Study of the Impact of Organic Manures and Biofertilizers on growth of Phaseolus aureus Roxb.

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Abstract
Weeds are wild plants growing where they are not wanted, and they compete with the cultivated crop for nutrition. Though they are seen as agricultural waste throughout the year, they are rich sources of nutrients. They grow in abundance during the rainy season, but as the season ends these biomasses get wasted. In the present investigation, Tephrosia hamiltonii Drumm belonging to family Fabaceae, and Achyranthes aspera L. belonging to the family Amaranthaceae were used as a nutrient source to develop crop Phaseolus aureus Roxb. Weed manures, Vermicompost and Compost, were prepared by using weeds T. hamiltonii Drumm and A. aspera L. in 1:1 proportion. Chemical analysis of weed and weed manures were done before administering it into the soil. Neem cake was also used as one of the organic manures. In the experiment, a single dose of biofertilizers Azotobacter and Phosphate solubilizing bacteria were mixed with Weed Vermicompost, Weed Compost and Neem cake; and in one of the treatments, only biofertilizers were used indouble dose. Treatments were given to the crop as ATVB, ATCB, NCB, BioD, NPK, and Control in a randomized block design of experimental plot size 1.5X 1.5 m. The use of chemical pesticides or fertilizers was completely avoided except for NPK treatment plots. Single plant analysis of pulse crop P. aureus Roxb. was done. Observations were recorded in the forms of fresh weight and dry weight of root, stem, leaves, leaf (4th number), and legumes. Total fresh yield (kg ha⁻¹), DM (kg ha⁻¹) increase over control, and Nitrogen efficiency ratio were recorded. Results showed that %DM (an increase over control) and DM kg ha⁻¹ recorded highest in ATVB treatment and the highest N efficiency ratio was in BioD. The present investigation emphasized reducing the input cost of the farm products along with protection of the environment and natural resources.

Keywords: Biofertilizer, Neemcake, Organic agriculture, P. aureus, Weed compost, Weed Vermicompost.

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Introduction
Recent agricultural trends are focused on both reducing the usage of inorganic fertilizers by using organic manure and applying biofertilizers such as vermicompost and phosphatic biofertilizers [1]. Microbial activities play a key role in agriculture because they are significant in the movement and availability of minerals required for plant growth and ultimately lower the use of chemical fertilizers [2]. The maintenance of nutrients in the soil is most important for healthy plant growth [3]. Biofertilizers enhance soil health and crop yield. They improve fertility of soil, nutrient uptake, decomposition of crop residue, and microbial diversity of soil. They also reduce the requirement of chemical fertilizers [4]. The use of excessive chemical fertilizer, however, causes hazardous effects on the soil, leading to serious problems; thus, biofertilizers are important alternative sources of nutrients. They are biologically active microorganisms, like bacteria, algae, fungi; they can provide nutrients to crops [5, 6]. Among Biofertilizers, beneficial bacteria are Azotobacter, Azospirillum, Rhizobium, symbiotic fungi Mycorrhizae; they are essential in crop production. Biofertilizers improve plants’ resistance to an unfavorable environment [7]. The biological manure helps to increase crop yields, and also plays a vital role in the nutrient accessibility in soil by improving the physical, chemical, and biological structure of soil, and it enhances the utilization of applied fertilizers [8]. In developing countries, residue management is very important as the amount of nutrients in crop residue is several times higher than the quantities of these nutrients applied as high cost fertilizer [9]. Weed plants compete with the agricultural crops; they cause a tremendous reduction in crop yields and increase their production costs. Several scientists have estimated such losses in crop yields in different parts of India. A very broad-based
average of these estimates show that weeds reduced productivity of wheat by 15-30%, rice by 30-35%, and maize, sorghum, pulses and oilseeds by 18-85% each. Many cases of complete crop failure due to weeds particularly in upland rice and vegetable crops were recorded [10, 11, 12]. *T. hamiltonii Drumm.* and *A. aspera* L. are the weed plants used in the present study. The present study emphasized conversion and utilization of weeds beneficially by using them for the preparation of compost and vermicompost. Neem cake is a residue left after the extraction of neem oil and used as an organic fertilizer. With the utilization of these organic manures along with biofertilizers like Azotobacter and Phosphate solubilizing bacteria for the cultivation of Pulse crop *P. aureus Roxb.* belonging to the family Fabaceae.

We can minimize the cost of production, increase output per hectare by using organic manures like compost, vermicompost prepared from weed biomass, Neem cake, and biofertilizers for the production of crops and for sustainable agriculture.

**Material and Methods**

The experiment was conducted during March 2008. A summer variety of *P. aureus* Roxb. was cultivated at college campus of New Arts, Commerce and Science College, Shevgaon District Ahmednagar, (Maharashtra), India. Shevgaon extends between 19°13 North latitude to 75°35 East longitude and between 75°01 East longitudes to 75°37 East longitude.

**Weed collection and preparation of manures**

The fresh vegetation of weeds i.e. *Aghada* (*A. aspera* L.) and *Unhali* (*T. hamiltonii* Drumm.) were collected from different localities and chopped into small pieces (2-3 cm) by locally available iron cutter. Equal amount (6944+6944 kg ha⁻¹) 1:1 proportion mixture of weed pieces were used for the preparation of compost and vermicompost. To prepare compost this material was placed into pit (90x90x90 cm) and then added cow dung, soil and weed plant material layer by layer and sprinkled with water per requirement. Finally, the compost pit was sealed with dung-mud mixture to prevent loss of heat and moisture. After partial decomposition first turning was given after 15 days for homogeneous decomposition, subsequent turnings were given after every 15 days interval. Sufficient water was sprinkled to maintain moisture. Finally, amorphous, dark brown, well fermented compost was obtained within 70 days. Fresh weight of compost obtained from pit was 33 kg. Same procedure was applied for vermicomposting, only with the addition of the worms in the pits after 15 days (Worms’ variety *Eudrilus eugeniae* and *Iceniaps foetida*). Identification of earthworms was done by the method prescribed in Fauna of India and Adjacent countries [13]. The prepared vermicompost was used for field trials. Fresh weight of vermicompost obtained from pit was 32 kg. The uniformly mixed samples (100 g) were collected immediately from the pit for nutrient analyses. Chemical analyses of weeds and weed manures and Neem cake were done using oven dried and pulverized powder of samples. All the manures compost, vermicompost and neemcake (1000 kg ha⁻¹) were mixed with biofertilizer *Azotobacter* and Phosphate solubilizing bacteria at the rate 25 kg ha⁻¹ (recommended dose); and only Biofertilizer double dose treatment 50 kg ha⁻¹ in two split doses were applied to appropriate plots except chemical fertilizer (NPK) plots. The Mung (*P. aureus* Roxb.) Variety “Raj Biotech” Balwan R.J. Biotech, Pvt Ltd. Siddharth Arcade, Station Road, Aurangabad was sown in the research plots of size 1.5 x 1.5 m. at the rate of 20 kg ha⁻¹.

**Application of Inorganic Fertilizers**

The inorganic fertilizers were supplied to the experimental plots as Nitrogen (N), Phosphorus (P) and Potassium (K) through urea, single super phosphate (SSP) and muriate of potash at the rate of 25 kg N, 50 kg P and ‘0’ K kg ha⁻¹ (25:50:0) only for fertilizer treatment plots. Entire amount of P₂O₅ and K₂O and N was applied at the time of sowing. The crop supplemented with irrigation during periods of growth and whenever necessary weeding was done. Use of insecticides and pesticides was completely avoided.

Seeds were planted in rows at a distance 30 cm x 10 cm. Soil was murum so the crop was grown under frequent irrigation after each 8-10 days. Sample from each plot was brought into laboratory chopped into 3-4 cm pieces. Measured amount of biomass was kept in digital electrical oven separately in pre-weighted tray at 95±5°C for 48 hours or more till constant weight. Weight of dried samples were reported as DM. Results were used to calculate %DM, DM Kg ha⁻¹, increase over control and Nitrogen efficiency ratio of crop.
Results

Table 1. Analyses of weeds administered in experimental plots through compost and vermicompost weed manures. Here, weeds used were A. aspera L. and T. hamiltonii Drumm. Kg plot\(^{-1}\) (Plot size 1.5 m X 1.5m)

| Weed Name | Kg plot\(^{-1}\) | Kg ha\(^{-1}\) | % | DM | Kg ha\(^{-1}\) | % N | Kg ha\(^{-1}\) | Ash | P | K | C | C.N |
|-----------|----------------|-------------|---|-----|-------------|-----|-------------|-----|---|---|---|-----|
| Achyrantes | 1.56 | 6944 | 19.29 | 1339.50 | 2.03 | 27.19 | 17.43 | 0.123 | 0.43 | 10.11 | 4.99 |
| Tephrosia | 1.56 | 6944 | 22.40 | 1555.46 | 1.94 | 30.18 | 18.57 | 0.115 | 0.51 | 10.77 | 5.54 |

Table 2. Analyses of weed manure and Neem cake amendment along with biofertilizer. Here, ATV=B=Achyranthes, Tephrosia vermicompost mixed with Biofertilizer single dose, ATCB=A=Achyranthes, Tephrosia compost along with Biofertilizer single dose, NC=Neem cake along with Biofertilizer single dose

| Treatments | Fresh weight | Fresh weight | % | DM | Nitrogen | % |
|------------|--------------|--------------|---|-----|----------|---|
| ATV        | 2.00         | 8889         | 67.21 | 5974.30 | 0.42 | 25+4.485 | 0.13 |
| ATCB       | 2.06         | 9169         | 65.07 | 5966.27 | 0.50 | 30+4.485 | 0.12 |
| NCB        | 0.23         | 1000         | 97.94 | 0979.40 | 1.96 | 19+4.485 | 0.81 |

(Amount of Nitrogen fixed by single dose of biofertilizer is 4.485 kg ha\(^{-1}\) as according to N balance method[14]). These values added in N kg ha\(^{-1}\) of other treatments and amount of N kg ha\(^{-1}\) fixed by Azotobacter biofertilizer double dose was 8.97 kg ha\(^{-1}\).

Table 3. C:N ratio of organic amendments. Here, ATV=A=Achyranthes, Tephrosia vermicompost, ATC=A=Achyranthes, Tephrosia compost, NC=Neem cake

| Treatments | Ash | % | C | N | C:N |
|------------|-----|---|---|---|-----|
| ATV        | 32.00 | 18.56 | 0.42 | 44.56 |
| ATC        | 36.50 | 21.17 | 0.50 | 42.36 |
| NC         | 74.93 | 43.46 | 1.96 | 22.17 |

Table 4. Fresh wt and DM analyses of Single Plant of Phaseolus (at 56 DAS). Here, ATV=B=Achyranthes, Tephrosia vermicompost mixed with Biofertilizer single dose, ATCB=A=Achyranthes, Tephrosia compost mixed with Biofertilizer single dose, NC=Neem cake along with Biofertilizer single dose. BioD=Biofertilizer double dose, NPK=Inorganic fertilizer, CON=Control, (DAS=Days after sowing)

| Treatment | Plant Fresh wt in gm | DM in gm |
|-----------|----------------------|----------|
| ATV       | 48.50 | 112.9 | 12.39 | 25.55 | 5.83 | 0.21 | 1.44 | 2.99 | 0.82 | 7.99 | 2.71 |
| ATCB      | 48.25 | 109.7 | 12.56 | 20.83 | 4.21 | 0.19 | 1.17 | 2.77 | 0.60 | 6.47 | 3.03 |
| BioD      | 60.50 | 12.87 | 3.92 | 24.37 | 5.44 | 0.24 | 1.40 | 3.25 | 0.70 | 8.30 | 3.49 |
| NCB       | 39.25 | 06.79 | 1.66 | 12.71 | 2.65 | 0.13 | 0.74 | 1.80 | 0.43 | 4.20 | 1.67 |
| NPK       | 51.75 | 08.38 | 2.15 | 15.09 | 2.63 | 0.20 | 1.08 | 2.21 | 0.53 | 4.76 | 1.60 |
| CON       | 19.15 | 03.49 | 1.14 | 07.26 | 1.72 | 0.08 | 0.47 | 1.02 | 0.27 | 2.33 | 1.41 |
| S.E       | 0.05 | 0.37 | 0.44 | 2.61 | 0.68 | 0.02 | 0.16 | 0.34 | 0.10 | 0.95 | 0.36 |
| C.D.      | 0.12 | 1.32 | 2.92 | 5.56 | 1.45 | 0.05 | 0.33 | 0.73 | 0.21 | 2.02 | 0.76 |

Table 5. Increase over control of total biomass and Nitrogen efficiency ratio (73DAS). It is the ratio of the crop Nitrogen uptake to the total input of Nitrogen fertilizer.

| Treatment | FW Kg ha\(^{-1}\) | Increase Over Con | % | DRY WT Kg ha\(^{-1}\) | Increase Over Con | % | N Efficiency Ratio |
|-----------|----------------|------------------|---|---------------------|------------------|---|-------------------|
Analyses of weeds were done on dry matter basis. Observations of weed analyses are recorded in the Table 1. Fresh weight were used, 1.56 kg plot⁻¹ (6944 kg ha⁻¹) of each weed, for preparations of manures. % DM of Achyranthes weed was higher (22.4), followed by Tephrosia (19.29). The DM kg ha⁻¹ of Tephrosia weed was higher (1555.46) followed by Achyranthes weed (1339.50). % N was higher in Achyranthes (2.03) followed by Tephrosia (1.94) (Tephrosia weed was collected from comparatively non fertile land and Achyranthes from fertile land with ample domestic waste nearby. So, Nitrogen percent of Tephrosia was less than Achyranthes though it is leguminous weed). N kg ha⁻¹ of Tephrosia weed was higher (30.18) it was followed by Achyranthes (27.19). % Ash of Tephrosia weed was higher (18.57); it was followed by Achyranthes (17.43). % P was higher in Achyranthes (0.123) followed by Tephrosia (0.115). % K was higher in Tephrosia (0.51) followed by Achyranthes (0.43). % Cof Tephrosia weed was higher (10.77); it was followed by Achyranthes (10.11). C:N ratio of Tephrosia weed was higher (5.54), and it was followed by Achyranthes (4.99).

Analyses of Achyranthes and Tephrosia weed manure and Neemcake were done; it is presented in Table 2. Fresh weight of weed compost (ATC) was administered at the rate of 2.06 kg plot⁻¹ (9169 kg ha⁻¹) and weed Vermicompost (ATV) was added at the rate of 2.00 kg plot⁻¹ (8889 kg ha⁻¹). Fresh weight of Neem cake (NC) was used at the rate of 0.23 kg plot⁻¹ (1000 kg ha⁻¹). All the manures treatment was mixed with single dose of biofertilizer i.e. 25 kg ha⁻¹. Double dose of biofertilizer 50kg ha⁻¹ was given to the biofertilizer treatment (BioD). % DM of ATV (on 211th day) was 67.21 %; it was followed by ATC (on 211th day) was 65.07% and Neemcake 79.94%. DM kg ha⁻¹ was highest in Vermicompost (5974.30) followed by Compost (5966.27) and lowest in Neemcake (979.40). % N and N kg ha⁻¹ was highest in ATCB (0.5%, 30 kg) followed by ATV (0.42%, 25 kg) and NCB (1.96%, 19 kg). Single dose of biofertilizer fixed 4.485 Nkg ha⁻¹; so, input of N was 29.485 for ATV, 34.485 for ATCB and 23.485 for NCB. %Phosphorus recorded highest in Neem cake (0.81) and % Potassium in Neemcake (0.48); and %Ca was highest in ATC (4.3).

Amount of Nitrogen fixed by single dose (recommended dose) of biofertilizer was 4.485 kg ha⁻¹ and amount of N kg ha⁻¹ fixed by Azotobacter biofertilizer double dose was 8.97 kg ha⁻¹ in according to N balance method [14]. As per Table 3, % ash % C and % N were highest in NC (74.93, 43.46 and 1.96, respectively) followed by ATC (36.5, 21.17, & 0.5) and lowest in ATV (32, 18.56 & 0.42). C:N ratio was highest in ATV (44.56), followed by ATC (42.56) lowest in Neemcake (22.17).

In Table 4, Analyses of Fresh weight and Dry weight of single plantare presented. Fresh weight of root was highest in BioD (0.60) followed by NPK, ATV, ATCB, NCB and lowest in the CON (0.19), statistically significant in all the treatments. The fresh weight of stem leaves and 4th leaf and total plant was highest in BioD followed by ATV, ATCB, NPK, NCB and lowest in the CON (0.19), statistically significant in all the treatments except in NCB and NPK. The DM of root was highest in BioD (0.24) followed by ATV, NPK, ATCB, NCB and lowest in the CON (0.08), statistically significant in all the treatments. DM of stemwas highest in ATV (1.44) followed by BioD, ATV, NPK, NCB and lowest in CON (0.47), statistically significant in all the treatments except in NCB. DM of leaves, 4th leaf and total plant was highest in BioD followed by ATV, ATCB, NPK, NCB and lowest in CON (0.47), statistically significant in all the treatments except in NCB. DM of leaves, 4th leaf and total plant was highest in BioD (3.49) followed by ATCB, ATV, NPK and lowest in CON (1.41), statistically not significant in NCB for 4th leaf and total plant. DM of legume was highest in BioD (5.83) followed by BioD, ATV, ATCB, NPK and lowest in CON (5.0), statistically not significant in all the treatments except in ATV and NPK. The DM of legume was highest in BioD (5.83) followed by BioD, ATV, ATCB, NPK and lowest in CON (1.41), statistically not significant in all the treatments except in ATV and NPK.

In Table 5, Percent increase over control and nitrogen efficiency ratio is presented. The percent increase over control in Phaseolus for fresh weight was found highest in ATV (55.71) followed by ATCB (43.31), BioD (37.44), NCB (15.64), and minimum in NPK (12.33). Similarly, dry matter percentage (DM%) was found maximum with the treatment ATV (72.02) followed by ATCB (55.96), BioD (46.01), NCB (19.17) and minimum in NPK (10.47).DM kg ha⁻¹ recorded highest in ATV (1660) followed by ATCB, BioD, NCB, NPK and lowest in CON (965), statistically significant in ATV, ATCB, BioD, but statistically not significant in NPK and NCB. The nitrogen efficiency ratio for fresh weight was found highest in BioD (125.60) followed by ATV (56.85), ATCB (37.79), NCB (20.04) and lowest
in NPK (14.84). Similarly, the nitrogen efficiency ratio for Dry matter (DM) was found highest in BioD (49.50) followed by ATVB (23.57), ATCB (15.66), NCB (7.88) and lowest in NPK (4.04). Highest Fresh weight and DM kg ha\(^{-1}\) was recorded in Treatment ATVB.

**Discussion**

*Azotobacter* treated seedlings of knolkhol showed the highest whole plant weight [15]. Biofertilizers such as *Azotobacter*, *Azospirillum*, PSB, and a mixture of Aza + Azo + PSB were administered to crops which showed the increased plant fresh weight, dry weight [16]. Similar results showing fresh weight and dry weight of BioD treatment was recorded highest at 56 DAS. Combined inoculation of soybean by symbiotic bacteria improved the dry weight of soybean [17]. Vermicompost and phosphate biofertilizer showed improved growth and yield in *Anise (Pimpinella anisum L)* [1]. Vermicompost and PSB when applied together was found helpful in developing production and yield in anise [18]. *Azotobacter* increases the production of agriculture crop plants by 10-12%. *Azotobacter* can also improve growth and grain yield in wheat crops. *Azotobacter* act as one of the vital biofertilizers in the case of rice and some cereals could be applied by seed dipping and seedling root dipping methods [19]. Maize hybrid seed priming with *Azotobacter* showed the highest grain yield (7.01 ton/ha) and DM accumulation (2019 gr/m²) in treatment compound SC-434 [20]. Panchgavyawas found to contribute to better growth and yield of *Pisum sativum* as compared to NPK [21].

In biochemical analyses of the total biomass of plant, Nitrogen, and total crude protein was recorded highest in ATVB [22]. The findings of the present experiment showed that Fresh weight and Dry weight was recorded highest in Biofertilizer double dose at 56 DAS. But at harvesting 73 DAS maximum Fresh and Dry Yield was recorded highest in Weed vermicompost + Biofertilizer *Azotobacter* and Phosphate solubilizing bacteria treatment (ATVB).

**Conclusion**

The results of this investigation concluded that weed vermicompost, weed compost along with a single dose of biofertilizer and biofertilizer double dose can effectively be used as a nutrient source to increase crop yield and soil fertility. Weed manures and Neem Cake with biofertilizers worked more efficiently as compared to the chemical fertilizers (NPK) to improving the quality of the crop; it could reduce the input cost of the farm produce as well in addition to protecting the environment and natural resources.

**Author’s Contribution**

CB is responsible for all the data collection, conceptualization, writing – original draft preparation, review & editing the final draft of the manuscript. CB read and approved the final manuscript.

**Competing Interests**

No competing interests

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**Ethical Approval and Consent**

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