INDO AMERICAN JOURNAL OF PHARMACEUTICAL RESEARCH

INFLUENCE OF *TRICHODERMA HARZIANUM T22* TO ENRICH THE MINERALS REQUIRES FOR PLANT GROWTH IN VERMICAST

M. Agnes Sharmila, M. Jegadeesan*

Department of Environmental and Herbal Science, Tamil University, Thanjavur, Tamilnadu.

ARTICLE INFO

Article history

Received 27/04/2017

Available online 08/04/2017

Keywords

Bagasse,

Trichoderma Harzianum T22,

AAS.

ABSTRACT

In several developing countries bagasse is one of the major source of bio fertilizer and animal feed production. About 25 million tonnes of bagasse are available every year but 60 –75 % of dry matter of bagasse acquires in the form cellulose and hemicelluloses and its digestibility is very poor because of the presence of lignin. It is regarded as a cheap substrate, collected at the site of processing and constant supply generated within the sugarcane industry. *Trichoderma harzianum T22* produce ligno cellulolytic enzyme which is used to break the indigestive compounds and help earthworm to easily digest food matters in the bagasse. In the current research three different groups each group had three different combinations of the composting beds with the size 3L×2B×1H were prepared. The first group had three different combinations of composting beds with bagasse and cow dung; then second group had three different combinations of composting beds with bagasse, cow dung and earthworm; finally third group had three different combinations of composting beds with bagasse, cow dung, earthworm and *Trichoderma harzianum T22* were constructed and the beds were maintained at optimum temperature and pH. The presences of the minerals were analysed using standard method and heavy metals were analysed using the Atomic absorption spectroscopy at regular interval 30th, 60th, and 90th day of composting respectively. Finally, the results revels that the vermicast from *Trichoderma harzianum T22* inoculated composed beds especially T3c have higher quantity of essential minerals with lesser heavy metals when compare to others. We concluded that the *Trichoderma harzianum T22* plays a major role to enrich the essential minerals required for various plants growth in the vermicast.

Corresponding author

M. Agnes Sharmila

Department of Environmental and Herbal Science,

Tamil university,

Thanjavur, Tamilnadu.

Please cite this article in press as ***M. Agnes Sharmila*** *et al. Influence of Trichoderma Harzianum T22 to Enrich the Minerals Requires for Plant Growth in Vermicast. Indo American Journal of Pharmaceutical Research.2017:7(04).*

INTRODUCTION

Bagasse is the fibrous residue of the sugar cane stalk coming from the mill after crushing and extraction of juice. This bagasse maintains 50-52% moisture at the mill outlet due to the imbibitions technique adopted to extract the juice from the cane and is generally known as 'mill wet bagasse'. The bagasse percentage on cane can vary from 23% to 37% (ICRA 2006). This depends on the fiber percentage cane, which normally ranges from 12 to 19%. The rest of the bagasse is made up of trapped dissolved matter, trash, and water. The properties of bagasse are outlined in Table 1.3, while Table 1.4 gives the proximate and ultimate analysis of bagasse (Hugot 1986).

Vermicomposting is a biotechnological process in which earthworms are employed to convert the organic wastes into humus like material known as vermicompost. Certain earthworm species are capable of consuming a wide range of organic wastes from sewage sludge, animal wastes, agricultural residues, domestic wastes, to industrial wastes. Under favourable conditions of temperature and moisture, earthworms maintain the aerobic conditions in the vermicomposting process ingest organic waste materials and egest a humus-like substance which is more homogeneous than the organic wastes or raw materials used (Arancon et al. 2003; Edwards and Burrows 1988). The actions of the earthworms in this process are both physical and biochemical. The physical actions include fragmentation, turnover and aeration. Whereas biochemical actions include enzymatic digestion, nitrogen enrichment, transport of inorganic and organic materials (Edwards and Lofty 1972). During this process, important plant nutrients such as nitrogen, potassium, phosphorus and calcium present in the waste materials are converted through microbial action into such chemical forms which are much more soluble and available to the plants than those in the parent substrate (Ndegwa and Thompson 2001).

Trichoderma spp. long has been known for their abilities to control plant-pathogenic fungi. Mechanisms primarily have included direct effects upon target fungi via competition, mycoparasitism, and antibiosis. In addition, these fungi have been shown to directly increase plant growth by unknown mechanisms, even under axenic conditions. They also have abilities to solubilise plant nutrients and to increase plant nutrient uptake. Among the most useful Trichoderma strains are those that are rhizosphere competent (i.e., able to colonize and grow with root systems of most plants). T. harzianum strain T22, when applied as a seed or soil treatment, colonizes the entire root system of most plants and increases the depth of rooting of maize even when plants are fully mature. This effect occurs across many soil types and pH levels and results in a variety of beneficial effects, including resistance to abiotic stresses such as drought and improved nitrogen fertilizer use efficiency. It also protects plants from a variety of pathogens when the organism is applied as a foliar, soil, or root protectant. However, even though the organism is becoming widely used in commercial agriculture, knowledge of its multiple modes of action is sketchy at best.

MATERIALS AND METHODS

The chemicals were used from sigma (analytical grade), Hi-media and completely sterilized glassware were used in every step.

Strain collection:

The *Trichoderma harzianum T22* strain was collected from department of marine science, bharadidhasan university, Trichy.

Source collection:

In this present study the sugarcane waste (bagasse) will be collected from the sugarcane industry in Thanjavur. E*isenia fetida* were collected from Periyar Maniammai University, vallam, thanjavur, tamilnadu, India.

Composting methods:

All the composting beds were constructed as open bed method have 10Kg of quantity with the 3 feet length, 2 feet breadth, and 1 feet height.

Control bed:

The control bed (C) is constructed by using only 10 Kg bagasse.

First Composting bed (T1):

The T1 is constructed by using bagasse (substrate) and cow dung. The first bed is split into three beds for the depth analysis of which ratio is best for the composting. T1a contains 2.5 Kg of substrate and 7.5 Kg of cow dung, T1b contains 5 Kg of substrate and 5 Kg of cowdung, and T1c contains 7.5 Kg of substrate and 2.5 Kg of cow dung.

Second Composting bed (T2):

The T2 is constructed by using bagasse (substrate), cow dung, and Earthworm. The second bed is split into three beds for the depth analysis of which ratio is best for the composting. T2 a contains 2.5 Kg of substrate, 7.5 Kg of cow dung and 100gm of Earthworm, T2b contains 5 Kg of substrate, 5 Kg of cow dung and 100gm of Earthworm, and T2c contains 7.5 Kg of substrate, 2.5 Kg of cow dung and 100gm of Earthworm.

Third Composting bed (T3):

The T3 is constructed by using bagasse (substrate), cow dung, Earthworm and . The Third bed is split into three beds for the depth analysis of which ratio is best for the composting. T3a contains 2.5 Kg of substrate, 7.5 Kg of cow dung, 100gm of Earthworm and 1.2ml of *Trichoderma harzianum* T 22, T3b contains 5 Kg of substrate, 5 Kg of cow dung, 100gm of Earthworm and 1.2ml of *Trichoderma harzianum* T 22, and T3c contains 7.5 Kg of substrate, 2.5 Kg of cow dung, 100gm of Earthworm and 1.2ml of *Trichoderma harzianum* T 22.

Incubation Period:

All the beds were allow incubating for 3 months to find out the composting level and complete observation was made for the 3 months incubation period. All the important parameters like pH, moisture etc., should be maintained in all the beds for the entire incubation period. All beds were free from all other contaminants and put all safety measures like steel fencing were carried out for the period of incubation, this is for avoiding the animals, birds etc., were disturbing the composting beds.

Physico-chemical characteristics:

The quality of the compost will be characterized by characteristics like temperature, pH, electrical conductivity, alkalinity, etc., along with elemental analysis. Samples were collected at 10 days intervals, randomly in 5 different places of the pile, at 1ft depth. Collected samples were mixed together and dried by exposing to sunlight followed by hot air oven at 105º C. Dried samples were crushed well and sieved through 0.2 mm mesh. The powdered samples were then used for further physico-chemical and spectral characterization Temperature (°C) was noted during the collection of compost samples at each 10 days interval. Moisture content (%) was estimated by gravimetric method. pH and Electric conductivity was measured for the 1:5 compost: water suspensions. Organic carbon (%) and Organic matter (%) were estimated using Walkey and Black method. Water holding capacity (%) was measured using filtration method. Phosphorus (ppm) was determined through Vanadomolybdate method. Carbonates, bicarbonates and alkalinity (mg CaCO3/L) were estimated by titration of compost: water (1:5) extract with standard H2SO4 solution using Methyl orange and Phenolphthalein as indicators. Calcium and Magnesium were estimated by titration of compost: ammonium acetate (1:5) extract with standard EDTA solution using Eriochrome Black-T and Patton-Reeder's indicators. Brucine method was used for the estimation of nitrate-nitrogen, sodium and potassium concentrations were determined using Flame photometer. Humification Index was estimated through UV-Visible spectrophotometry. For estimation of heavy metals like lead, manganese, zinc, copper and iron etc, triacid (nitric acid: sulphuric acid: perchloric acid in 9:2:1 ratio) extract of the compost sample were used for Atomic Absorption Spectrophotometry

Analysis:

The samples were taken in all the composting beds and analyzed periodically at 30th day, 60th day and 90th day respectively. The samples were subjected to analyze for the presence of heavy metals named cadmium, copper, lead, and zinc using Atomic Absorption Spectroscopy.

Atomic absorption spectroscopy (AAS):

Using AAS the presence of heavy metals in the composts were analyzed. Here the AAS used were shimadzu AA - 7000 model and the air used were acetylene and the temperature around 2000ºc.

RESULT AND DISCUSSION

Table: 1: Physico-chemical characteristics.

S.NO	Parameters		T1a	T1b	T1c	T2a	T2b	T2c	T3a	T3b	T3c
1.	Total carbon %	30 d	30.4	23.0	39	36.5	26	34.6	35.3	37.2	48.3
		60 d	32	25.2	47.2	42.2	28.2	39.0	40.3	43.4	51.2
		90 d	35	28	52.0	48.0	34.4	45.4	48.5	50.2	55.0
2.	Total nitrogen %	30 d	0.11	0.32	0.13	0.21	0.4	0.32	0.35	0.40	0.54
		60 d	0.12	0.36	0.18	0.30	0.45	0.43	0.49	0.53	0.61
		90 d	0.15	0.40	0.22	0.32	0.5	0.46	0.50	0.51	0.82
3.		30 d	15	20	22	12	15	13	11	13	24
	C/N ratio	60 d	15.4	18	15	9	16	11	14	17	25
		90 d	15.8	15	13	7	18	8	15	18	27
4.		30 d	1.8	2.0	1.9	2.0	1.8	2.3	1	0.9	0.7
	Phosphate	60 d	1.5	1.8	1.4	1.5	1.4	2.0	0.9	0.7	0.6
		90 d	1	1.5	1.3	1.3	1.2	1.9	0.7	0.7	0.5
5.		30 d	0.43	0.34	0.46	0.32	0.30	0.23	0.20	0.15	0.15
	Potassium	60 d	0.25	0.30	0.41	0.26	0.32	0.20	0.18	0.15	0.13
		90 d	0.5	0.26	0.35	0.20	0.30	0.18	0.15	0.14	0.1

Table: 2: AAS Results on 30th day.

Sample ID	Cd	Cu	Pb	Zn	Unit
T1a	Nil	70.3182	Nil	54.3421	ppm
T1b	Nil	75.4523	Nil	54.0435	ppm
T1c	Nil	62.9116	Nil	49.9231	ppm
T2a	Nil	65.5603	Nil	41.2666	ppm
T2b	Nil	60.3027	Nil	44.0349	ppm
T2c	Nil	65.3956	Nil	42.5412	ppm
T3a	Nil	52.4254	Nil	42.8554	ppm
T3b	Nil	42.0457	Nil	40.0818	ppm
T3c	Nil	37.4677	Nil	39.7317	ppm

Table: 3: AAS Results on 60th day.

Sample ID	Cd	Cu	Pb	Zn	Unit
T1a	Nil	52.3181	Nil	35.6311	ppm
T1b	Nil	46.0257	Nil	37.7117	ppm
T1c	Nil	38.4667	Nil	41.0815	ppm
T2a	Nil	55.3856	Nil	43.2546	ppm
T2b	Nil	50.2027	Nil	45.0321	ppm
T2c	Nil	44.4533	Nil	50.3415	ppm
T3a	Nil	50.4554	Nil	41.3321	ppm
T3b	Nil	41.2176	Nil	40.0235	ppm
T3c	Nil	36.6603	Nil	32.6554	ppm

Table: 4: AAS Results on 90th day.

Sample ID	Cd	Cu	Pb	Zn	Unit
T1a	Nil	34.4617	Nil	32.4530	ppm
T1b	Nil	31.1257	Nil	26.7127	ppm
T1c	Nil	30.3677	Nil	35.0180	ppm
T2a	Nil	42.5408	Nil	40.2604	ppm
T2b	Nil	50.3022	Nil	42.0913	ppm
T2c	Nil	31.3696	Nil	25.5104	ppm
T3a	Nil	45.3174	Nil	34.3241	ppm
T3b	Nil	36.2523	Nil	32.1230	ppm
T3c	Nil	28.9011	Nil	31.5231	ppm

CONCLUSION

Finally, the results revels that the vermicast had higher quality of minerals when compare to others. In addition to that the vermicast from *Trichoderma harzianum T22* inoculated composed bed T3c had maximum quantity of minerals with lesser heavy metals when compare to others. This result confirms that baggase is very suitable substrate for vermicomposting and *Trichoderma harzianum T22* plays a major role to enrich the essential minerals required for various plants growth in the vermicast.

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54878478451**170551**