ISOLATION, IDENTIFICATION AND CULTURAL OPTIMIZATION OF INDIGENOUS FUNGAL ISOLATES AS A POTENTIAL BIOCONVERSION AGENT OF MUNICIPAL SOLID WASTE

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Received November 30, 2010; in final form March 25, 2011, accepted May 12, 2011

ABSTRACT

Native populations of fungi were isolated from different areas of garbage and their diversity was characterized. In all habitats, total colony-forming units (cfus) of fungi varied significantly (p=0.05) and were almost positively correlated with the characteristics of the habitat. On the basis of cultural and microscopic characteristics, the isolated strains were identified as *Trichoderma viride, Aspergillus niger, Aspergillus fumigatus, Curvularia* sp. and *Fusarium* sp. A potato dextrose broth medium was suitable for massive growth of *Trichoderma viride, Aspergillus fumigatus* whereas the Czapeck Dox broth medium was suitable for *Aspergillus niger* and *Curvularia* sp. The optimum ranges of pH and temperature were 7 - 8 and 30 - 70^{o}C, respectively. In *Trichoderma viride*, *Aspergillus niger* and *Aspergillus fumigatus*, a suitable incubation period was 3 to 4 days but in *Curvularia* sp. and *Fusarium* sp. it was 6 to 7 days. These selected fungi were tested for their potential to bioconvert municipal solid waste. A fungal suspension was found to be more effective than culture disc methods.

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Keywords: Serial dilution, cultural optimization, fungal suspension, culture disc, bioconversion.

1. INTRODUCTION

Bacteria and fungi are frequent microbes in soil, manure and decaying plant tissues that are able to degrade domestic wastes and their distribution patterns are correlated with the substrate organic matter [1]. Fungi use wastes for their metabolism and produce some simple and useful compounds that are important for soil health, plant growth and overall to keep a natural ecosystem well balanced. In biological processes, aerobic microorganisms biodegrade or mineralize solid waste completely into C, H compounds and mineral salts [2]. Micro-organisms from relevant environments previously exposed to hydrocarbons possess greater degradation capabilities on related wastes in the biodegradation process [3]. Fungi play an important role in bioconversion/ composting of organic waste and can be an important contributor to optimal agricultural waste bioconversion [4].

Jabalpur is a densely populated city. Every day a huge quantity of solid waste emanates from its urban area. The municipal solid waste in the urban centers of Jabalpur is generated by domestic, commercial and industrial sources [5]. It contains mostly organic wastes that can be decomposed by composting. Composting is the microbial decomposition of biodegradable solid waste in which microorganisms convert waste into a stable end product (compost). The main objectives of composting are to reduce the solid volume, weight and moisture content, minimize odor, decrease pathogens and the spread of disease and increase potential nutrients for agricultural applications. Therefore, composting is emerging as a popular waste management alternative both in developed and developing countries including cities like Jabalpur [6-8].

Implementation of composting technology has great potential for mitigating several problems related to an ecological imbalance due to loss of nutrients from ecosystems and the disposal of organic wastes that cause water, soil and air pollution and corresponding health hazards. Therefore, the present investigation was carried out to isolate effective fungal strains from different municipal solid waste in order to identify and optimize their culture conditions. Further, these fungal strains were applied to decompose municipal solid waste (MSW) *in vitro* to determine their potential as bioconversion agents.

2. MATERIALS AND METHODS

2.1. Sampling

For isolation of effective fungal strains, different types of samples, such as garbage and MSW were collected from different places in Jabalpur City from March to September, 2009. These samples were obtained by digging to 15 cm depth with sterile spoons and were collected in sterile plastic pots. The samples were transported to the laboratory and stored at 4°C until use.

2.2. Determination of Moisture Content (%) and pH of Samples

Freshly collected samples were taken in bottles and the initial weight was recorded. These moisture content of the samples were measured in a hot air incubator at 105°C to constant weight [9]. The sample acidity (pH) was measured using the published method of ref. [10].

2.3. Chemical Analysis

The organic wastes collected from different sources were also analyzed for electrical conductivity, organic carbon, total nitrogen, total phosphorus and total potassium using standard procedures. Total micro-nutrients were analyzed with a standard inductively coupled plasma emission spectroscopy (ICPS) [10,11].

2.4. Isolation of Fungi

A serial dilution technique was used for isolation of fungi. In this technique, a sample suspension was prepared by adding 1.0 gm sample to 100 mL distilled water and shaking for 15 minutes in an orbital shaker [12,13]. Immediately afterwards, each suspension was serially diluted to 10^{-6}. From this 10^{-6} gm/L sample, 0.1 mL was pipetted onto plates with potato dextrose agar media, spread with a glass spreader and incubated at 30°C for observation. Each colony that appeared on the plate was considered as one colony forming unit (cfu). Chloramphenicol antibiotic (0.03 mg/L) was added to media to avoid bacterial contamination.

2.5. Identification

The fungal isolates were identified by morphological examination and its characteristics with established methods of ref [14-16].

2.6. Optimization of Culture Conditions

Three broth media (Potato dextrose broth, Czapeck Dox broth and Richards's broth) were used to optimize the culture media of isolated fungi. The media were adjusted to pH 4, 5, 6, 7 and 8. For optimization of the incubation period and temperature, the culture flasks were incubated at 30, 40, 50, 60 and 70°C for 4 to 7 days [17,18].

2.7. Preparation and Application of Fungal Cultures

2.7.1. Disc for Garbage Decomposition

At first the fungal strains were cultured on their preferred medium at 40-50°C for 4-7 days. Culture discs (5 mm diameter) were cut with a cork borer for each test organism and added to autoclaved MSW at 5 culture discs/100gm MSW. Control treatments performed with no inoculation were incubated at 30°C. Sterile thermometers were pushed through a cotton plug in each bottle [19], which was later sealed with parafilm and labeled. All procedures were conducted under running laminar airflow.

2.7.2. Preparation and Application of Fungal Suspensions for Garbage Decomposition

Ten mL of autoclaved distilled water was added to a 4 to 7 day-old fungal culture tube and shaken well to get a fungal suspension. Five mL of this suspension was mixed with 100g of sterilized garbage. Control treatments performed without adding any inoculums were incubated at 30°C. The following observations were made for the decomposed solid waste: changes of temperature, pH, moisture, volume and weight losses of decomposed organic solid waste [19,20]. Data were recorded in 5 day intervals up to 55 days. For volume loss the following formula was used: $V = \pi r^2 h$, where V = volume of the garbage, r = radius of the bottle, h = height of the decomposed garbage. Volume loss (%) = $(V-V_1/V) \times 100$, where V = initial volume, V_1 = final volume. Percentage weight loss of decomposed garbage was also calculated employing the following formula: Weight loss (%) = $(W-W_1/W) \times 100$, where W = initial weight and W_1 = final weight.

2.8. Statistical Analysis

The experiment was conducted using a randomized complete block design with three replications. Results of all analyses were judged for significance at the 5% level. All data were analyzed with Microsoft- Excel® [21].

3. RESULTS AND DISCUSSION

3.1. Effect of Physical and Chemical Characteristics on Populations of Fungi

Total cfu of fungi recovered from different samples were high and unequal in number. Colony-forming units of fungi in the different habitats were significantly (P=0.05) different (Table 1). Populations of fungi ranged from 152 $\times 10^{-6}$/gm up to 225 $\times 10^{-6}$/gm. The highest number of cfus was found in garbage sample 8 and the lowest in sample 1. The pH ranges were from 5 to 7. Maximum pH was obtained from sample 2 and the minimum in sample 10. Sample acidity showed positive and significant (P=0.05) correlation with the population of fungi (Table 1). Most of the fungi need pH between 6 to 7 for growth. However, a few fungi prefer more extreme pH values for growth [22].

The range of % moisture content varied from 35 to 57%. The maximum moisture content was for sample 8 and the minimum for sample 5 (Table 1). The results indicate that the moisture contents of the different samples habitats were significant (P=0.05) and positively correlated with the population of fungi. The fungal population of various garbage samples is closely correlated with moisture content: the maximum fungal density is found in regions of fairly high moisture content: the optimum level for aerobic fungi growth often is 50-75% of the garbage moisture holding capacity [23]. Fungal growth also depends on other physiochemical conditions such as substrate, pH, temperature, incubation period, and carbon source. Conditions in which fungi grow in natural habitats should be studied before so scaling up for use as decomposers.

In the present study, organic matter, N, P and K contents varied in different habitats. Organic matter, P and K contents showed positive and significant correlation with the population of fungi. However, no significant correlation of N content was noted with the fungal population [20].

3.2. Isolation and Medium Selection of Sample Fungi

A total of fifty-five fungal strains were obtained from different samples. Only five strains were cultured on a large scale and these strains were further cultured in potato dextrose agar, Czapek-Dox-Agar and Richards's agar media. It was observed that the basic Czapek-Dox-Agar medium was suitable for massive growth of *Fusarium* sp. and *Curvularia* sp. strains; the potato dextrose medium was suitable for the massive growth of *Trichoderma viride, Aspergillus niger, Aspergillus fumigatus* and *Curvularia* sp. strains. Previous work reported that potato dextrose agar was suitable for maximum growth of fungi [24]. *Aspergillus niger, Aspergillus fumigatus* and *Curvularia* sp. grew well in the Czapek-Dox-Agar medium [25].

3.3. Identification of Selected Fungi

The five selected strains were cultured on their preferred media, their cultural characteristics were observed and the results were summarized. The selected strains were identified as *Trichoderma viride, Aspergillus niger, Aspergillus fumigatus, Curvularia* sp. and *Fusarium* sp. [26].

3.4. Optimization of Culture Conditions

Different cultural conditions were studied for massive growth of fungi and the most favorable condition was selected before mass production. pH is a key factor for growing fungi in artificial media. In this study, the pH of potato dextrose broth, and Czapek-Dox broth media were optimized for culturing the strains. The ranges of pH 7 to 8 of two media were suitable for growth of fungi: Czapek-Dox broth medium with pH 7, and potato dextrose broth medium with pH 7 were optimum for the maximum growth of isolated fungal strains (Table 2). It is relevant from the results that the pH of samples from which the strains *Trichoderma viride, Aspergillus niger, Aspergillus fumigatus*, *Curvularia* sp. and *Fusarium* sp. were isolated exhibited pH 4, 5, 6, 7 and 8, respectively and possibly for this reason, these strains also grew well in *in vitro* conditions at alkaline pH [27].

Fungi tolerate a soil reaction between pH 4 to 10, but the most favorable pH for the majority is just on the alkaline side of neutrality. In the present investigation, the five strains isolated were incubated at 30, 40, 50, 60 and 70ºC and the most massive growth of all the strains was found at 40-60ºC in their selected media (Table 3). The optimum temperature range for the growth of selected fungi was 25-35ºC. A great number of fungi may grow quite well over the temperature range 10-40ºC [28]. Sultana observed that 33 ± 4ºC was ideal for the growth of fungi [29]. On the other hand, Sarker observed 30ºC for maximum growth of the isolated strains [30].

The strains obtained in this study were incubated for 1-6 days. The results show that 7 days of incubation was suitable for growth of fungal strains and 4 to 7 days was suitable for *Trichoderma viride, Aspergillus niger* and *Aspergillus fumigatus* strains (Table 3).

Table 1 Source, pH, moisture content, chemical characteristics and total cfu (10^{-6}/gm) of different garbage samples from which fungal strains were isolated

Sample No.	Location	pH	Moisture (%)	Organic matter (%)	Total N (%)	P mg/kg	K mg/kg	cfu (10^{-6}/gm)
S/1	Zone 1	7.15±0.13	45.3	27.9	0.84	0.25	8	152
S/2	Zone 2	7.65±0.21	52.2	29.6	0.98	0.12	7	158
S/3	Zone 3	6.93±0.18	36.1	24.2	1.02	0.54	9	181
S/4	Zone 4	7.42±0.26	38.3	25.3	0.59	0.34	5	167
S/5	Zone 5	6.81±0.30	35.3	28.5	0.87	0.28	8	176
S/6	Zone 6	6.35±0.26	48.2	30.0	0.78	0.23	6	192
S/7	Zone 7	7.68±0.15	55.3	32.5	1.23	0.32	8	198
S/8	Zone 8	7.59±0.24	57.4	34.7	1.27	0.35	9	225
S/9	Zone 9	7.38±0.17	38.6	22.7	0.86	0.45	6	165
S/10	Zone 10	6.16±0.21	39.6	29.0	0.98	0.42	5	186

Table 2 Effect of pH on the growth of isolated strains on potato dextrose, Czapek-Dox and Richards's broth media

Media	pH of medium	Strains	Incubation period (days)						
			1	2	3	4	5	6	7
Potato dextrose broth	4	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	-	++	++	++++	++++
		Aspergillus fumigatus	-	-	+	+	+	+++	+++
		Curvularia sp.	-	-	+	+	+	+	+
		Fusarium sp.	-	-	-	+	+	++	+++
	5	*Trichoderma viride*	-	+	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	+	++	+++	++++
		Aspergillus fumigatus	-	-	+	+	++	+++	++++
		Curvularia sp.	-	-	-	+	+	++	+++
		Fusarium sp.	-	-	-	+	+	++	+++
	6	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus fumigatus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	7	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus fumigatus	-	-	+	+	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	8	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	-	-	-	-
Czapek-Dox broth	4	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	+	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++

- No growth, + Poor growth, ++ Moderate growth, +++ Good growth, ++++ Massive growth.

Table 2 Effect of pH on the growth of isolated strains on potato dextrose, Czapek-Dox and Richards's broth media *(continued)*

Media	pH of medium	Strains	Incubation period (days)						
			1	2	3	4	5	6	7
	5	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	6	*Trichoderma viride*	-	-	+	+	++	+++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	7	*Trichoderma viride*	-	-	+	+	++	+++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	-	-	-	-
	8	*Trichoderma viride*	-	-	-	-	+	++	+++
		Aspergillus niger	-	-	-	-	+	++	+++
		Aspergillus fumigatus	-	-	-	-	+	++	+++
		Curvularia sp.	-	-	-	-	+	++	+++
		Fusarium sp.	-	-	-	-	-	-	-
Richards's broth	4	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	+	++	+++	++++
		Aspergillus fumigatus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	5	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus fumigatus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	6	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus fumigatus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	7	*Trichoderma viride*	-	-	-	+	++	+++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	-	+	++	+++
		Fusarium sp.	-	-	-	-	-	-	-
	8	*Trichoderma viride*	-	-	-	-	+	++	+++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus fumigatus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-

- No growth, + Poor growth, ++ Moderate growth, +++ Good growth, ++++ Massive growth.

Table 3 Effect of temperature on the growth of isolated strains

Media	Temperature, °C	Strains	Incubation period (days)						
			1	2	3	4	5	6	7
Potato dextrose broth	30	*Trichoderma viride*	-	-	++	+++	++++	++++	++++
		Aspergillus niger	-	-	-	++	++	++++	++++
		Aspergillus	-	-	+	+	+	+++	+++
		Curvularia sp.	-	-	+	+	+	+	+
		Fusarium sp.	-	-	-	+	+	++	+++
	40	*Trichoderma viride*	-	+	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	+	++	+++	++++
		Aspergillus	-	-	+	+	++	+++	++++
		Curvularia sp.	-	-	-	+	+	++	+++
		Fusarium sp.	-	-	-	+	+	++	+++
	50	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-				
		Fusarium sp.	-	-	-				
	60	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus	-	-	+	+	+++	++++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-
	70	*Trichoderma viride*	-					+	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-
Czapek-Dox	30	*Trichoderma viride*	-	+	++	+++	++++	++++	++++
		Aspergillus niger	-	-	+	+	++	+++	++++
		Aspergillus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	40	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	50	*Trichoderma viride*	-	-	+	+	++	+++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	60	*Trichoderma viride*	-	-	+	+	++	+++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-
	70	*Trichoderma viride*	-	-	-	+	+	++	+++
		Aspergillus niger	-	-	-	-	+	++	+++
		Aspergillus	-	-	-	-	+	++	+++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-

- No growth, + Poor growth, ++ Moderate growth, +++ Good growth, ++++ Massive growth.

Table 3 Effect of temperature on the growth of isolated strains *(continued)*

Media	Temperature, °C	Strains	Incubation period (days)						
			1	2	3	4	5	6	7
Richards's broth	30	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	+	++	+++	++++
		Aspergillus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	40	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	+	++	+++	++++
		Fusarium sp.	-	-	-	+	++	+++	++++
	50	*Trichoderma viride*	-	-	+	++	+++	++++	++++
		Aspergillus niger	-	-	+	++	+++	++++	++++
		Aspergillus	-	-	+	++	+++	++++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-
	60	*Trichoderma viride*	-	-	-	+	++	+++	++++
		Aspergillus niger	-	-	-	+	++	+++	++++
		Aspergillus	-	-	-	+	++	+++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-
	70	*Trichoderma viride*	-	-	-	-	+	++	+++
		Aspergillus niger	-	-	-	-	++	+++	++++
		Aspergillus	-	-	-	-	++	+++	++++
		Curvularia sp.	-	-	-	-	-	-	-
		Fusarium sp.	-	-	-	-	-	-	-

- No growth, + Poor growth, ++ Moderate growth, +++ Good growth, ++++ Massive growth.

Fungi can grow at 50ºC for 4-7 days and show good growth at 50ºC for 4-7 days of incubation. At least 7 days incubation at 50ºC for substantial growth of *Curvularia* sp. and *Fusarium* sp is required [21].

3.5. Bioconversion of Solid Waste Using Culture Discs

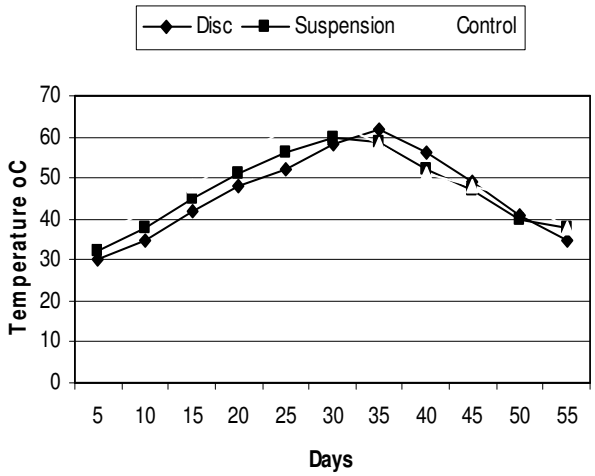
Changes of odor and color of decomposed solid waste were observed after 5 days and up to 55 days (data not shown). The odor of decomposed garbage changed from a sweet bad smell to odorless after 5-10, 15-25 and 55 days in all treatments except the *Aspergillus niger* control, where the bad smell was emitted after 55 days of incubation for decomposition. The color of garbage turned brownish to deep-black-brownish in *Trichoderma viride* treatment but in other treatments it was a greenish brown to brownish color. In the control it was greenish brown to brownish green [31]. A five degree temperature increase was observed after 20 days in all treatments (Fig. 1) but the temperature decreased to 50ºC after 20 days. In the control temperature increased by 3ºC after 20 days [26].

The initial pH of garbage was 7.32 but after 55 days of treatments the pH of decomposed garbage changed to 7.89, 7.54, 7.99, 8.10 and 7.65 when treated with *Trichoderma viride, Aspergillus niger*, *Aspergillus fumigatus, Curvularia* sp. and *Fusarium* sp., respectively. Treatment with *Trichoderma viride* showed the highest volume and weight loss, 67 and 55%, respectively after 55 days, whereas in the control it was 41 and 26%, respectively (Fig. 2).

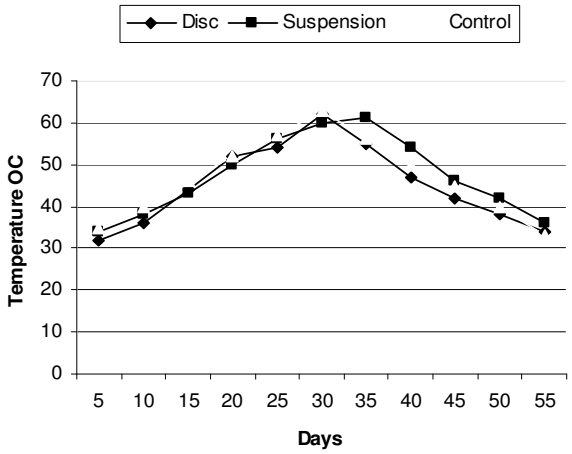
3.6. Bioconversion of Solid Waste Using A Spore Suspension

The changes of odor and color of treated solid waste were as in the culture disc treatment: A temperature increase after 20 days in *Trichoderma viride* treatment (Fig. 1) but a temperature decrease up to 36 ºC after 55 days. In control the temperature was increased by 3ºC after 20 days. The initial pH of garbage was 7.32

but after 30 days of inoculation the pH of decomposed garbage changed to 7.9, 7.6, 7.7, 8.6 and 8.2 when it was treated with *Trichoderma viride Aspergillus niger*, *Aspergillus fumigatus, Curvularia* sp. and *Fusarium* sp., respectively. The highest volume loss and weight loss was observed to be 64.3 and 52.2%, respectively, after 55 days in *Trichoderma viride* treatment and the lowest were 40.0 and 26.5 %, respectively, in the control (Fig. 2 and 3).

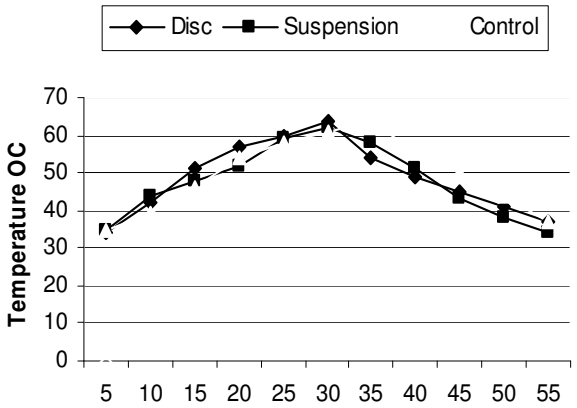


(a) *Trichoderma viride*

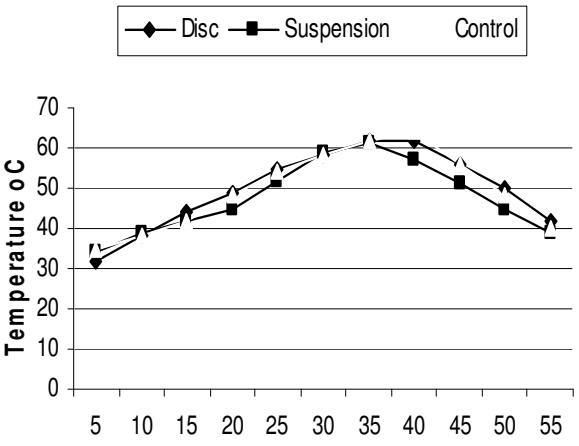


(b) *Aspergillus niger*

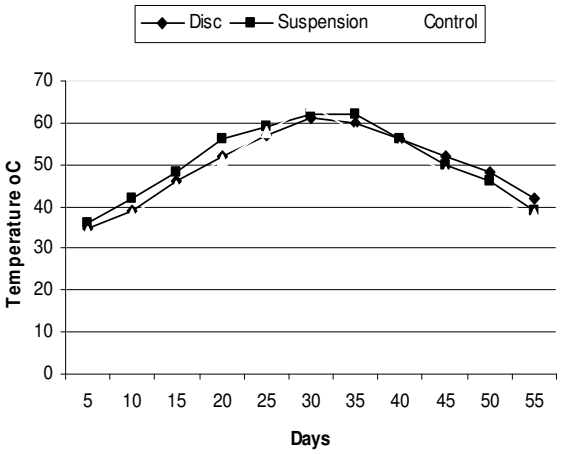
Figure 1 Changes of temperature (^{o}C) during decomposition of municipal solid waste at 5 day intervals using fungal culture disc and suspension of five fungal strains (a) *Trichoderma viride*, (b) *Aspergillus niger*, (c) *Aspergillus fumigatus,* (d) *Curvularia* sp., (e) *Fusarium* sp.



(c) *Aspergillus fumigatus*



(d) *Curvularia* sp.



(e) *Fusarium* sp

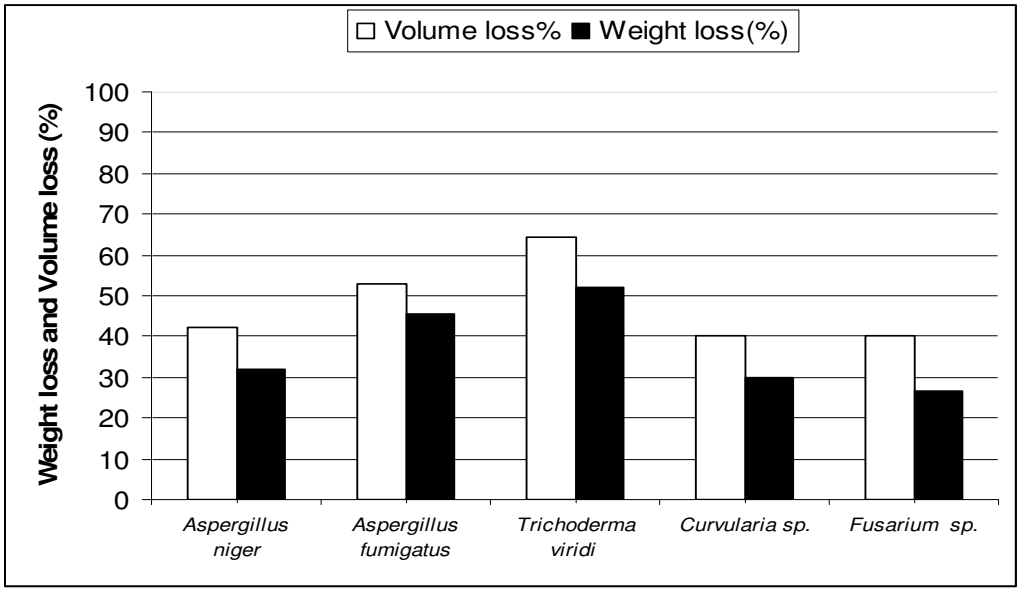


Figure 2 Volume loss (%) and Weight loss (%) of decomposed municipal solid waste with culture discs of different fungal strains after 45 days

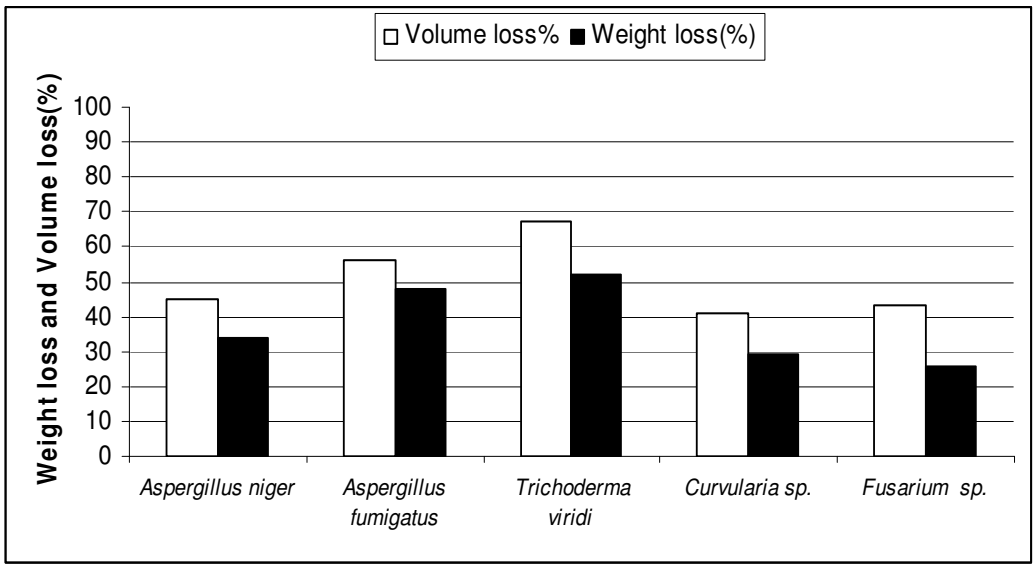
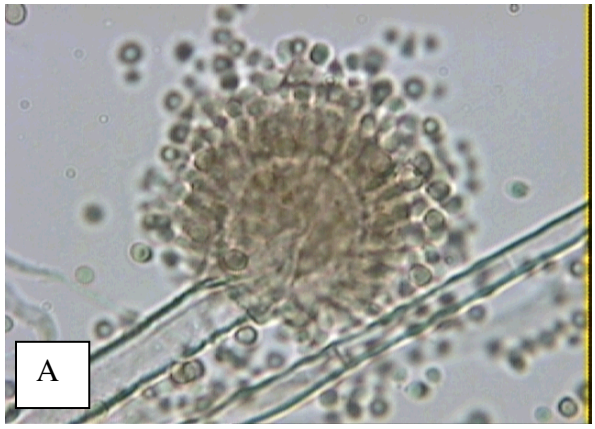
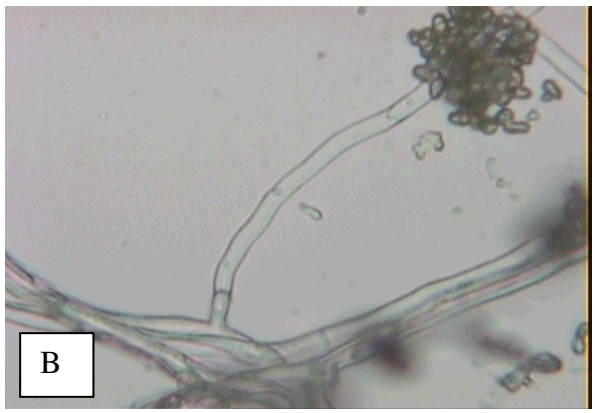


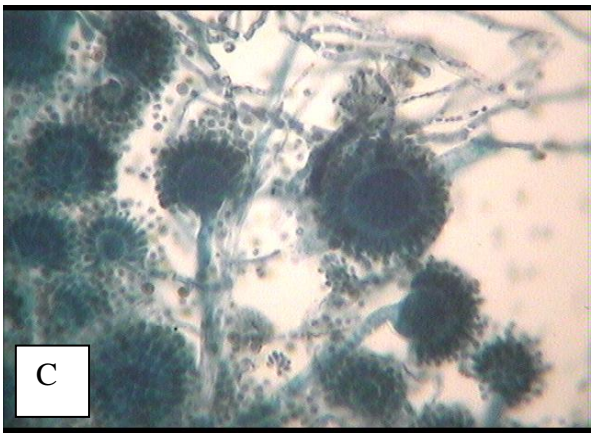
Figure 3 Volume loss (%) and Weight loss (%) of decomposed municipal solid waste with fungal suspension of different fungal strains after 45 days

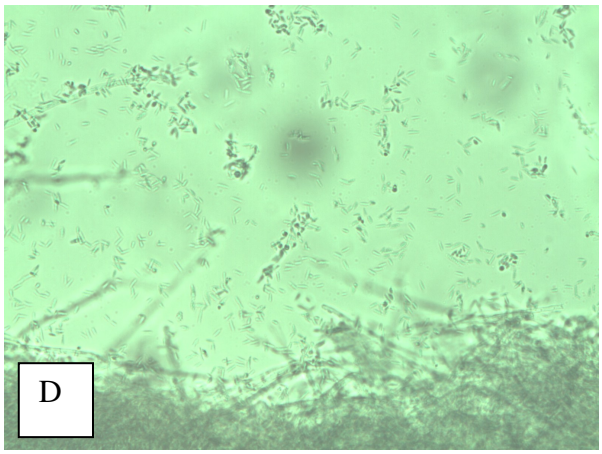
For decomposition of organic solid waste either using a culture discs or a spore suspension no bad smell was emitted after 55 days. This indicates the possible complete degradation of organic waste [20]. For control solid waste, bad smell continued even after 55 days, indicating slow degradation. Rahman also reported similar results in his solid waste decomposition experiments [31]. When microbes degrade wastes, heat is produced and the temperature increases with decomposition; and rising temperatures accelerates plant residue degradation [24]. Tchobanoglous also reported temperature increases from the initial day of composting up to two or three weeks but the temperature remained constant after that [32].

Among the five fungal strains used in the present study, *Trichoderma viride* is the most effective strain on the basis of volume loss (%) and weight loss (%) data. A fungal suspension was more effective than the culture disc method. It also was observed that *Trichoderma* sp. were the most effective strain for solid waste decomposition. Zheng and Shetty reported that *Trichoderma viride* is capable of producing various polysaccharide degrading enzymes, which may help to degrade long-chain carbon compounds, particularly materials normally incorporated for composting of cellulosic materials [33]. Martin and Dale studied the biodegradation of turf thatch with wood decay fungi [34]. They used weight loss measurements to estimate tissue degradation of Bermuda grass (*Cynodon dactylon* L.) and zoysia grass (*Zoysia japonica* Steud., 'Mayer') stolons, and showed that some wood decay fungi can decompose certain turf thatch components. Bari reported that *Trichoderma harzianum* was the most effective strain for solid waste decomposition and showed the highest weight loss (31.8%) when the culture disc approach was used [35]. The present results are partially in accordance with the findings of Zheng and Shetty, and Martin and Dale [34,35].









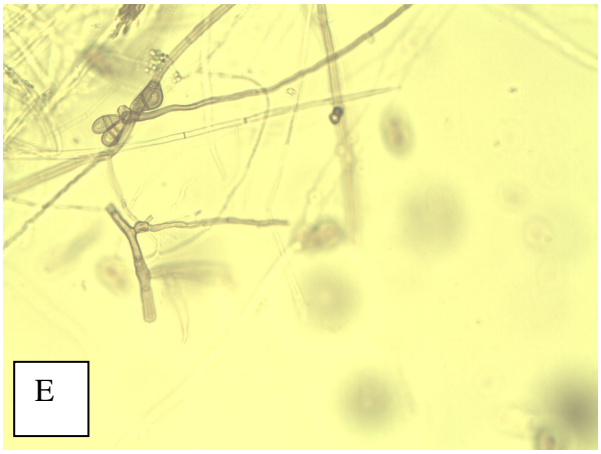


Figure 4 Microscope images show A, *Aspergillus fumigatus,* B. *Trichoderma viride*, C. *Aspergillus niger*, D. *Fusarium* sp., E. *Curvularia* sp.

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4. CONCLUSION

We conclude that significant numbers of fungi were correlated (P = 0.05) with the physiochemical characteristics of the samples. The identified genus was *Trichoderma viride, Aspergillus niger, Aspergillus fumigatus, Curvularia* sp. and *Fusarium* sp. Their optimum growth was observed in basic media in the temperature range 50-60°C. *Trichoderma viride* was the most potent strain for bioconversion, exhibiting 15.3% greater weight loss than the control when spore suspension was used. Fungal suspensions were more effective than culture disc treatments. Natural sources are rich in fungi and these fungi may promote degradation of solid waste. *Trichoderma viride* was the favored strain for degradation and can be used effectively to prepare compost within a short time period.

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