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Full Length Research Paper

Studies on effect of organic amendments and bio-stimulants on morphology, yield and quality of *Gymnema sylvestre* R.Br

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The experiment on effect of organic amendments and growth promoters on morphology and yield of *Gymnema sylvestre* was laid out in split plot design with three replications. The main plot consisted of different combinations of basal application of organic amendments and the sub plots consist of the foliar spray of biostimulants such as, humic acid, panchagavya and Manchurian mushroom. The results revealed that the treatment combination, M_1S_4 that is, FYM (25 t/ha) + recommended dose of fertilizer (90:45:35 kg/ha of NPK/ha) combined with foliar spraying of panchagavya and Manchurian mushroom extract each at 3% and humic acid at 0.3%, recorded highest plant height (227.53, 286.47, 300.1 and 334.54 cm), number of leaves (62.0, 70.0, 82.0 and 95.0) number of branches (36.048.055.058.0), leaf area (12.60, 15.52, 17.50 and 18.92 cm^2), fresh biomass (2.55, 3.22, 3.88 and 4.10 kg/plant) and dry biomass (0.638, 0.782, 0.890 and 0.913 kg/plant) at 180, 240, 300 and 360 days after transplanting, respectively. Regarding the quality parameters, the treatment combination, M_6S_4 (FYM (25 t/ha) + Vermicompost (5 t/ha) + Neem cake-250 kg) combined with foliar spraying of panchagavya and Manchurian mushroom extract each at 3% and humic acid at 0.3%, registered the highest crude gymnemic acid content of 485.74 mg 100 g^{-1} dry weight.

Key words: *Gymnema*, vermicompost, humic acid, panchagavya, Manchurian mushroom, gymnemic acid.

INRODUCTION

Gymnema sylvestre R.Br. is a woody perennial that grows in the tropical forests of central and southern India and has a deep history rooted in Ayurvedic medicine. Through out India in dry forests up to 600 m, it is distributed in Asia, Tropical Africa, Malaysia and Srilanka (Keshavamurthy, 1990). Currently, it is one of the major botanicals being administered for those suffering from diabetes, in boosting insulin levels and controlling healthy blood sugar levels. The leaves of *Gymnema* are used medicinally for its unique property to directly mask the tongue's ability to taste sweet foods and at the same time suppress glucose absorption from the intestine. This is the reason why it is known in Hindi as gurmar, or "destroyer of sugar". *Gymnema sylvestre,* a plant used in the Ayurvedic medicine of India for the treatment of diabetes mellitus, has been known from antiquity also to have an antisaccharin taste effect. The active principles are glycosides (several Gymnemic acids) which shows selective anaesthetic effect (Warren, 1969). Gymnema sylvestre has been used in India for the treatment of diabetes for over 2,000 years. It is interesting to note that gymnema extract given to healthy volunteers does not produce any blood sugar-lowering or hypoglycemic effects (Baskaran et al., 1990). In India, this plant is found growing in abundance in the forests of Karnataka, Tamil Nadu and Bihar. Due to its rising demand in south-east Asian countries like India and China, the plant is becoming endangered. Adoption of intensive farming practices such as application of high dose of chemical fertilizers particularly nitrogenous fertilizers, pesticides and fungicides are becoming more common in the traditional areas.

Residual toxicity and microbial contamination has been reported in crops due to such chemical intensive farming.

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There has been an exponential growth in the market for organic produce especially for culinary herbs and botanicals all over the world. The European countries hold the major share in import of organic produce. A recent estimate for current herbal medicine market world-wide may be around 30 – 60 billion dollars. The Indian herbal drug market is estimated to be around one billion dollars. The demand for herbal and organic products is increasing every year and by taking the advantage of rising green consumerism, India can acquire enviable position in the world. The organic cultivation of *Gymnema sylvestre*, being a medicinal herb, is highly beneficial to boost the exports and to get premium price in the world market. With this concept in view, the present investigation was made to study the effect of organic amendments and biostimulants on growth, yield and quality of *G. sylvestre*.

MATERIALS AND METHODS

The experiment was conducted at the Medicinal Plants Unit, Department of Spices and Plantation Crops, HC and RI, TNAU, Coimbatore during the period 2007 - 08. The experiment was laid out in split plot design comprising basal application of organic manures as main plots and foliar spraying of growth promoters in the sub plots. The following are the treatment details.

Main plot

M_1- FYM (2.5 kg/plant/ year) + Recommended dose of NPK/plant /year (Chemical check: 90:45:35 kg/ha)
M_2- FYM (2.5 kg/plant/ year) + Vermicompost (80 g / plant / month)
M_3- FYM (2.5 kg/plant/ year) + Vermicompost (160 g / plant / month)
M_4- FYM (2.5 kg/plant/ year) + Neem cake - 250 kg
M_5- FYM (2.5 kg/plant/ year) + Vermicompost (80 g / plant / month) + Neem cake-250 kg
M_6- FYM (2.5 kg/plant/ year) + Vermicompost (160 g / plant / month) + Neem cake - 250 kg

Sub plot

S_1- Foliar application of Panchagavya (3%) at monthly intervals (six sprays at 6, 8, 10, 12 and 14 months after planting)
S_2 - Foliar application of humic acid (3%) at monthly intervals (six sprays at 4, 6, 8, 10, 12 and 14 months after planting)
S_3 -Foliar application of Manchurian mushroom (3%) at monthly intervals (six sprays at 4, 6, 8, 10, 12 and 14 months after planting)
S_4 - Combination of all three (S_1 + S_2 + S_3)

Observations on morphological (plant height (cm), plant spread, biomass, fresh and dry weight) and yield parameters at different stages of plant growth such as 180, 240, 300 and 360 days after transplanting were recorded. The quality attribute and total gymnemic acid was estimated at 360 DAT as per the methods suggested by Rajpal (2002). About 3.00 gm of the leaf extract was weighed into a beaker. 10% of hydrochloric acid was added to the filtrate to make the pH 1.5. Later, the filtrate was distilled with water and the precipitate dissolved in 20 ml of 80.0% v/v ethanol or methanol. Filtrate was dried at 70°C to a constant weight to enable the calculation of the percentage of total gymnemic acid.

RESULTS AND DISCUSSION

Morphological parameters

The results revealed that basal application of organic manures and foliar spraying of biostimulants resulted in significant increase in plant height, number laterals and number of leaves in all the four stages of crop growth. The treatment combination, M_1S_4 that is, FYM (25 t/ha) + recommended dose of fertilizer (90:45:35 kg/ha of NPK/ha) combined with foliar spraying of panchagavya and manchurian mushroom extract each at 3% and humic acid at 0.3%, recorded highest plant height (227.53, 286.47, 300.1 and 334.54 cm), number of laterals (40.0, 52.0, 58.0 and 62.0) and number of leaves (62.0, 70.0, 82.0 and 95.0) at 180, 240, 300 and 360 days after transplanting, respectively (Tables 1, 2 and 3). The possible reason for this accelerated rate of growth pertaining to morphological parameters might be due to the increased availability of nitrogen supplied through inorganic sources of fertilizers.

Nitrogen, the chief constituent of protein, is essential for the formation of protoplasm, which leads to cell division and cell enlargement. Likewise, application of nitrogen (10 g/plant) promoted vegetative growth of *G. sylvetsre* (Subbaraj, 1997). In gymnema, since leaves are the economic part of the plant, integrated application of FYM along with inorganic sources might have boosted the crop growth significantly by increasing the availability of macro and micro nutrients as suggested by Sharma, 1999. Increase in primary and secondary branches, leaf area, and fresh and dry weight of vine was observed on application of equal proportion of NPK mixture (17:17:17) at 125 kg/ha (Prasad et al., 2002). Addition of any form of organics has been found to improve the soil health, buffering capacity, water retention capacity, chelation, release of micronutrients, microbiological process, etc., (Saravana Pandian et al., 2005).

Application of biostimulants would deliver naturally occurring non toxic growth promoting substances that would react synergistically with nutrients through various fermentation technologies and a variety of plant extracts (Anonymous, 2001). The synergistic effect of these three biostimulants carries considerable amount of nitrogen, which would be utilized for the protein synthesis, eventually resulting in stimulated growth. The humic acid contains gibberellins like substances, which may lead to increased plant growth as reported by Vaugham et al. (1985). The results of enhanced biometric parameters due to the application of panchagavya are in consonance with the findings of Kanimozhi (2004) in *Bacopa monnierri,* Sanjutha et al. (2006) in kalmegh and Ramasamy (2006) in periwinkle.

Yield parameters

Yield is a complex phenomenon which is influenced both

Table 1. Effect of organic amendments and biostimulants on plant height (cm) at different growth stages of *Gymnema sylvestre.*

Treatments	180 DAT	240 DAT	300 DAT	360 DAT
M_1S_1	225.40	284.24	298.57	332.50
M_1S_2	223.98	283.50	295.55	331.10
M_1S_3	222.45	280.84	292.14	330.78
M_1S_4	227.53	286.47	300.10	334.54
M_2S_1	174.10	207.62	237.4	254.33
M_2S_2	173.56	203.84	236.32	250.20
M_2S_3	172.50	200.12	235.47	248.32
M_2S_4	174.24	210.10	238.44	257.22
M_3S_1	185.78	244.40	264.84	290.85
M_3S_2	184.54	242.36	262.22	287.47
M_3S_3	182.75	241.24	261.78	284.35
M_3S_4	186.55	245.36	266.52	292.78
M_4S_1	179.84	231.31	255.54	273.34
M_4S_2	178.00	229.20	249.65	268.2
M_4S_3	177.54	228.45	246.35	265.56
M_4S_4	180.57	232.52	258.47	276.77
M_5S_1	189.22	260.24	276.77	310.47
M_5S_2	188.88	254.54	273.55	306.24
M_5S_3	187.62	250.89	271.18	302.32
M_5S_4	190.17	262.52	278.45	312.44
M_6S_1	199.62	274.47	285.56	323.10
M_6S_2	197.70	272.63	283.44	320.44
M_6S_3	196.45	270.54	280.35	318.48
M_6S_4	220.10	275.42	288.25	325.47
SED	12.463	18.132	17.244	14.316
CD (0.05)	25.280	36.773	34.976	29.039

Table 2. Effect of organic amendments and biostimulants on number of laterals at different growth stages of *Gymnema sylvestre.*

Treatments	180 DAT	240 DAT	300 DAT	360 DAT
M_1S_1	40.0	51.0	57.0	62.0
M_1S_2	38.0	50.0	56.0	61.0
M_1S_3	38.0	50.0	56.0	60.0
M_1S_4	40.0	52.0	58.0	62.0
M_2S_1	31.0	40.0	46.0	48.0
M_2S_2	30.0	38.0	45.0	47.0
M_2S_3	30.0	38.0	44.0	47.0
M_2S_4	31.0	40.0	46.0	48.0
M_3S_1	32.0	42.0	49.0	50.0
M_3S_2	31.0	41.0	48.0	49.0
M_3S_3	31.0	41.0	47.0	48.0
M_3S_4	32.0	42.0	49.0	51.0
M_4S_1	33.0	46.0	52.0	54.0
M_4S_2	32.0	45.0	50.0	53.0
M_4S_3	32.0	44.0	50.0	52.0
M_4S_4	34.0	46.0	52.0	54.0
M_5S_1	30.0	36.0	44.0	46.0
$M5S_2$	29.0	35.0	43.0	45.0

Table 2. Cont'd.

M_5S_3	28.0	35.0	42.0	45.0
M_5S_4	30.0	36.0	44.0	46.0
M_6S_1	36.0	48.0	54.0	57.0
M_6S_2	35.0	47.0	52.0	56.0
M_6S_3	35.0	46.0	52.0	55.0
M_6S_4	36.0	48.0	55.0	58.0
SED	1.877	2.805	2.802	2.116
CD (0.05)	3.807	5.691	5.683	4.292

Table 3. Effect of organic amendments and biostimulants on number of leaves at different growth stages of *G. sylvestre*.

Treatments	180 DAT	240 DAT	300 DAT	360 DAT
M_1S_1	62.0	69.0	82.0	94.0
M_1S_2	61.0	68.0	81.0	93.0
M_1S_3	60.0	67.0	80.0	92.0
M_1S_4	62.0	70.0	82.0	95.0
M_2S_1	47.0	53.0	66.0	72.0
M_2S_2	45.0	52.0	65.0	70.0
M_2S_3	45.0	52.0	65.0	70.0
M_2S_4	47.0	53.0	66.0	73.0
M_3S_1	52.0	59.0	70.0	80.0
M_3S_2	51.0	57.0	68.0	79.0
M_3S_3	50.0	57.0	68.0	78.0
M_3S_4	52.0	60.0	70.0	81.0
M_4S_1	58.0	65.0	77.0	89.0
M_4S_2	57.0	64.0	76.0	87.0
M_4S_3	56.0	63.0	75.0	87.0
M_4S_4	59.0	65.0	78.0	90.0
M_5S_1	49.0	55.0	68.0	76.0
M_5S_2	48.0	54.0	66.0	75.0
M_5S_3	48.0	54.0	66.0	74.0
M_5S_4	49.0	55.0	68.0	77.0
M_6S_1	56.0	62.0	73.0	84.0
M_6S_2	55.0	61.0	72.0	83.0
M_6S_3	53.0	61.0	71.0	82.0
M_6S_4	56.0	62.0	74.0	85.0
SED	3.445	4.349	3.478	3.969
CD (0.05)	6.988	8.822	7.054	8.051

by genetic as well as environmental factors. In any crop management programme, the ultimate objective is to achieve the maximum yield with high quality. Combined application of inorganic fertilizers along with organic manures and growth promoting chemicals were found to significantly influence the biomass yield of Gymnema sylvestre. From the present study, it is evident that application of FYM (25 t/ha) + recommended dose of fertilizer (90:45:35 kg/ha of NPK/ha) combined with foliar spraying of panchagavya and manchurian mushroom extract each at 3% and humic acid at 0.3% (M_1S_4), registered the highest values for fresh (2.55, 3.22, 3.88 and 4.10 kg/plant) and dry biomass (0.638, 0.782, 0.890 and 0.913 kg/plant) yield at 180, 240, 300 and 360 days after transplanting, respectively (Tables 4 and 5). Application of nitrogen (10g/plant) promoted the growth of

Table 4. Effect of organic amendments and biostimulants on fresh biomass (kg plant^{-1}) at different growth stages of *G. sylvestre.*

Treatments	180 DAT	240 DAT	300 DAT	360 DAT
M_1S_1	2.48	3.18	3.82	4.06
M_1S_2	2.36	3.14	3.79	4.02
M_1S_3	2.34	3.12	3.76	4.00
M_1S_4	2.55	3.22	3.88	4.10
M_2S_1	1.75	1.93	2.38	2.59
M_2S_2	1.73	1.91	2.35	2.55
M_2S_3	1.72	1.90	2.33	2.54
M_2S_4	1.78	1.94	2.40	2.60
M_3S_1	1.97	2.15	2.85	3.00
M_3S_2	1.95	2.10	2.80	2.96
M_3S_3	1.95	2.08	2.76	2.95
M_3S_4	1.98	2.20	2.88	3.07
M_4S_1	2.22	2.94	3.62	3.77
M_4S_2	2.20	2.88	3.54	3.54
M_4S_3	2.18	2.85	3.44	3.47
M4S4	2.25	3.00	3.65	3.95
M5S1	1.86	2.00	2.50	2.72
M5S2	1.84	1.98	2.49	2.68
M5S3	1.82	1.97	2.47	2.65
M5S4	1.89	2.01	2.54	2.75
M6S1	2.08	2.60	3.17	3.30
M6S2	2.05	2.51	3.14	3.24
M6S3	2.02	2.44	3.10	3.12
M6S4	2.10	2.64	3.20	3.35
SED	0.116	0.163	0.175	0.132
CD (0.05)	0.235	0.331	0.356	0.268

Table 5. Effect of organic amendments and biostimulants on dry biomass (kg plant^{-1}) at different growth stages of *G. sylvestre.*

Treatments	180 DAT	240 DAT	300 DAT	360 DAT
M_1S_1	0.634	0.764	0.874	0.913
M_1S_2	0.627	0.754	0.8703	0.907
M_1S_3	0.625	0.744	0.867	0.900
M_1S_4	0.638	0.782	0.890	0.913
M_2S_1	0.460	0.492	0.565	0.640
M_2S_2	0.455	0.487	0.557	0.637
M_2S_3	0.430	0.48	0.550	0.620
M_2S_4	0.467	0.495	0.587	0.657
M_3S_1	0.538	0.622	0.718	0.743
M_3S_2	0.527	0.617	0.714	0.723
M_3S_3	0.520	0.61	0.7103	0.713
M_3S_4	0.541	0.625	0.72	0.753
M_4S_1	0.610	0.711	0.818	0.860
M_4S_2	0.605	0.706	0.812	0.840
M_4S_3	0.600	0.702	0.8103	0.820
M_4S_4	0.612	0.712	0.822	0.877
M_5S_1	0.510	0.564	0.655	0.690
M5S2	0.499	0.553	0.642	0.690

Table 5. Cont'd.

M_5S_3	0.495	0.544	0.635	0.687
M_5S_4	0.514	0.587	0.674	0.700
M_6S_1	0.570	0.682	0.777	0.810
M_6S_2	0.564	0.673	0.754	0.790
M_6S_3	0.555	0.655	0.745	0.787
M_6S_4	0.587	0.686	0.788	0.810
SED	0.0335	0.0467	0.0298	0.0376
CD (0.05)	0.06898	0.0947	0.0606	0.0763

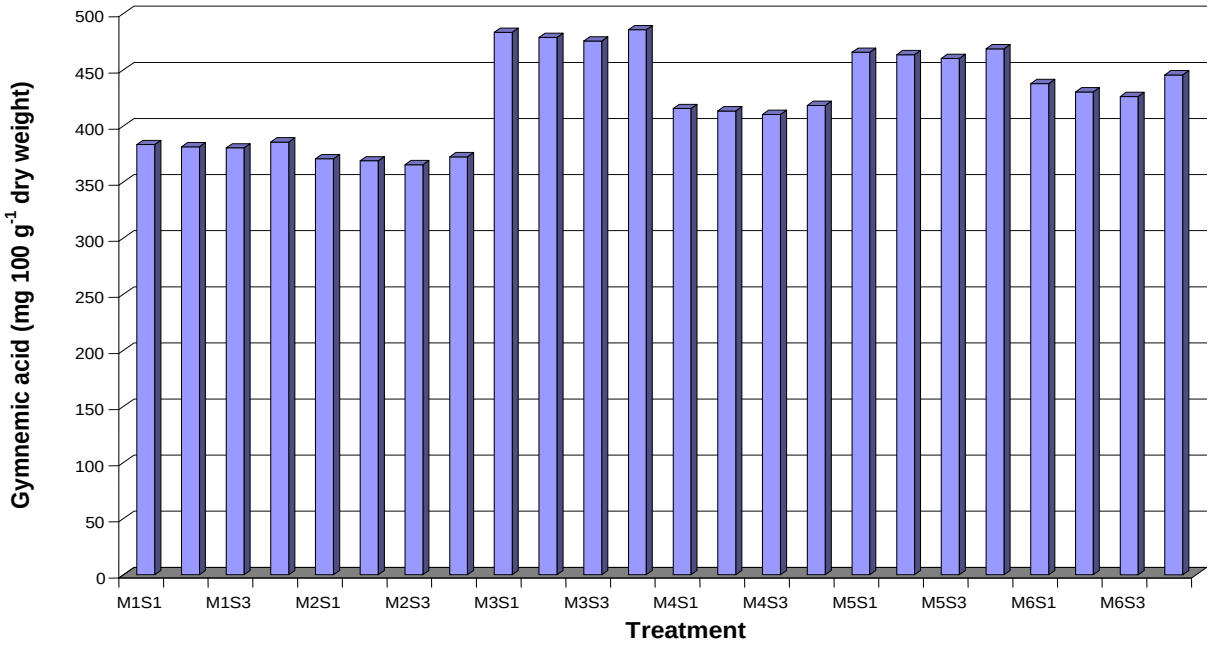

Figure 1. Effect of organic amendments and growth stimulants on gymnemic acid (mg 100 g-1 dry weight) of *Gymnema sylvestre.*

G. sylvestre significantly (Subbaraj, 1997). Application of organic manures along with inorganic fertilizers not only improves the physical status of the soil, but also increases the organic status of the soil. This results in rapid multiplication of beneficial soil microbes promoting the availability and uptake of nutrients by the plants, favouring an increase in yield (Gill et al., 1999).

Generally, the nutrients applied in the form of foliar spray are in readily available form and it was noted that the nutrients in liquid forms can be taken up by the plants quickly leading to increased yield. The growth promoting activity (greater movement and availability of phosphorus and micronutrients to the plant) of humic substances in biostimulants might also be a possible reason for higher yield by mobilization of the reserve food materials to the sink through increased activity of hydrolyzing and oxidizing enzymes.

This would have helped the better availability and utilization of nutrients. The growth hormones such as, IAA, GA_3 and cytokinin are rich in biostimulants leading to greater mobilization of nutrients, thereby increasing the leaf production (Sharma and Anandaraj, 2003). Prasad et al. (2002) reported increased primary and secondary branches, leaf area, fresh and dry leaf weight of vine in gymnema on application of equal proportion of NPK mixture (17:17:17) at 125 kg/ha.

Quality parameters

In *Gymnema*, the leaves are found to contain the active compounds in the form of acids group termed as gymnemic acids having antidiabetic, antisweetener and anti-inflammatory properties. Regarding the quality parameters, the treatment combination, M_6S_4 (FYM (25 t/ha) + Vermicompost (5 t/ha) + Neem cake - 250 kg

combined with foliar spraying of panchakavya and manchurian mushroom extract (fermented extract of mushroom) each at 3% and humic acid at 0.3% registered the highest gymnemic acid content of 485.74 mg $100g^{-1}$ dry weight (Figure 1). The gymnemic acid content is present abundantly in leaves. It is obvious that organic farming practices by using organic supplements enhance the quality attributes of a crop compared to their quantitative traits. In the present study, combined application of organic manures and growth promoters substantially improved the gymnemic acid contents of the crop. establishment and multiplication of beneficial symbiotic microbes which helps in fixing nitrogen in the soil, Vermicompost serves as a very good base for besides enhancing the availability of phosphate and nitrogen, and uptake of phosphate by plants (Kale, 1995). Similarly, application of biostimulants like humic acid and panchakavya would have induced the innate potential of the plant to synthesize the hormones like auxins and gibberellins resulting in better recovery of alkaloid content (Muscolo et al., 1999). The present finding of increased saponin content on application of panchakavya corroborates with the results of Cynthia (2003) in ashwaganda, who indicated increased withaferine content on application of panchakavya.

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