Effect of bio-inoculants on growth and yield of betel vine (*Piper betle*)

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(Manuscript Received: 08-05-2020, Revised: 10-08-2020, Accepted: 20-09-2020)

Abstract

Betel vine (*Piper betel*) cultivation and the consumption of betel leaves is a very traditional and widespread practice in India and many South-East Asian countries. The marginal and small farmers generally cultivate betel vine in their small holdings which provide them with a means of alternative cash earning to meet their day to day livelihood. Chewing betel leaves is an old habit of the people residing in subtropical countries. It is used in several traditional remedies for the treatment of stomach ailments, infections and as a general tonic. As betel leaf is directly chewed, there is a need to improve the leaf yield and to optimize the usage of manures for the leaf production. A study was taken up to assess the efficacy of some of the bio-inoculants in combination with inorganic fertilizers in betel vine cv. SGM 1 under an open system of deep trench method. Among the various treatments, *Azospirillum* @ 5 kg coupled with 100 kg each of P and K ha⁻¹ had recorded the highest vine length (195.6 cm), more number of laterals per vine (22.3) and highest leaf yield ha⁻¹ (44.7 lakh).

Keywords: Bio-inoculants, leaf yield, nitrogenous fertilizer, *Piper betle*, shelf life

Introduction

Betel vine is an important commercial crop in India, Bangladesh and Sri Lanka. It is also cultivated in almost all the South-East Asian countries on a limited scale. The betel vine (*Piper betle* L.) is a perennial climber, cultivated for its leaf which is used as a masticatory. Pan chewing is an old habit, prevalent amongst all classes of people in South-East Asian countries. It is an antiseptic, carminative, curing cough and cold, pains and sores in throat and chest. These leaves are also in great demand in several other countries of the world. It is cultivated intensively for its young leaves, which are consumed by about 15-20 million people in the country. Some evidence suggests that betel leaves have immune-boosting properties as well as anti-cancerous properties.

Betel vine is grown as an important cash crop in southern parts of India, mainly in the states of Andhra Pradesh, Karnataka, Kerala and Tamil Nadu. It is also cultivated in Assam, Bihar, Madhya Pradesh, Maharashtra, Orissa, Meghalaya, Tripura, Uttar Pradesh and West Bengal. This crop is cultivated in about 55,000 hectares with an annual production worth about ₹ 9000 million (Kaleeswari and Sridhar, 2013). Betel leaves worth ₹ 30-40 million are exported to other countries. Thus it is a most promising commercial leafy crop capable of attracting a substantial amount of foreign exchange to the country. In India, Tamil Nadu with an area of about 5000 hectares, ranks fourth in the total area under cultivation of betel vine. Classified as a plantation crop, betel vine occupies 2 per cent of net cultivated area in the state. It offers perennial employment and income to the small and marginal farming community because of its capital and labour intensive characteristics. Betel vine cultivation is highly intensive and particularly suited to small holding, maybe 5 to 10 decimal land (1/100 of an acre). In the total production of betel leaves in India,
Tamil Nadu’s share is about to 46.5 per cent. Revenue generated will easily exceed if agronomic practices are scientifically explored (Kaleeswari and Sridhar, 2013).

Applications of nutrients play a pivotal role in the growth, production and quality of the crops. Continuous and heavy application of chemical inputs resulted in a major shift in soil microbial load, nutrient imbalance, fast depletion of soil fertility and continuous deterioration of soil physical and chemical properties and accumulation of toxic contaminants (Srinivasan and Thomas, 2003). Now, there is a growing realization that the adoption of ecological and sustainable farming practices can only reverse the declining trend in global productivity and environmental protection. Research on the use of organic inputs has shown that there is scope to improve the yield potential of many horticultural and field crops besides improving the soil physical and chemical properties. This will ensure a continuous supply of nutrients to the crop, which ultimately improves the yield and quality of the crops.

Plant nutrition is one of the important components in betel vine cultivation. Among the various essential nutrients, nitrogen plays a pivotal role in improving the productivity and quality of betel vine. Being a perennial crop, the production of economic part viz., the leaf requires ‘N’ throughout the crop growth (Arulmozhiyan, 1996). The fertilizer use efficiency of inorganic nitrogenous fertilizer urea is only 30-40 per cent, which can be improved by combining with other organic sources, which has an impact on productivity. Use of bio-inoculants as the biological nitrogen-fixing source can act as an organic source of nitrogen, for improvement in yield. Hence, the present study was taken up to assess the efficacy of some of the bio-inoculants in combination with inorganic fertilizers in betel vine cv. SGM 1 under an open system of deep trench method.

Materials and methods

A field experiment was undertaken during 2016-17 and 2017-18 in farmer’s field at Banglamedu, Periyakulam, Theni District. Betel vine var. SGM 1 cuttings were treated with bio-inoculants (Azotobacter, Azospirillum and Phosphobacterium each @ 5 kg ha⁻¹), in combination with farmyard manure (FYM @ 100 kg ha⁻¹) before planting. The cuttings were trained on Sessania grandiflora which provide support and shade to betel vine. The soil status of the experimental field was clay loam with low available soil N of 158.7 kg ha⁻¹ and medium status of P and K (21.2 kg and 372.00 kg ha⁻¹, respectively). The soil pH was 8.3, with EC at 0.43 dSm⁻¹. The experiment was carried out in Randomized Block Design with seven treatments replicated three times. Each plot consisted of 50 vines measuring 5 m² in a paired row system. The treatments were as follows:

| Treatment | Description |
|-----------|-------------|
| T₁ | Azotobacter 5 kg + 100 kg P + 50 kg K ha⁻¹ |
| T₂ | 150 kg N + Phosphobacteria 5 kg + 50 kg K ha⁻¹ |
| T₃ | Azotobacter 5 kg + Phosphobacteria 5 kg + 50 kg K ha⁻¹ |
| T₄ | 200:100:100 kg NPK ha⁻¹ as inorganic form |
| T₅ | 150:100:50 kg NPK ha⁻¹ in organic form |
| T₆ | Azospirillum 5 kg + 100 kg each of P and K ha⁻¹ |
| T₇ | Recommended NPK of 150:100:50 kg ha⁻¹ |

Fertilizers and bio-inoculants were applied in three split doses at 45 days interval from 60th day after planting. Azotobacter is an aerobic free-living bacteria and fixes atmospheric nitrogen (Rangaswami and Bagyaraj, 2005). The most abundant species is Azotobacter chroococcum and was taken for the experiment. The inoculum was mixed with FYM and applied @ 5 kg ha⁻¹ at the base of the plant. A full dose of P₂O₅ and K₂O were applied 15 days after Azotobacter application. Phosphobacter spp. helps in solubilizing phosphorus that is immobilized and fixed in the soil to utilizable form and helps in easy uptake (Krishnamurthy and Rema, 2004). The inoculum of this microorganism was mixed with FYM and applied @ 5 kg ha⁻¹ at the base (rhizosphere) of the plant. For the treatment T₅, organic manures applied were in the form of FYM for N, rock phosphate for P and press mud for K. The fertilizer dose of 200:100:100 kg NPK ha⁻¹ (T₄) is followed in North India, while 150:100:50 kg NPK ha⁻¹ (T₇) is followed in South India, applied as urea, single super phosphate (SSP) and muriate of potash. Splash irrigation was given daily for a week and subsequently in alternate days. Mud plastering of bed was done at monthly intervals. Lowering or coiling of the vine was done once in nine months. Harvesting was done during the month.
Table 1. Effect of bio-inoculants on growth of betel vine

| Treatments | Vine elongation (cm month\(^{-1}\)) | Vine length (cm) | Lateral vine\(^{-1}\) (Nos.) | Internodal length (cm) | Leaf petiole length (cm) | Leaf length (cm) | Leaf breadth (cm) |
|------------|-------------------------------------|-----------------|-------------------------------|------------------------|------------------------|-----------------|------------------|
| T\(_1\)    | 8.0                                 | 96.0            | 12.60                         | 5.60                   | 6.4                    | 12.52           | 9.22             |
| T\(_2\)    | 9.4                                 | 112.8           | 14.30                         | 5.90                   | 6.7                    | 14.43           | 11.78            |
| T\(_3\)    | 10.9                                | 130.8           | 15.50                         | 6.15                   | 7.5                    | 14.89           | 12.47            |
| T\(_4\)    | 6.9                                 | 82.8            | 12.30                         | 5.80                   | 7.4                    | 15.01           | 12.68            |
| T\(_5\)    | 12.5                                | 150.0           | 17.58                         | 6.32                   | 7.7                    | 16.34           | 12.92            |
| T\(_6\)    | 16.3                                | 195.6           | 23.30                         | 6.60                   | 7.8                    | 16.93           | 13.21            |
| T\(_7\)    | 8.3                                 | 99.6            | 14.60                         | 5.20                   | 7.1                    | 14.25           | 12.18            |
| Mean       | 10.33                               | 123.9           | 15.74                         | 5.94                   | 7.2                    | 14.91           | 12.07            |
| SE         | 0.57                                | 1.32            | 0.67                          | 0.05                   | 0.02                   | 0.41            | 0.16             |
| CD(P=0.05) | 1.77                                | 2.77            | 2.07                          | 1.50                   | 0.06                   | 0.87            | 0.35             |

Results and discussion

The important growth parameters like vine length, number of laterals and internodal length influenced the growth and productivity of the crop (Table 1). These growth parameters were differentially influenced by the bio-inoculants, which contributed to the growth, yield and quality of the crop. The highest vine length (195.6 cm) was recorded by the application of *Azospirillum* @ 5 kg coupled with 100 kg each P and K ha\(^{-1}\) (T\(_6\)), and the lowest vine length (82.0 cm) was recorded by the application of 200:100:200 kg NPK ha\(^{-1}\) as an inorganic form. The maximum elongation of vine per month (16.3 cm) was also noticed in the treatment applied with *Azospirillum* @ 5 kg coupled with 100 kg each P and K ha\(^{-1}\) (T\(_6\)). As per reports of Selvaraj *et al.* (2003), the growth of thyme was vigorous by the combined application of organic manures and bio-fertilizers. Among the different treatments, the number of laterals per vine ranged from 23.3 in T\(_6\) (*Azospirillum* 5 kg + 100 kg each of P and K nuts ha\(^{-1}\)) to 12.3 in T\(_4\) (200:100:200 kg NPK ha\(^{-1}\) as an inorganic form). The application of *Azospirillum* @ 5 kg coupled with 100 kg each P and K ha\(^{-1}\) (T\(_6\)) registered significantly higher leaf length of 16.93 cm, followed by T\(_5\) which recorded 16.34 cm. The treatment T\(_6\) recorded the highest leaf breath (13.21 cm), which was on par with T\(_4\) (12.92 cm). Leaf length and leaf breadth are reliable yield determinants in many crops. Higher leaf length will be helpful for better exposure of the leaf to the sun and thus, a higher rate of photosynthesis (Joseph *et al.*, 1981). Among different treatments, the treatment T\(_6\) recorded the highest internodal length (6.60 cm) and petiole length (7.80 cm).

Growth parameters like vine length, number of laterals, internodal length, leaf length and leaf breadth were improved due to the application of bio-fertilizers in combination with chemical fertilizers in this experiment. In general, biofertilizers are reported to supply or build up the soil N up to 100-300 kg ha\(^{-1}\) year\(^{-1}\) in case of legume crops and up to 30-50 kg ha\(^{-1}\) year\(^{-1}\) in non-legume crops. *Sesbania grandiflora* is a legume crop used as support tree for betel vine. This support crop also increased the N content in the soil. Enhanced nutrient uptake by plants by inoculation of *Azospirillum* and P-solubilizing bacteria (PSB) probably caused increased cell elongation and cell multiplication, and hence, the increased plant height (Preethi *et al.*, 1999). This can also be attributed to the production of plant growth-promoting substances by the inoculated microorganisms in the root zone. Significant increase in plant height and spread due to combined application of *Azospirillum*, PSB and inorganic fertilizers have been reported earlier in crossandra (Narashimha Raju and Haripriya, 2001) and jasmine (Manonmani, 1992).
The leaf is the economic part of betel vine. Significant differences were recorded among the treatments in respect of the number of leaves per vine and estimated leaf yield ha\(^{-1}\) (Table 2). Treatment T\(_6\) recorded the highest number of leaves per vine (63.86) while the treatment T\(_1\) (Azotobacter 5 kg + 100 kg P + 50 kg K ha\(^{-1}\)) recorded the lowest number of leaves per vine (45.43). The estimated leaves ha\(^{-1}\) were highest in treatment T\(_6\) (44.70 lakhs) and on par with T\(_5\) (38.70 lakhs). The shelf life was not influenced by any of the above treatments (Table 2). The leaves were said to be less pungent in the vines treated with bio-inoculants while it was highly pungent in the vines that received inorganic fertilizers. Increased yield attributes noticed in the application of Azospirillum 5 kg + 100 kg each of P and K ha\(^{-1}\) may due to bio-fertilizer addition reduced the loss of nitrogen by improving the structure of the soil by more aggregation, increasing water holding capacity and air permeability. Similar trends were found in Coleus (Ravikumar et al., 2013); turmeric (Roy and Hore, 2011) and Anthurium (Padmadevi, 2004).

The increase in yield due to the application of Azospirillum, which is a microaerobic symbiotic bacteria, was due to fixing atmospheric nitrogen in host plant viz., betel vine (Arulmozhiyan, 1996). Azospirillum fixes nitrogen through the enzyme nitrogenase and also produces growth hormones viz., IAA, GA and cytokinin (Hubbel et al., 1979; Govindan and Nair, 1986), that can cause changes in root length (Pandey and Kumar, 1980), branching of root hairs (Jain and Partquin, 1984), production of more lateral roots (Barbieri et al., 1986), enhanced rate of cell division and differentiation in meristematic tissues (Fallick et al., 1989) and nitrogen uptake (Pandey and Kumar, 1980). It was also reported that application of Azospirillum to horticultural crop plants enhanced yield as a substitute for nitrogenous fertilizers (Jeeva, 1987; Thangaselvabai, 1989). Marimuthu et al. (1986) reported betel vine cuttings inoculated with Azospirillum recorded more marketable leaves and leaf weight. Thus, it can be concluded that Azospirillum at 5 kg coupled with 100 kg each of P\(_2\)O\(_5\) and K\(_2\)O ha\(^{-1}\) had recorded the highest growth and yield attributes in betel vine var. SGM 1 which is a bushy type under an open system of deep trench system under Tamil Nadu conditions.

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### Table 2. Effect of bio-inoculants on yield and quality of betel vine

| Treatments | No. of leaves vine\(^{-1}\) year\(^{-1}\) | Estimated leaf yield ha\(^{-1}\) (lakh) | Hundred leaf weight (g) | Shelf life (days) | Organoleptic test* |
|------------|---------------------------------|---------------------------------|------------------|----------------|------------------|
| T\(_1\)    | 45.43                           | 31.8                            | 244.9            | 12.9           | 3                |
| T\(_2\)    | 47.86                           | 33.5                            | 249.7            | 13.3           | 3                |
| T\(_3\)    | 49.86                           | 34.9                            | 255.4            | 13.4           | 3                |
| T\(_4\)    | 48.57                           | 34.0                            | 238.2            | 12.1           | 1                |
| T\(_5\)    | 55.29                           | 38.7                            | 267.0            | 13.3           | 1                |
| T\(_6\)    | 63.86                           | 44.7                            | 280.5            | 15.6           | 3                |
| T\(_7\)    | 48.14                           | 33.7                            | 259.6            | 13.3           | 1                |
| Mean       | 51.29                           | 35.9                            | 256.47           | 13.41          | 2                |
| SE         | 1.61                            | 1.37                            | 3.536            | 0.45           | -                |
| CD(P=0.05) | 3.27                            | 4.25                            | 11.61            | NS             | -                |

* 1: Highly pungent; 2: Pungent; 3: Less pungent
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