** | **Cellulose** | 9.00 | 0.009 | 12 | 0.108 |
| **Total** | | | | | **51.808** |
| **Total expenses in preparing the raw material for pellets (A+B)** | | | | | **76.98** |
| **Cost of 1 pellet (600 pellets/ l)** | | | | | 0.128 |
| **C.** | **Cost of Pelletization** | | | | |
| **1.** | **Total No. of pellets/ day** |  |  | 2000 (@250 pellets/ hr) | 256.00 |
| **2.** | **Labor cost** |  |  |  | 425.00 / 8 hrs |
| **Total Expenses in pelletization** | | | | | **681.00** |
| **Selling price (Rs. 0.60/pellet)** | | | | | **1200.00** |
| **B:C ratio** | | | | | **1: 1.76** |
| **Profit margin per pellet** | | | | | **0.76** |

1. **CONCLUSION**

The present study clearly demonstrates that the use of pelletized biofertilizer formulations enriched with microbial consortia—Azospirillum, Phosphobacteria, and VAM—in combination with Orgafol and the plant growth regulator NAA, significantly improves the rooting success, survival rate, and vegetative growth of mulberry (Morus indica L. cv. V1) cuttings. Among the tested treatments, T9 consistently outperformed all others by recording the highest rooting percentage (79.75%), survival rate (91.60%), shoot length (50.08 cm), root length (38.99 cm), number of leaves (13.42), and leaf area (160.82 cm²) at 75 days after planting. In addition to its agronomic benefits, the pellet-based application method proved to be economically viable. With a production cost of ₹0.128 per pellet and a market value of ₹0.60, the technology generated a net daily profit of ₹516.84 per laborer and a favorable Benefit-Cost (B:C) ratio of 1.8. The low cost, ease of application, and improved microbial viability make the pellet form especially suitable for nursery and field-level adoption. These findings strongly support the integration of microbial biofertilizers into mulberry cultivation as a sustainable alternative to chemical fertilizers. The use of such eco-friendly inputs not only enhances early plant establishment and growth performance but also reduces input costs, contributes to soil health, and aligns with environmentally sustainable sericulture practices. Hence, biofertilizer pellets hold significant promise for improving both the productivity and profitability of mulberry-based farming systems.

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