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Effect of Azotobacter chroococcum Inoculated Lantana camara Compost on Wheat Crop Yield

Shivani Chauhan^{*} & Saneel K. Thakur**

^{*}Department of Veterinary Pharmacology and Toxicology, COVAS, CSKHPKV, Palampur, Kangra (H.P.), India ^{**}Department of Chemistry, Sri Sai University, Palampur, Kangra (H.P.), India 176061

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ABSTRACT: The manurial utility of abundantly available lush green succulent biomass of *Lantana camara* was investigated through two waste management practices *i.e.* directly incorporated as well as composting in wheat crop. The compost was inoculated with *Trichoderma viride* and *Azotobacter chroococcum* cultures for rapid quality composting. Direct incorporation of green biomass and utilization as Lantana compost proved beneficial in improving grain and straw yields of wheat. Taking mean values of the yield of wheat obtained at four levels (N₀, N₆₀, N₉₀, N₁₂₀) of applied nitrogen it was observed that utilization of Lantana both as green matter and compost had significant effect on grain and straw yield in wheat crop over no Lantana. Grain and straw yields increased with increasing levels of nitrogen upto the level of 120kg N/ha. Also, the incorporation of Lantana as green matter or compost increased the grain protein of wheat crop. Results of the studies brought out conclusively that the abundantly available green biomass of Lantana with exorbitant power of regeneration could be directly utilized as organic manure provided the crop rotation as well as the weather conditions permit the green matter to decompose for atleast twenty days before sowing of the next crop. Out of the two practices the utilization of Lantana as compost proved to be better.

Key words: Lantana camara, compost, Azotobacter chroococcum, nitrogen fixing bacteria

INTRODUCTION

Depletion of non-renewable sources of energy, escalating cost of fertilizers and environment quality of aspects necessitated the review of various approaches focusing on the use of available renewable sources of plant nutrition for sustainable agricultural production. As a result renewed research efforts are being made systematically to evaluate the feasibility and efficiency of bio-degradable wastes of produced at farms, agroindustries and cities, in re-furnishing soil productivity and improving the efficiency of chemical fertilizers.

Compositing is one of the few natural processes capable of stabilizing organic wastes. Development of technologies of composting and co-composting for the production of quality manures from wastes of diverse origin and nature has therefore, become a vital issue for not only meeting the requirements of organic matter for sustaining higher productivity of crops but also to converse our resources. Compost when properly prepared and used can help to promote low-input agricultural system to become more sustainable and productive (Golabi *et al.*, 2004). Composted organic material contains essential nutrients for plant growth, especially N and P Beltran *et al.*, (2002) and Nyamangara *et al.* (2003) reported that the organic waste composted manure application even enhance the use efficiency of mineral nitrogen fertilizer by crops when the two were applied in combination.

Manure is readily available with farmers but its nutrient concentration is low. There is possibility of improving the nitrogen content of compost by inoculation with nitrogen fixing microorganisms, which convert dinitrogen to ammonia. One of the most interesting non-symbiotic bacteria that have great potential for use in production of biofertilizer is *Azotobacter chroococcum*, having ability to fix atmospheric nitrogen (N₂) into ammonia in a reaction catalysed by nitrogenase enzyme complex (Sabra *et al.*, 2000; Bakulin *et al.*, 2007).

Lantana camara (common name Lantana, Wild sage, locally known as *Bara Phulnoo* in Himachal Pradesh) is one of the most widely occurring shrub that has shown alarming growth in recent years, in the state of Himachal Pradesh. The plant has now spread to almost all states in India. It belongs to the family *Verbenaceae*, is an exotic ornamental shrub. Complete eradication of Lantana is both difficult and costly (Kohli and Rani, 1994; Shrestha Vaidya *et al.*, 2005).

Lantana biomass has potential for utilization as organic manure, has antimicrobial, insecticidal and medicinal properties also. Lantana camara makes available huge nitrogen rich succulent biomass, which has potential to be utilized as a substrate for organic recycling. Recycling of Lantana camara has been practiced sometimes as unprocessed *i.e.* direct incorporation of plant in the field or processing it as compost. No study has been carried out to compare the relative efficiency of processed and unprocessed biomass utilization of Lantana in crop production. The addition of compost or green manure is an important way to improve the soil in degraded areas since nitrogen and other nutrients, as well as organic matter which improves soil structure, is added with the organic material (Caravaca et al. 2002; Muthukumar and Udaiyan, 2000, Nziguheba et al. 2000; Shrestha Vaidya et al. 2007).

Keeping in view the importance and scope of composting and ever spreading menace of Lantana as described above, the present investigation was planned to study the relative efficiency of Lantana compost and fresh biomass application of Lantana with graded doses of nitrogen fertilizer on the grain and straw yields as well as grain protein in wheat crop.

MATERIALS AND METHODS

Procurement and Maintenance of Culture: Cultures of *Azotobacter chroococcum* and *Trichoderma viride* were obtained from the Department of Microbiology, College of Basic Sciences, CSKHPKV Palampur, Kangra (H.P.) and the microorganisms were maintained on Jensen's agar slants and Potato dextrose agar slants, respectively.

Survival of *Azotobacter chroococcum* **during composting:** Survival of *Azotobacter* was monitored at 60th and 90th day of composting by most probable number given by Alexander (1982).

pH: The pH was determined by using digital pH meter (Decibel, India) by diluting the sample in the ratio of 1:10 with water (w/v) as described by Jackson (1972).

Moisture: Moisture was determined by the method of AOAC (1970).

Organic Carbon: Organic carbon was estimated by combustion method as given by Black (1982) by ashing the material in muffle furnace at 650° C for 5 hours. The loss in organic matter was divided by 1.724 to determine the organic carbon content.

Total Nitrogen: Total nitrogen was determined by Kjeldahl's method given by Black (1982) using Kjeldatherm (Gerhardt, Germany).

Field Evaluation of *Lantana camara*: Two practices of utilization of green matter of *Lantana camara* were used

(i) Green Biomass of Lantana

Clippings of *Lantana camara* including leaves and succulent twigs were collected. These were manually cut to about 5cm size and incorporated @5t/ha on the surface soil (0-15cm) of the respective experimental plots with the help of spade in the Research Farm of the Dept. of Microbiology, College of Basic Sciences, CSKHPKV, Palampur Kangra (H.P.). The field was irrigated and the incorporated material was allowed to decompose for 20 days before sowing of wheat crop.

(ii) Lantana camara compost preparation

Compost was prepared from green biomass of Lantana supplemented with dung, soil, and FYM (Table 1) in the ratio of 8:1:0.5:0.5 (on dry weight basis) respectively (Anonymous, 1993). The preparation of compost was done in 1 m³ cemented pits. The pits were filled layer by layer using substrates, microbial inoculants, as per Table 2. On the basis of quality testing (unpublished data) the compost of T₄ treatment was used for field evaluation. The compost was applied @5t/ha in the respective experimental fields as per treatments (Table 3). Just before trial chemical compostion of Lantana green & Lantana compost was also analysed (Table 4).

Table 1. The chemical profile of substances used for the preparation of compost.

| Parameters | eters Values (on dry weight basis) | | | |
|--------------|------------------------------------|-------|-------|-------|
| | Lantana | Dung | Soil | FYM |
| Carbon (%) | 54.88 | 44.81 | 0.79 | 22.6 |
| Nitrogen (%) | 1.78 | 1.02 | 0.07 | 0.81 |
| C:N ratio | 30.83 | 43.93 | 11.28 | 27.90 |
| Moisture (%) | 69.10 | 76.23 | 27.18 | 69.21 |
| pH (1:10) | 6.92 | 6.71 | 5.78 | 7.22 |

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Table 2. Compost Treatments.

| Treatments | Descriptions |
|----------------|--|
| T ₁ | Lantana camara |
| T ₂ | $T_1 + Trichoderma viride$ |
| T ₃ | $T_1 + Azotobacter chroococcum$ |
| T ₄ | $T_1 + Trichoderma viride + Azotobacter chroococcum$ |

Table 3. Description of Field Treatments.

| Treatments | Description |
|-----------------|---|
| T ₁ | Direct incorporation of green biomass of Lantana @5t/ha |
| T ₂ | $T_1 + 60 \text{ kg N/ha}$ |
| T ₃ | $T_1 + 90 \text{ kg N/ha}$ |
| T_4 | $T_1 + 120 \text{ kg N/ha}$ |
| T ₅ | Compost of Lantana @5t/ha |
| T ₆ | $T_5 + 60 \text{ kg N/ha}$ |
| T ₇ | $T_5 + 90 \text{ kg N/ha}$ |
| T ₈ | T ₅ + 120 kg N/ha |
| T ₉ | 0 kg N/ha |
| T ₁₀ | 60 kg N/ha |
| T ₁₁ | 90 Kg N/ha |
| T ₁₂ | 120 Kg N/ha |

Table 4. Chemical Composition of Lantana Green and Lantana Compost.

| Parameter | Biomass Incorporated as direct | Biomass Incorporated through compost |
|--------------|---------------------------------------|---|
| Carbon (%) | 55.30 | 30.78 |
| Nitrogen (%) | 1.75 | 2.18 |
| C:N ratio | 31.60 | 14.11 |

The wheat crop (*Triticum aestivum*) variety HPW-89 was raised using twelve treatments (Table 4) in Randomized Block Design with three replications. The plot size used was $5x2 \text{ m}^2$. The crop was sown and raised under irrigated conditions. One half of the total nitrogen was applied at the sowing time through urea (46% N), the remaining half dose of nitrogen was top-dressed after full germination of seeds. All the standard agricultural practices were followed during growth of the crop. The crop was raised upto maturity, harvested and data on the grain and straw yields of the crop was recorded. Grain and straw samples were collected separately, oven dried at $70^{\circ}C \pm 5^{\circ}C$ ground in Wiley mill and stored in paper bags, for subsequent analyses.

Statistical analysis: The data obtained from field and laboratory studies were statistically analysed by the method of Gomez and Gomez (1984). The treatment effects were characterized on the basis of CD at 5% level of significance.

RESULTS

Survival of *Azotobacter* **during composting:** The survival of *Azotobacter chrooococcum* was monitored periodically at 60 and 90 days of composting with initial *Azotobacter* population of 88 x10⁶ cell/g. *Azotobacter* was inoculated only in treatment T_3 and T_4 (Table 2). Initially there was decrease upto 60 days and then it got stabilized subsequently upto 90 days (Table 5).

| Table 5. Surviva | l of Azotobacter | during | Composting. |
|------------------|------------------|--------|-------------|
|------------------|------------------|--------|-------------|

| Treatment | Azotobacter Population (X10 ² cells/g | | |
|---------------------------------|--|----|--|
| | Days of Composting | | |
| | 60 | 90 | |
| Lantana+Azotobacter chroococcum | 78 | 7 | |
| Lantana+Azotobacter+Trichoderma | 96 | 9 | |

Field evaluation of green matter and compost of Lantana

Grain and straw yield: The perusal of data on grain and straw yields of wheat crop revealed that Lantana manures significantly increased the grain and straw yields over no Lantana. The highest grain and straw yields were recorded by Lantana compost, which was significantly higher than the straw and grain yield obtained by Lantana green. Nitrogen levels significantly affected the grain and straw yields of wheat crop. The Grain and straw yields increased significantly with increase in level of nitrogen from 0-120kgN/ha. The highest grain and straw yields were obtained by application of 120kgN/ha (N_{120}) which was significantly higher than N_{90} and N_{60} .

Application of Lantana green or Lantana compost along with 60kgN/ha recorded yields statistically at par with the yield obtained by the application of 90kgN/ha alone. Similarly, application of Lantana green or Lantana compost along with 90kgN/ha recorded straw yield statistically at par with the yields of obtained by the application of 120kg N/ha alone (Table 6).

Table 6. Effect of Lantana manures and fertilizer nitrogen on the yield of wheat.

| Wheat yield (kg/ha) | | |
|---------------------|-------|-------|
| | Grain | Straw |
| Lantana manures | | |
| No Lantana | 2397 | 3524 |
| Lantana green | 2572 | 3898 |
| Lantana compost | 2760 | 4226 |
| CD(<0.05) | 102 | 124 |
| Nitrogen Levels | | |
| N ₀ | 1654 | 2469 |
| N ₆₀ | 2454 | 3729 |
| N ₉₀ | 2906 | 4409 |
| N ₁₂₀ | 3290 | 4923 |
| CD(<0.05) | 126 | 146 |

Effect of Lantana manures and fertilizer nitrogen on the grain protein in wheat: The data revealed that Lantana manures significantly affected the grain protein over no Lantana. Further, both Lantana green as well as Lantana compost had almost similar effect on grain protein (Table 7). The data revealed that Lantana manures significantly affected the grain protein over no Lantana. Nitrogen application at different levels (N_{60} , N_{90} and N_{120}) significantly effects on the grain protein over no nitrogen. The grain protein content was observed to increase with increase in nitrogen levels. Therefore, maximum grain protein was recorded with the application of 120kgN/ha (N_{120}) followed by 90kgN/ha (N_{90}).

Table 7. Effect of Lantana manures and fertilizer nitrogen on the grain protein in wheat.

| | `Grain protein (%) | |
|------------------|--------------------|--|
| Lantana manures | | |
| No Lantana | 11.5 | |
| Lantana green | 11.9 | |
| Lantana compost | 11.9 | |
| CD(<0.05) | 0.3 | |
| Nitrogen Levels | | |
| N ₀ | 11.1 | |
| N ₆₀ | 11.8 | |
| N ₉₀ | 12.0 | |
| N ₁₂₀ | 12.1 | |
| CD(<0.05) | 0.4 | |

DISCUSSION

Survival of Azotobacter chroococcum during Composting: There is possibility of enriching the compost with regard to its nitrogen content by inoculating Azotobacter. But such enrichment can be done only after the material has decomposed partly and not before composting, since, Azotobacter chroococcum does not have the ability to survive at high temperature that prevail during the first month of decomposition process. The survival of Azotobacter inoculated at 30th day of composting was thus monitored at 60 and 90 days of composting. The initial Azotobacter population was recorded to be 88×10^6 cells g^{-1} . The population decreased from initial to final stages tremendously, which may be due to the inhibition of microbial activity. The Azotobacter population however, decreased upto 60 days of composting and then it got stabilized subsequently at 90 days of composting *i.e.* $7x10^2$ cells g⁻¹ and $9x10^2$ cells g⁻¹ in treatment T₃ and T₄ respectively. These findings are in accordance with (Kapoor et al. 1983; Thakur and Sharma, 1998; Banta and Dev, 2009).

Field evaluation of green matter and compost of *Lantana*: Addition of organic matter such as green manure is a common practice to improve soil nutrient content and soil structure. Organic residues from plants such as *Tithonia diversifolia* and *Lantana camara* have been found to be especially beneficial since they are reported to have a high content of N and P, which is mineralized rapidly from the organic material (Nziguheba *et al.* 2000)

Taking mean values of the yield of wheat obtained at four levels of applied nitrogen it was observed that utilization of Lantana both as green matter and compost had significant effect on grain and straw yield in wheat crop over no Lantana. The useful effect of green matter of Lantana can be attributed to its high nitrogen content and succulent nature (Bhardwaj and Kanwar, 1991). It improves the microenvironment in root zone (Rhizosphere), which in turn, increased the N and P use efficiency by wheat crop, which was ultimately reflected in the yield data Tripathi et al., (1992). Nziguheba et al. (2000) found that P is released more rapidly from such organic residues than from triple superphosphate. Nitrogen content of the plant residues plays an important role in its transformations; Organic substances containing more than 1.8% N. upon decomposition, immediately increase the inorganic nitrogen level in the soil. Here mineralization rate exceeds the immobilization rate. A temporary removal of inorganic nitrogen follows the application of plant material with 1.2 to 1.3% N, but the initial period is followed by a stage in which mineralization exceeds

immobilization so that the ammonia or nitrate accumulate in soil (Christen, 1986). The significant results obtained from the incorporation of the Lantana compost show that it is better alternative to conserve the plant nutrients.

Compost is prepared in the month of August when the plant shows peak vegetative growth. At this time cultivated lands are occupied by crops and there is no possibility of applying the green matter directly to the soil. Production of compost thus appears to be the feasible alternative to utilize the abundant green biomass of Lantana at this time of the year. The compost production at this time by mixing animal dung and green biomass of Lantana serves as stable manure for the crops sown in the succeeding rabi season. Animal dung serves as a good inoculant material to promote microbial activity in compost (Bhardwaj, 1983). Also, *Azotobacter* inoculation helps to improve the nitrogen content of the compost thus resulting in good compost (Banta and Dev, 2009).

In the present study, grain and straw yields increased with increasing levels of nitrogen upto the level of 120kgN/ha in wheat. These observations are in conformation with the results obtained by Bhardwaj and Gaur (1985) and Kapur *et al*, (1986). The increase in growth attributes due to increasing levels of nitrogen application is quite obvious as at higher nitrogen levels the growth activity of the crop is likely to be greater.

The incorporation of Lantana as a green matter or compost increased the grain protein, similar results were earlier reported by Tisdale *et al.* (1995). Since, Lantana is rich in nitrogen and phosphorus was uniformly applied as per its recommend dose, it might have resulted in profuse and extensive root growth resulting in higher concentrations of grain protein.

The organic wastes originating from plant resources are subjected to intense state of biotransformations resulting in biodegradation of larger tissues and molecules to smaller ones, which later recombined to form complex humic molecules in the soil. Soil microorganisms play a dominant role in all these bio chemical processes not only as silent scavengers but also as eminent biochemists and biotechnologists.

The biodegradation process of the added organic wastes and residues is carried out by different groups of heterotrophic microorganisms, *viz*, bacteria, actinomycetes, fungi and protozoa. The role of cellulolytic and lignolytic microorganisms is of prime importance. Microorganisms involved in the process derived their energy and carbon from the decomposition of carbonaceous materials. Heterotrophs which utilize oxygen, decompose organic residues and assimilate some of carbon, nitrogen, phosphorus, sulphur and other nutrients for synthesis of their cell - protoplasm, resulting in the production of carbon dioxide, humic substances and release of available plant nutrients. Carbon serving as energy source and basic material for cell synthesis is assimilated in the greater amount than nitrogen. In general, for every ten parts of carbon one part of nitrogen is required for building up the cell protoplasm. About two-third of the carbon is respired/evolved as CO_2 and the remaining one-third is combined with nitrogen in the livings cells (Gaur *et al.*, 1984).

Further, the different groups of microflora respond differently to the type of organic residues. The fresh undecomposed carbonaceous material significantly alter their population as compared to the stable manures, bacteria being affected to a greater extend then fungi and actinomycetes. The bacterial population, in general, shows an early increase, fungi and actinomycetes dominating at mid and later stages of decomposition. Besides, the addition of easily decomposable material to the soil to stimulate the microorganisms that they not only attack the readily decomposable carbon of these materials but also decompose some relatively resistant native organic matter (Bhardwaj and Gaur, 1985; Bhardwaj and Patil, 1982).

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