Bioconversion of Shrimp Shell Waste into Compost Preparation and its Plant Growth Study

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ABSTRACT

Background: The fishing sector plays an important role in the global economy. The development of this sector is related to environmental issues, in particular to waste management. Indeed, the quantity of the halieutic waste are considered at several thousand tons of waste a year. Waste management is a solution to maintain an area with ecological harmony, but still can produce economic benefits that are beneficial to social welfare. Chitinases are enzymes that degrade chitin. Chitinase contribute to the generation of carbon and nitrogen in the ecosystem from shrimp shell wastes. Chitin and chitosan are naturally-occurring compounds that have potential to be used in agriculture. The present study was designed to prepare the shrimp shell waste compost (SSC) and also to analyse the enhancement growth of Ladies finger plant by using shrimp shell waste compost (SSC), shrimp shell waste compost (SSC) +cow dung (CD) and along with coir pith (CP).

Methods: This experiment was conducted within the period of 2017-2018 in Department of Microbiology, Kamaraj college, Thoothukudi. 1Kg of dried shrimp shell waste powder was used for the compost preparation using chitinolytic bacteria *Bacillus licheniformis* SSCL10. The present study shrimp shell wastes were composted into manure with adding chitinolytic *Bacillus licheniformis* SSCL10. After 90 days of composting, the compost was dried at room temperature and used as manure for plant study. The growth study of ladies finger (*Abelmoschus esculents* L.) was analyzed in shrimp shell compost and also along with other composts (SSC + CD and SSC + CD+CP).

Result: In our study, nitrogen, phosphorus and potash content were increased in the shrimp shell composts (SSC) with adding chitinase producing bacterium (*Bacillus licheniformis* SSCL10) when compared with control soil. The maximum plant growth performances were showed in the SSC+CD (cow dung) compared to combination of CP (coir pith), shrimp shell composts (SSC) alone and control. So shrimp shell compost is used as supplements for other compost to induce plant growth performances.

Key words: Bacillus licheniformis, Bio-fertilizer and agriculture, Chitin, Coirpith, Cow dung, Shrimp shell waste.

INTRODUCTION

Farmers face the problem of climate change and the limitation of arable land suitable for growing crops. The global population is expected to increase by 40% over the next 40 vears with global agricultural crops struggling to keep pace. Agricultural sector is facing an intimidating challenge to feed this rapid burst in population by applying chemical fertilizers. Applications of chemical fertilizers have robbed the soil fertility, decline in biodiversity, nutrient and pesticide runoff and have resulted in health and environmental hazards (Boussemart et al., 2013). Hence, the alternative way is to reduce the use of inorganic fertilizers by recycling the organic wastes as fertilizers. This will cover the way for sustainable solid waste management and agriculture (Balraj et al., 2014). Generally, Soil is deficient in all necessary plant nutrients, but large quantities of such nutrients contained in agricultural by-products and domestic wastes are wasted. In nature, a number of organisms have the ability to convert organic waste into valuable compost (Vennila et al., 2012). The compost preparation is a new method for the complete utilization of this waste, which also serves as a means of raw verging, recycling nutrients and also keep away from disposal problem and following environmental consequences (Amar et al., 2006).

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The shrimp processing industry produces over millions tons of waste shell. Peeling process, which involves the removal of the shell from the tail of prawn, increases the total waste production up to 40-45% (Ravi Kumar, 2000). This plentiful waste may create environmental hazard due to the easy deterioration (Mejia-Saules *et al.*,2006). A wide range of microorganisms have the ability to degrade chitin by producing chitinases for nutrition, antagonism and combating parasites (Faramarzi *et al.*, 2009). Bacteria employ several proteins, including chitin-binding proteins, to degrade chitin, but the hydrolysis by chitinase is the key step in the solubilization and mineralization of chitin (Anuradha and Revathi, 2013).

The biotransformation of waste seems to be the most suitable to resorb these problems. It corresponds to the elaboration of beneficial products, of natural origin, usable as bio-fertilizing for grounds (Taiek et al., 2014) in substitution of artificial fertilizers. Moreover, excessive application of artificial fertilizers is one of the most important agricultural processes in the world, it cause volatilization of ammonia in the air, pollution of water resources causing their eutrophication, degradation of ground water by their pollution (Xu et al., 2008). Hu et al. (2008) reported that microorganisms alter organic waste materials into compost by breaking down to simple compounds and reforming them into new complex compounds during the composting process. This process is cost effective due to the abundance of soil microbes done the function (Krishnaveni and Ragunathan, 2014). These organisms utilize the complex nutrients of the compost and liberate the essential minerals and elements into soil which in turn offers nutrients to the crops and improves their yield (Kalpana et al., 2011). Fish manure contains NPK and micronutrients that are necessary for plant growth. Chitinases are considered to be useful in management of converting chitinous wastes into fertilizers generated by sea food manufacturing industries (Sakai et al., 1998). The increase in the human consumption of fish results in the production of waste which can be reutilized into ecofriendly compost preparation. A shift from chemical fertilizers recycled organic waste will represent a sustainable method of agriculture. Hence, the present study is to evaluate the potential effects of shrimp shell waste compost on the growth characteristics of Ladies finger (Abelmoschus esculentus L.).

MATERIALS AND METHODS

This study was conducted in Kamaraj College, Thoothukudi, Tamilnadu for nutrient content analysis of Shrimp shell wastes for bioactivator development, nutrient content analysis and plant composts application on Ladies finger (*Abelmoschus esculentus* L.) plant.

Microorganisms

Chitinolytic *Bacillus licheniformis* SSCL10 was isolated from our previous study (Abirami *et al.*, 2016), used for compost preparation study. Chitinolytic properties of the selected strain were assessed periodically by checking zone formation on colloidal chitin agar plates.

Shrimp shell waste

Raw shrimp shells were collected from shrimp processing plants in Thoothukudi, Tamil Nadu, India. The wastes were thoroughly washed with tap water to remove its impurities. The whole parts of shrimp shell including the head and legs were used in the experiment. The shrimp shells were dried to remove the water content. They were then milled using mechanical grinder into powder form. The powder was then placed in the sealed plastic bottle and kept in refrigerator. 1Kg of powder was used for the compost preparation. The soil and shrimp shell wastes were sterilized by the process of Tyndalization for three days. Sterile sandy (earthen) pot was used for the study.

Experimental study for compost preparation

In this experimental setup, first layer of about 5cm was filled with sterile soil from the base of the pot, after that 250gm of sterile powdered prawn shell was layered on the sand and 5ml (10x10⁸ cfu/ml) of chitinolytic *Bacillus licheniformis* (SSCL10) inoculums was added. The same was repeated to form many layers. Finally, pot was loosely covered with sterile cloth to minimize evaporation and prevent attack of insects and others. DDT was applied around the pots to prevent the attack of ants. The pot soil was stirred and moisture was added on weekly basis to bring the proper mixing of soil and compost, to enhance the decomposition.

Periodically sterile water was poured into the pot to maintain the moisture of the compost. pH and temperature was checked periodically. The pot was maintained at 40°C for 90 days, during which regular mixing was performed (every weeks). Then the compost was dried at room temperature and used as manure for plant study. The Control (without inoculation) was also maintained. The experiment was done repeatedly. A shrimp shell-based compost should heat up quickly (within 36 hours) and can easily reach temperatures of 50°C and higher. The periodic stirring will add air and keep the composting going on and generally, for on-farm use, any material which has composted for eight weeks will be sufficiently (Spargo, 2004).

Analysis of shrimp shell compost

The shrimp shell compost was harvested and placed in shade, allowed to air dry for chemical analysis together with untreated soil and using standard procedure for pH, EC, Total Nitrogen, available Phosphates and Potash. The analysis of the compost soil and control soil was done at SPIC, Thoothukudi, Tamil Nadu, India.

Growth of crops using shrimp shell waste compost

(Abelmoschus esculentus L.) known as lady's finger is a heat loving plant of Malvaceae family and is one of the most important summer vegetable in India. In the pot study experiment of Ladies finger fine sand was sieved with a 2 mm wire gauzed metallic tray, in order to remove stones, plant debris and generally unwanted materials that could hinder plant growth. After that, 500 g of Shrimp shell compost was weighed and mixed with different forms of compost. Each type of Shrimp shell compost was properly mixed with 1 kg of garden soil in the ratio of 1:1 except control (1 kg garden soil). Each mixture was packed into sand pot and adequately watered as reported by lwase et al. (2000). Twenty five seeds of lady's finger were properly placed in every mixture and control pots. The pots were maintained under environmental conditions that persist in the home garden and were irrigated with well water uniformly throughout the study period of experiment. Only the required

quantity of water alone was poured into each pot in order to avoid draining of excess water from the pots that can result in elimination of nutrients. The growth and growing stages of the plants were carefully monitored and the morphometric data were also taken at every week (upto 4 weeks). For each data entry, three plants were randomly collected from each treatments and the observation of exo-morphological characters was done as reported by Rekha *et al.* (2013). The treated groups were maintained in triplicates with a separate control (Jonathan *et al.*, 2013). The observation was made up to 30 days and only growth parameters were monitored.

Observations

In the compost prepared from shrimp shell waste, the nutrient content analysis was carried out. Every week, observations on plants shoot length, root length, internodes length, leaf width, wet and dry weight of the plant leaves were also carried out. The chlorophyll content in Ladies finger leaves were estimated by the Bansal *et al.* (1999) method.

Statistical analysis

The results obtained in the present investigation were analysed statistically using standard deviation (SD) and Duncan's Multiple Range Test (DMRT) (Zar, 1984). The data were compiled using statistical package for social science program (SPSS 16th version) and subjected to analysis of variance (ANOVA) with post hoc Duncan's test comparison (Duncan, 1955) (one-way). P< 0.05 was considered as significance.

RESULTS AND DISCUSSION

Intensive application of chemical fertilizers in agriculture has caused harm to the ecological state of the agricultural systems (Khan et al., 2018). in the present study, nitrogen, phosphorus and Potash content of the composts were increased on adding chitinase producing bacterium (Bacillus licheniformis SSCL10) when compared with control soil (Table 1). The C:N ratio was an indication of litter degradation of soil and indicator of efficient and effective composting. The final value was important to determine the value of the completed compost as a soil improvement for growing crops (Ansari and Jaikishun, 2010). Carbon provides the primary energy source and nitrogen which is critical for microbial growth. As the composting process proceeds, the carbon content decreased because of its conversion to carbon dioxide while nitrogen was kept intact. Very high C:N ratio (above 80) greatly reduced the rate of natural composting (Stoffella and Kahn, 2000). Composting shrimp waste has also become more common in coastal areas. The fresh waste is usually mixed in a 1:2 ratio with woodchips or other high carbon by-products and turned daily, while composting for 60 to 90 days. The compost is then added as a soil amendment to tillable soil (Cato, 1992). The present study shrimp shell wastes were composted into manure by adding chitinolytic *Bacillus licheniformis* SSCL10.

Bell, (2000) reported that chitin-containing compost reduced populations of parasitic nematodes. Partly degraded chitin is recognized as an elicitor of plant defence mechanisms and a growth suppressor of pathogenic fungi. The shrimp waste-based composts have significantly high levels of disease suppressive chitin derivatives than commercially available composts (Roy *et al.*, 2013).

The shrimp shell was completely composted to form good manure after 90 days. During composting, pH and temperature were analyzed every week. In the first week of the compost, pH value slightly increased upto pH 8.0 After then, pH value slowly decreased (pH value was 6.8). During the starting of the composting process, proteolytic and mineralisation process released ions and ammonia that increases the pH value. After then (third and fourth week), pH value slowly increased upto 8.2. But the chitin of the shrimp shell undergoes decomposition that decreases the pH value. Similarly soil pH significantly increased for all the vermicompost (Ansari and Jaikishun, 2010).

During composting process, the temperature of the compost increases upto 50°C, but most of the time, compost was maintained at 40°C (Table 2). Baruah and Sarma (2013) reported that vegetable crop demand has been increasing day by day. In our work, one of the vegetable crop (Abelmoschus esculents L.) plant was selected for the compost growth study. Ladies finger growth study of different compost were analyzed for shoot length (cm), root length (cm), length of internodes (cm), leaves width (cm), leaves fresh weight (g), dry weight (g) and chlorophyll content (mg/ g) were measured over the period of four weeks. Chitin and chitosan are naturally-occurring compounds that have potential in agriculture with regard to controlling plant diseases. These molecules were shown to inhibit fungal growth and development. They were reported to be active against viruses, bacteria and other pests. In addition to most reviewers mention, release of nematicidal compounds from decomposing materials such as chitin and chitin derivatives stimulated natural enemies of nematodes and improved plant growth and tolerance to nematodes (Thoden et al., 2011).

Sharma and Mittra (1991) reported that the use of organic manure (cow dung) is advocated for maintaining the soil fertility over a long period of time. The nutrients contained in organic manures are released more slowly and

Table 1: Analysis of macronutrients in soil treated with manures.

Compost Sample	рН	E.C	Nitrogen (N)	Phosphorous (P)	Pottash (K)
Control	8.4	0.36	105	4.3	135
SSC	8.2	0.66	102	13.1	450
SSC + CD	8	0.73	106	12.2	500
SSC + CD+CP	8	0.83	104	13.4	460

Table 2: Analysis	of pH	and	Temperature	variation	in	compost
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maturation		
Week	рН	Temperature
First	8	40°C
Second	6.8	50°C
Third	8.6	45°C
Fourth	8.2	40°C

are stored for a longer time in the soil, thereby ensuring a long residual effect. In the present work, shrimp shell compost was mixed with cow dung has shown considerable increase in plant growth. Microbial consortium along with organic manure improves the physical, chemical and biological properties of the soil. Vipitha and Geethakumari (2016) reported that growth and productivity of Amaranthus was influenced by the performance of inoculation of microbial

Table 3: Measurement of shoot length (cm) in lady's finger over the period of four week.

Characters	I Week	II Week	III Week	IV Week
Control	3.33 ± 0.18 ^d	5.57 ± 0.34 °	8.46 ± 0.49 ^b	11.43 ± 0.61 ª
SSC	5.08 ± 0.22 d	9.42 ± 0.33 °	12.80 ± 0.80 ^b	16.10 ± 0.76 ª
SSC+CD	5.79 ± 0.54 ^d	10.16 ± 0.31 °	13.98 ± 0.67 ^b	16.66 ± 0.98 ^a
SSC+CD+CP	5.09 ± 0.21 ^d	8.45 ± 0.67 °	13.82 ± 0.82 ^b	15.43 ± 0.55 °

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

Table 4: Measurement of root length (cm) in lady's finger over the period of four week.

I Week	II Week	III Week	IV Week
2.23 ± 0.32 °	2.55 ± 0.23 b	2.78 ± 0.33 b	3.46 ± 0.15 ª
3.71 ± 0.26 °	4.50 ± 0.15 ^b	4.68 ± 0.24 ^b	5.54 ± 0.31 ª
4.20 ± 0.15 °	5.22 ± 0.32 ^b	5.55 ± 0.42 ^b	5.83 ± 0.32 ª
2.93 ± 0.23 °	4.25 ± 0.29 b	4.59 ± 0.28 ^b	4.84 ± 0.35 °
	2.23 ± 0.32 ° 3.71 ± 0.26 ° 4.20 ± 0.15 °	$2.23 \pm 0.32^{\circ}$ $2.55 \pm 0.23^{\circ}$ $3.71 \pm 0.26^{\circ}$ $4.50 \pm 0.15^{\circ}$ $4.20 \pm 0.15^{\circ}$ $5.22 \pm 0.32^{\circ}$	$2.23 \pm 0.32^{\circ}$ $2.55 \pm 0.23^{\circ}$ $2.78 \pm 0.33^{\circ}$ $3.71 \pm 0.26^{\circ}$ $4.50 \pm 0.15^{\circ}$ $4.68 \pm 0.24^{\circ}$ $4.20 \pm 0.15^{\circ}$ $5.22 \pm 0.32^{\circ}$ $5.55 \pm 0.42^{\circ}$

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

Table 5: Measurement of leaf width (cm) in lady's finger over the period of four week.

Characters	I Week	II Week	III Week	IV Week
Control	5.1±0.20 °	6.0± 0.09 b	7.2± 0.09 ^b	8.5±0.25 ª
SSC	6.2±0.20 °	7.3 ± 0.02 ^b	9.4 ± 0.03 ^b	10.61 ± 0.03 ª
SSC+CD	5.96 ± 0.31 °	7.50 ± 0.29 ^b	7.43 ± 0.15 ^b	7.76 ± 0.15 ª
SSC+CD+CP	5.07 ± 0.30 °	6.03 ± 0.26 ^b	6.15 ± 0.14 ^b	6.70 ± 0.42 ª

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

Table 6: Measurement of internodes length (cm) in lady's finger over the period of four week.

Characters	I Week	II Week	III Week	IV Week
Control	2.12 ± 0.20 °	3.12 ± 0.24 b	3.30 ± 0.21 ^b	3.91 ± 0.19 °
SSC	3.02 ± 0.20 °	5.21 ± 0.25 b	5.24 ± 0.18 ^b	5.58 ± 0.13 ª
SSC+CD	5.96 ± 0.30 °	7.50 ± 0.26 ^b	7.43 ± 0.14 ^b	7.76 ± 0.42 ^a
SSC+CD+CP	5.07 ± 0.30 °	6.03 ± 0.26 ^b	6.15 ± 0.14 ^b	6.70 ± 0.42 ^a

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

Table 7: Measurement of plant leaf dry weight (g) in lady's finger over the period of four week.

I Week	II Week	III Week	IV Week
0.03 ± 0.01 °	0.06 ± 0.01 ^b	0.08 ± 0.02 b	0.17 ± 0.07 ª
0.45 ± 0.02 °	0.58 ± 0.03 b	0.61 ± 0.04 ^a	0.65 ± 0.06 a
1.31 ± 0.09 °	1.97 ± 0.04 ^b	2.05 ± 0.06 b	2.32 ± 0.11 ª
0.96 ± 0.07 °	1.39 ± 0.09 ^b	1.48 ± 0.10 ^b	1.70 ± 0.11 ª
	0.03 ± 0.01 ° 0.45 ± 0.02 ° 1.31 ± 0.09 °	$0.03 \pm 0.01^{\circ}$ $0.06 \pm 0.01^{\circ}$ $0.45 \pm 0.02^{\circ}$ $0.58 \pm 0.03^{\circ}$ $1.31 \pm 0.09^{\circ}$ $1.97 \pm 0.04^{\circ}$	$0.03 \pm 0.01^{\circ}$ $0.06 \pm 0.01^{\circ}$ $0.08 \pm 0.02^{\circ}$ $0.45 \pm 0.02^{\circ}$ $0.58 \pm 0.03^{\circ}$ $0.61 \pm 0.04^{\circ}$ $1.31 \pm 0.09^{\circ}$ $1.97 \pm 0.04^{\circ}$ $2.05 \pm 0.06^{\circ}$

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

Table 8: Measurement of plant leaf fresh weight (g) in lady's finger over the period of four week.

Characters	I Week	II Week	III Week	IV Week
Control	0.19 ± 0.02 °	1.08 ± 0.08 ^b	1.24 ± 0.09 ^b	1.54 ± 0.10 ª
SSC	1.37 ± 0.27 °	1.49 ± 0.14 ^b	1.83 ± 0.28 ^b	2.29 ± 0.20 ª
SSC+CD	2.17 ± 0.08 °	2.95 ± 0.18 ^b	3.02 ± 0.20 ^b	3.57 ± 0.16^{a}
SSC+CD+CP	1.41 ± 0.12 °	2.37 ± 0.18 ^b	2.51 ± 0.22 ^b	3.10 ± 0.21 ª

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

Table 3. Measurement of children with content (mg/g) in lady 3 miger over the period of roat week.				
Characters	I Week	II Week	III Week	IV Week
Control	0.13 ± 0.03 c	1.09 ± 0.01 b	1.47 ± 0.06 b	2.02 ± 0.01 a
SSC	1.16 ± 0.06 c	2.01 ± 0.01 b	2.3 ± 0.1 b	3.01 ± 0.01 a
SSC+CD	1.17 ± 0.06 c	2.05 ± 0.01 b	2.47 ± 0.06 b	3.07 ± 0.01 a
SSC+CD+CP	1.07 ± 0.06 c	2.02 ± 0.0 b	2.17 ± 0.06 b	2.93 ± 0.06 a

Table 9: Measurement of chlorophyll content(mg/g) in lady's finger over the period of four week

values are expressed in mean ± SE dissimilar alphabets in vertical column are significantly different at P< 0.05% level.

consortium along with bio-organic composite manure such as coir pith compost, poultry manure, neem cake, groundnut cake, ash, rock dust.

The result indicated that the maximum shoot length, root length, internodes length, fresh weight of leaves, dry weight of leaves and chlorophyll content were achieved in shrimp shell compost +cow dung followed by shrimp shell compost +cow dung + coir pith, shrimp shell compost and control. When compared with different compost, shrimp shell compost +cow dung was the best growth medium than that of shrimp shell compost alone. Shrimp shell compost induced the shoot length, root length, internodes length, fresh weight of leaves, dry weight of leaves and chlorophyll content significantly than compared to others. So it is used as supplements for other compost.

CONCLUSION

Chitin and chitosan are naturally-occurring compounds that have potential use in agriculture with regard to controlling plant diseases. Extensive usage of chemical fertilizers and pesticides caused number of harmful effects to the farmers. The poor farm management technique and improper use of agro-chemicals has also resulted in both soil quality and environmental degradation. The use of non-chemical fertilizers and pesticides is one of the common practices that have been introduced with alternative agricultural systems. The microorganisms and the nutrients present in the raw materials are very helpful in improving soil health. Shrimp shell wastes are rich sources of chitin and protein. Using the bacteria, Shrimp shell wastes was composed into manure. Composting is a simple, eco friendly safe and inexpensive alternative method of disposal of shrimp shell wastes. This compost can be used as a supplemental fertilizer for various crops.

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