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An effective biodegradation of organic solid waste by selective bacterial consortium

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ABSTRACT

Municipal solid waste is the very big menace for the environment. Due to rapid increase of human population, urbanization and the industrialization leads to the solid waste accumulation. Solid waste is usually managed through land filling and incineration processes. But both of these methods cause environmental pollution. Hence researchers are seeking alternate option to mitigate the solid waste pollution. In view of the above issues, the present study is focused to control and manage the municipal organic solid waste through biological way. In this present study the bacterial strains were isolated from the municipal waste land filling sites and the effective strains are identified and used to prepare selective bacterial consortium. The consortium with selective bacterial species were activated with two different sugar industry materials namely; (i) jaggery and (ii) molasses. The bacterial consortium activated with two different sources were sprayed on municipal organic waste at different quantity. After the sufficient time interval the biodegraded materials were harvested and subjected to physico chemical analysis. The result revealed that the municipal organic waste degraded by selective bacterial consortium (SBC) activated with molasses have high nutritive value and it degraded the waste effectively when compare to SBC activated with jaggery. The biodegraded waste subjected to seed germination assay to find out the phyto toxic effect of the biodegraded materials. The findings of phyto toxic assay proved that the SBC activated with molasses was effective one to biodegrade the organic waste quickly and efficiently and there was no phyto toxicity in that product. Hence we could recommend this product as an organic supplement to enhance the plant growth.

Keywords *– Municipal Solid Waste, biodegradation, bacterial consortia*

1. INTRODUCTION

Municipal solid waste (MSW) is the abridgment of the waste generated from domestic, commercial, and construction activities. Waste is classified as liquid waste, solid waste and toxic wate. It has become a major consequence of modernization and economic development (Tsiboe, *et al.*, 2004). Notably, the solid form of waste is fast becoming a menace in both developed and developing nations (Contreras, *et al.*, 2009). Exponential growth of population and urbanization, and the development of social economy, coupled with the improvement of living standard, have resulted in an increase in the amount of MSW generation throughout the world. It is estimated that, on the average developed countries generates 521.95–759.2 kg per person per year (kpc) and 109.5–525.6 kg per person per year (kpc) typically by developing countries. Recent estimates suggest that the MSW generation exceeds 2 billion tons per year around globally which is major potential threat to the environment (Musademba, *et al.*, 2011). The Municipal solid waste is a matter of serious concern especially in urban areas due to the improper disposal plans and facilities. The management of domestic solid waste is one of the huge challenges of the urban areas of all sizes. MSWM system involves four activities such as waste generation, collection, transportation, and disposal. (Bundela, *et al.*, 2010). These four activities of Municipal solid waste in most of the semi urban municipality remains at a very nascent stage. Improper disposal of Municipal solid waste by residents along with the improper coordination of collection and non-periodic removal of waste leads to accumulation of MSW. It occurs mostly at street corners, temples, hospitals and marriage halls (Cai, *et al.*, 2007). The part of waste disposal such as metals, plastic, coils, batteries, etc., of the municipal waste is picked up by the rag pickers and also by the passers which is disposed for money at waste disposal shops. The non-scientific disposal of waste causes an adverse impact on all components of the environment and in the human health (Rathi, 2006). Unregulated dumping and open burning are still considered as the ultimate waste disposal methods. Therefore, MSW management (MSWM) seems to be one of the key topics for environmental protection in present days and also in the future (Tanmoy Karak, Bhagat & Pradip Bhattacharyya, 2012; Nethaji Mariappan, Parthiban & Pynthamil Selvi 2018). The land filling and burning of MSW can cause environmental pollution. Hence the alternate method should be practiced to manage the MSW. In view of that the present study is focused to adopt biological method to dispose the MSW.

2. MATERIALS AND METHODS

2.1. Collection of soil samples

In the present study, soil samples were collected from two different dump yards in and around Thanjavur. The soil samples were collected from four locations namely Sreenivasapuram MSW disposal site (site - A) and Punnainallur MSW disposal site (site - B). Soil samples were collected by profile sampling at a depth of 30 cm using a low cost 18 x 1 inch core sampler. (APHA 1998).

2.2. Sample processing and analysis

Soil samples were brought to the laboratory and sieved. The sieved soil that has been stored in a labeled polythene bags and used for analysis of various physicochemical and microbial parameters. The Physicochemical parameters of the soil samples were analyzed using the standard analytical procedure of Attoe (1947).

2.3. Enumeration and isolation of bacteria

The microorganisms were isolated by serial dilution technique on Nutrient Agar Media (NAM). In this technique, a sample suspension was prepared by adding 1.0 g sample to 10 ml distilled water and mixed well for 15 min and vortexed and was serially diluted in sterilized distilled water to get a concentration 10^{-3} (Cappuccino & Sherman, 1999).

2.4. Identification of bacterial strains

The identification of soil bacterial strain was carried out based on morphological characteristics and biochemical tests (Bergey & Holt, 2000).

2.5. Development of Selective Bacterial Consortium (SBC)

The isolation was followed by identification and screening of bacterial species. The effective isolates of various bacterial species were mixed in different proportions to prepare bacterial consortia. The compatibility of the bacterial strains within the consortium was checked regularly using gram staining technique.

2.6. Activation of SBC

The consortia were activated within seven days by two different supplementary materials such as jaggery and molasses. The mixtures were prepared in the proportions of 6 litres pure water, 3 kg jaggery / molasses and 1litre of SBC properly. Then, the mixtures were then poured into a clean plastic containers and sealed airtight, so that little air is left in the container. The containers were kept under shade at an ambient temperature of 24–26°C for 7-10 days. After wards a white layer on the top of the solution accompanied with a sweetish sour, rather pleasant smell was observed which a characteristic feature of the efficient SBC. The products were ready when the pH had dropped below 4.0 after a week.

2.7. Preparation of municipal waste for decomposition

Municipal wastes were collected from primary municipal deposit points around Tanjore and all the secondary ingredients which included wood pieces, polythene, plastics, etc., were removed. Only organic portion of the municipal wastes was considered as a final substrate for treatment which was designated as "Municipal organic waste."

2.8. Ex situ biodegradation of Municipal organic waste

The segregated municipal organic waste subjected to aerobic ex situ biodegradation in which organic wastes are biodegraded inside the composting pin. There are three plastic pins of uniform size were taken and it was named as T1, T2 and C. The biodegradation process carried out in two different set.

2.8.1. Set I: Municipal organic waste treated with SBC solution supplemented with jaggery

5 kg of municipal organic waste was placed inside the pin T1. Subsequently the 1 liters of SBC solution supplemented with jaggery was sprayed over the waste in the pin T1.

2.8.2. Set II: Municipal organic waste treated with SBC solution supplemented with molasses

5 kg of municipal organic waste was placed inside the pin T2. Subsequently the 1 liters of SBC solution supplemented with jaggery was sprayed over the waste in the pin T2. The municipal organic wastes in the pin C considered as a control, hence it was not treated with any kind of SBC solutions. Control is common for both sets.

All three pins were covered with mosquito net to avoid flies attack and prevent maggot's formation. The wastes in the pins are mixed properly once in a day and SBC solutions were sprayed twice in a week. After 30 days of biodegradation the final products were harvested and subjected to further investigation.

2.9. Analysis of biodegraded product

The harvested biodegraded products from all three pins were subjected to physico chemical analysis as per the method prescribed by Tandon (1993).

2.10. Phyto toxic study

Seed germination assay was performed to determine the phyto toxicity of the biodegraded organic waste. For this experiment, 2 g of the biodegraded municipal organic waste was diluted in 30% deionized water. Then, the extract (with 85% moisture content) was kept it for 2 hours without any disturbance. There after the extract was centrifuged at 6000 rpm for 15 minutes and the supernatant was filtered with Whatman filter paper. A 3 ml of filtered extract was poured in the five petri plates and 10 viable VRI 2 ground nut seeds (Parentage: Spanish bunch JL 24 x CO2) were placed on the each petri plates. Now the seeds were incubated in room temperature for 72 hours (Cesaro, Belgiorno and Guida, 2015; Luo *et al.*, 2018). After the incubation the germinated seeds were identified and the percentage of germination was calculated by using below formula:

$$\text{Germination \%} = \frac{\text{Number of Seeds Germinated}}{\text{Total Number of Seeds}} \times 100$$

3. RESULTS AND DISCUSSION

In this present study, Municipal Solid Waste dump fill soils were collected from two different locations namely Sreenivasapuram MSW disposal site and Punnainallur MSW disposal site and the soil samples were designated as site-A and site-B respectively. The soil samples were subjected to physico-chemical analysis and the results obtained is correlated with the findings of Saritha *et al.*, (2014); Uma, Prem Sudha & Murali (2016). Moreover the current study proved that the soil collected from Punnainallur MSW disposal site (site-B) contain higher physico chemical parameters than Sreenivasapuram MSW disposal site (site-A) (Table 1).

Table 1: Physical and chemical characteristics of two different soil samples

S. No.	Parameters	Site - A	Site - B
1	pH	6.6 ± 0.05	7.3 ± 0.07
2	EC (ms/cm)	0.795 ± 0.0008	0.798 ± 0.0005
3	CEC (meq/ds^{-1})	13.43 ± 0.003	13.58 ± 0.005
4	Nitrate (ppm)	4.5 ± 0.05	4.9 ± 0.08
5	Phosphorous B1 (ppm)	143.8 ± 0.05	144.5 ± 0.05
6	Phosphorous B2 (ppm)	194 ± 0.47	194 ± 0.47
7	Potassium (Exchang) (ppm)	30 ± 0.47	33 ± 0.48
8	Calcium (Exchang) (ppm)	172.8 ± 0.47	173.6 ± 0.47
9	Magnesium (Exchang) (ppm)	302 ± 0.47	307 ± 0.42
10	Sulphur (Available) (ppm)	122.6 ± 0.05	123.2 ± 0.06
11	Sodium (Exchang) (ppm)	482 ± 0.46	484 ± 0.41
12	Zinc (Available) (ppm)	0.7 ± 0.08	0.8 ± 0.05
13	Manganese (Available) (ppm)	6.9 ± 0.005	7.04 ± 0.005
14	Iron (Available) (ppm)	18.78 ± 0.005	18.88 ± 0.004
15	Copper (Available) (ppm)	1.02 ± 0.004	1.05 ± 0.005
16	Boron (Available) (ppm)	0.1 ± 0.04	0.2 ± 0.05
17	Organic Matter (OM) (%)	3.15 ± 0.005	3.17 ± 0.008

Each value represents mean ± SEM of 3 samples

The collected soil samples were serially diluted and cultured in the laboratory to identify the predominant bacterial strains and this study results revealed that there are 42 white colored colonies (1.68 x 10^6 CFU/ml) enumerated in site-A soil sample where as there are 56 pale yellow colored colonies (2.24 x 10^6 CFU/ml) enumerated in site-B soil sample (Table 2).

Table 2: Microbial population dynamics of two different soil samples

S. No	Sample	Number of colonies	Colony color	Results (CFU/ml)
1	Site - A	42	White	1.68 x 10^6
2	Site - B	56	Pale yellow	2.24 x 10^6

The bacterial strains present in the colonies were identified through biochemical characterization and through the results we found that the bacterial isolates present in the both soil samples are *Staphylococcus aureus, Staphylococcus epidermis, Escherichia coli, Enterobacter aerogens, Klebsiella pneumonia, Shigella sp., Bacillus subtilis* & *Pseudomonas aeruginosa* (Table 3) and these findings matches with the works of Faith Efosa Oviasogie, Christopher Uche Ajuzie & Uyiosa Glory Ighodaro (2010) and Saha & Santra (2014).

Table 3: Biochemical characterization of bacterial isolates

Samples	Microorganisms	Morphology	O_2 Requirements	Gram straining	Motility	Indole	Methyl red	Voges-proskauer	Citrate test
Site-A	*Staphylococcus aureus*	Cocci	Anaerobic	+	Non Motile	-	+	+	+
	Staphylococcus epidermidis	Cocci	Anaerobic	+	Non Motile	-	-	+	-
	Escherichia coli	Rod	Anaerobic	-	Motile	+	+	-	-
	Enterobacter aerogenes	Bacilli	Anaerobic	+	Motile	-	-	+	+
Site-B	*Klebsiella pneumoniae*	Bacilli	Anaerobic	+	Non Motile	-	-	+	+
	Shigella sp.	Rod	Anaerobic	-	Non Motile	-	+	-	-
	Bacillus subtilis	Rod	Aerobic	+	Non Motile	+	+	+	+
	Pseudomonas aeruginos	Bacilli	Aerobic	-	Motile	+	+	-	+

The effective bacterial consortium was prepared from the bacterial isolates by the method prescribed by Jeya Bharathi *et al.*, (2017). The bacterial consortium is activated later by using two different sugar industry materials such as jaggery and molasses. Both can act as a carbon source for bacterial growth. After a week after bio-activation of bacterial consortium. Later SBC was applied on the municipal organic waste at different quantity (100, 150, 200 and 250 ml). After 30 days biodegradation the end products were harvested and subjected to physico chemical analysis. The result explain that the municipal organic waste biodegraded with SBC solution plus molasses showed high nutrient value when compare to the municipal organic waste biodegraded with SBC solution plus jaggery and control (Tables 4 & 5).

Table 4: Physical and chemical characteristics of Municipal Organic Waste biodegraded by different concentrations of SBC solution supplemented with jaggery

S. No.	Parameters	Control (C)	SBC solution supplemented with jaggery (T1)			
			100 ml	150 ml	200 ml	250 ml
1	pH	6.1 ± 0.05	7.5 ± 0.05	7.8 ± 0.05	8.2 ± 0.05	8.5 ± 0.05
2	EC (mS/cm)	3.11 ± 0.008	3.15 ± 0.008	3.25 ± 0.008	3.37 ± 0.005	4.39 ± 0.009
3	Nitrogen (%)	0.9 ± 0.05	1.5 ± 0.05	1.3 ± 0.05	1.9 ± 0.05	2.3 ± 0.05
4	Phosphorous (%)	1.15 ± 0.009	1.35 ± 0.009	1.44 ± 0.008	1.61 ± 0.009	2.1 ± 0.05
5	Potassium (%)	0.32 ± 0.008	0.53 ± 0.008	0.65 ± 0.009	0.72 ± 0.05	1.1 ± 0.09
6	Carbon (%)	41 ± 0.27	52 ± 0.27	61 ± 0.27	74 ± 0.27	82 ± 00
7	C:N ratio	21:1	21:1	24:1	28:1	30:1

Each value represents mean ± SEM of 3 samples

Table 5: Physical and chemical characteristics of Municipal Organic Waste biodegraded by different concentrations of SBC solution supplemented with molasses

S. No.	Parameters	Control	SBC solution supplemented with molasses (T2)			
			100 ml	150 ml	200 ml	250 ml
1	pH	6.1 ± 0.05	7.1 ± 0.05	7.3 ± 0.05	7.5 ± 0.05	8.3 ± 0.05
2	EC (mS/cm)	3.11 ± 0.008	4.15 ± 0.008	4.25 ± 0.008	4.37 ± 0.005	5.39 ± 0.009
3	Nitrogen (%)	1.3 ± 0.05	2.5 ± 0.05	2.6 ± 0.05	2.7 ± 0.05	3.0 ± 0.05
4	Phosphorous (%)	1.45 ± 0.009	1.65 ± 0.009	1.74 ± 0.008	1.81 ± 0.009	2.0 ± 0.05
5	Potassium (%)	0.42 ± 0.008	0.69 ± 0.008	0.77 ± 0.009	0.80 ± 0.05	1.2 ± 0.09
6	Carbon (%)	54 ± 0.27	65 ± 0.27	76 ± 0.27	88 ± 0.27	93 ± 00
7	C:N ratio	21:1	24:1	27:1	30:1	31:1

Each value represents mean ± SEM of 3 samples

The maturity and toxicity of the biodegraded products were determined through seed germination assay. The seed germination assay results showed that the municipal organic waste biodegraded with 150 ml of SBC solution with molasses has highly matured and not toxic to seeds (Table 6).

Table 6: Results of seed germination assay for evaluating the phyto toxicity of biodegraded municipal organic waste

S. No.	No. of seeds	Test	Quantity of SBC solution				Control (C)
			100 ml	150 ml	200 ml	250 ml	
1	10	T1	70 ± 0.4	80 ± 0.4	80 ± 0.4	70 ± 0.4	70 ± 0.4
2	10	T2	80 ± 0.4	90 ± 1.6	70 ± 0.4	50 ± 0.4	

Each value represents mean ± SEM of 3 samples

This findings were similar with the findings of the researchers Kalaivani, Amiya Kumar & Shanthi (2011) and Premalatha *et al.*, (2017). The present study results concluded that the municipal organic waste biodegraded with a selective bacterial consortium activated with molasses can make efficient and nutritive compost product and which may be used as an effective organic supplement for crop cultivation.

4. CONCLUSION

This study is an ecofriendly approach to the organic solid waste pollution abatement. The methods adopted in this study is cost effective and provide better solution to the pollution problem. Biodegradation organic solid waste produce large quantities of nutrition rich compost product and it may generate revenue also create job opportunity for economically weaken people. In a nutshell this study may mitigate the pollution in one hand and generate revenue in other hand.

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