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## Isolation, characterization and identification of predominant microorganisms from agro-waste

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### ABSTRACT

The study of biological degradation of agro waste materials by different microorganisms with a view to pollution control is an important strategy. The complex molecules in wastes are decomposed by many microorganisms, which are either soil borne or air borne. These organisms have many applications in the production of organic manure and bio-fertilizers. The isolation and identification of biodegrading microorganisms such as bacteria, fungi, actinomycetes were carried out on bagasse, press mud, farm yard and mushroom waste by gram staining and biochemical methods. *Pseudomonas* sp., *Bacillus* sp., *Aspargillus* sp., *Penicillium* sp., *Streptomyces* sp. and *Thermo actinomycetes* were isolated from these different organic wastes. These isolated microorganisms could be used for bio fertilizer production, for use as industrial microorganisms and for mitigation of pollution.

**Key words:** Agro waste, *Actinomycetes*, *Aspargillus*, Bagasse, Press mud, Mushroom waste

### INTRODUCTION

Highly toxic organic compounds have been synthesised and released into the environment directly or indirectly over a long period of time by industrial and agricultural activities [1]. Agro wastes include solids, liquids and gases. The production and improper disposal of agro wastes has become a major pollution issue round the globe [2]. Everyday huge quantity of waste is generated in all the developing and developed countries. Biological decomposition of organic waste such as fertilizers, pesticides and agro wastes are the most important and effective way to remove these compounds from the environment. Bacteria, *Actinomycetes*, fungi, algae and protozoa are the major microorganisms found in soil which decompose soil organic materials, of which bacteria are most prominent and most abundant [3]. Microbes use the waste for their own metabolism and finally produce some simple and useful compounds which are important for soil health, plant growth and overall eco-balance. Microorganisms have the ability to interact, both chemically and physically with substances, leading to the structural changes or complete degradation of the target molecules [4]. Therefore, the present study was aimed to focus at the importance of isolation, characterization and identification of microorganisms from waste dumping soils. Our results suggest that a number of microorganisms such as bacteria, fungi and *Actinomycetes* have the ability to degrade lignin, cellulose and hemicellulose content in these soils samples.

### MATERIALS AND METHODS

#### Determination of moisture from different organic wastes

Two grams of the sample was taken in a pre weighed glass crucible and was allowed to dry for 6 hours in a hot air oven. The crucible was weighed and the readings were noted. The moisture content was calculated from the readings.

**Determination of pH from different organic wastes**

The pH of organic waste was obtained by Elico model Lt-10T pH meter, by preparing 1:10 diluted sample suspension in water and stirring by means of glass rod. One gram of sieved sample was taken in a 50 ml glass beaker. To it, 10 ml of distilled water was added and mixed well by keeping in shaker for 60 mints. Then the mixture was filtered by using filter paper and the filtrate was stirred with glass rod. The pH value was found by using pH meter.

**Determination of organic carbon from different organic wastes**

Half gm of air dried sieved sample was taken in a 500 ml conical flask and 50 ml of 1N Potassium Dichromate solution was added. To that 50 ml of conc. Sulphuric acid was added and mixed well. The mixture was incubated at room temperature for 30 mints. To it, 200 ml of distilled water was added and made up to 500 ml. From this solution 50 ml of sample was taken in a fresh 500 ml conical flask and 15 ml of Ortho-phosphoric acid was added. One ml of Diphenylamine indicator was added before titrating with ferrous ammonium sulphate solution. The end point indicated by blue colour changing into dark green and the values were noted to calculate the Carbon content.

**Estimation of Nitrogen content**

Half gm of sieved sample was taken in a fresh test tube and 2.5 ml of conc. H<sub>2</sub>SO<sub>4</sub> was added. The test tube was heated until dark black was formed. To this, distilled water was added until the solution became colourless. This was filtered and the filtrate was separated. One ml of Nessler's reagent was added to the filtrate and the reddish brown colour was read at 490 nm.

**Estimation of Cellulose**

Half gm of sieved sample was taken in a 250 ml conical flask and 3 ml of Acetic/Nitric acid reagent was added and mixed well. Then the flask was kept in the water bath at 100 °C for 30 mints. It was cooled and centrifuges for 20 mints and the supernatant was discarded. The residue was washed with distilled water and 10 ml of 67% Sulphuric acid was added and allowed to stand for 60 mints. From 100 ml of this mixture 1 ml was sample and 10 ml of Anthrone reagent was added alternatively and mixed in a 15 ml test tube. Then the mixture was kept in the water bath for 10 minutes and the colour was measured at 630nm.

**Estimation of Hemicellulose**

Half gm of sieved sample was taken in a test tube and 10 ml of 24% Potassium Hydroxide was added. The sample was incubated at room temperature for 4 hrs. The sample was filtered and the filtrate was washed with distilled water and allowed to dry in hot air oven at 100 °C for 1 hr. The dried material was taken and weighed to calculate the hemicellulose content.

**Estimation of lignin**

Half gm of sieved sample was taken in a conical flask and 2 ml of 72% sulphuric acid was added and mixed at room temperature. It was allowed to cool and then diluted with 28 ml of distilled water. Then the mixture was shifted to 125 ml of conical flask and autoclaved. The sample was allowed to cool for 1 hr. The aqueous layer of the sample was centrifuged. The supernatant was discarded and the precipitate was washed with distilled water. The precipitate was dried in a glass plate in the hot air oven at 100<sup>0</sup> C for 1 hr. The dried material was weighed to calculate the lignin content.

**Estimation of Starch**

Half gm of sample was homogenised in hot 80% ethanol and centrifuged. The residue was washed with 80% ethanol and allowed to dry for few mint. To it 5 ml of water and 6.5 ml of 52% Perchloric acid was added alternatively and centrifuged at 0<sup>0</sup> C. Then the supernatant was taken and made up to 100 ml. 0.1 ml of supernatant was taken out by a pipette and made up to 1 ml. To this 4 ml of Anthrone reagent was added and kept in the water bath to increase the reaction. The solution was cooled and read for the intensity of dark green colour at 630 nm.

**Sterilisation**

Glassware and culture media were sterilised in an autoclave for 15 mints. and used for the isolation, characterisation and identification of organisms which are responsible for the degradation of different organic waste.

**Collection of samples**

The four different samples from different agro industrial wastes such as press mud, bagasse, farm yard and mushroom waste were collected in and around Chennai, India.

**Preparation of samples**

Dispensed one gram of organic sample in 10 ml of distilled water, mixed well by Vortexing and transferred one ml of suspension to another test tube to make  $10^{-5}$  dilution. Dilution procedure was continued up to  $10^{-6}$ .

**Spread plate methods**

Nutrient agar plates were prepared and 0.1 ml of suspension was pipetted from each dilution on the agar surface. The L rod was dipped in 95% alcohol which was taken in the beaker. The glass rod was removed from the beaker and the bent position was sterilised in the Bunsen burner flame. The rod was cooled for 10-15 sec. and softly touched on the agar and spread the suspension on the agar surface. The procedure was repeatedly carried out to prepare up to  $10^{-6}$  and then the plates were incubated in an inverted position at 25 °c for 24 to 48 hrs.

**Enumeration of colonies**

The method, Most Probable Number (MPN), was used for the enumeration of cultured colonies. The different colonies in the plate were counted manually. For each sample the counting was carried out and the count of bacteria, fungi and *Actinomyces* was tabulated.

**Identification of organisms**

After the growth of microbial colonies in the spread plates the various colonies were differentiated by colony morphology. Then the colonies are streaked onto the different agar slants by taking a loop full of culture. From those slants a single colony was inoculated into the sterile broths and incubated for 4 to 6 hrs. These were used for further experiment. The bacterial cultures were identified as positive or negative to gram staining. Gram staining is an old and reliable method for observing the bacteria. Gram negative bacteria were decolourised by alcohol, losing the purple colour of crystal violet stain. Gram positive did not decolourise and remained purple [5].

**Biochemical Identification**

Various biochemical tests such as Indole, Methyl Red, Voges-Proskauer, Citrate and Hydrogen Sulphide on TSI were done.

**Preliminary morphological identification of fungal culture****Lactophenol Cotton Blue staining**

A drop of Lactophenol Cotton Blue stain was dropped on a clean micro-concave glass slide. Then the fungal culture was teased using a teasing needle and kept in the drop of stain. A cover slip was carefully placed on the slide without formation of any air bubble and the slide was examined.

**Preliminary morphological identification of *Actenomyces* Culture**

A single identical colony was picked from the pure culture slants and made a thin uniform smear on a clean glass slide and it was allowed to dry. Methylene blue solution was flooded over the smear and stained for 1 min. The slide was washed with water and air dried.

**RESULTS**

The various activities of human beings such as domestic farming, agriculture and industry generate waste products which cannot be used profitably and discarded as waste which is available as crop residues, fertilisers, pesticides, and other agro industrial wastes such as bagasse, press mud, mushroom waste and coir pith etc. The major composition of these wastes is cellulose, hemicelluloses, lignin, carbohydrates, proteins and fatty materials.

The different organic waste samples were collected from agro based industries and the physio-chemical parameters like pH, moisture, organic carbon and organic nitrogen content and the chemical composition like cellulose, hemicelluloses, lignin and starch were determined. The different predominant microbial populations like bacteria, fungi and *Actinomyces* were isolated from different organic wastes are tabulated in Table 1.

**Table 1: Microbial populations of different organic wastes**

Bagasse						Mushroom Waste					
Fungi		Actinomycetes		Bacteria		Fungi		Actinomycetes		Bacteria	
$10^{-2}$	$10^{-3}$	$10^{-4}$	$10^{-5}$	$10^{-5}$	$10^{-6}$	$10^{-2}$	$10^{-3}$	$10^{-4}$	$10^{-5}$	$10^{-5}$	$10^{-6}$
22	11	26	15	49	72	12	4	22	18	87	60

  

Press Mud						Farm Yard Waste					
Fungi		Actinomycetes		Bacteria		Fungi		Actinomycetes		Bacteria	
$10^{-2}$	$10^{-3}$	$10^{-4}$	$10^{-5}$	$10^{-5}$	$10^{-6}$	$10^{-2}$	$10^{-3}$	$10^{-4}$	$10^{-5}$	$10^{-5}$	$10^{-6}$
27	7	34	27	65	40	18	4	26	15	41	23

Bacterial population in various soil samples was found to be closely correlated with the moisture content. The maximum bacterial density was found in regions of fairly high moisture content and the optimum level for the activities of aerobic bacteria often is 50-77% of the soil moisture. From our data it was found that bacterial population was much higher as compared to fungal and pure *Actinomyces* populations.

Table 2 shows the types of cultures of bacterial, fungal and *Actinomyces* isolated from different organic wastes such as mushroom waste, press mud and bagasse.

**Table 2 Isolation of different microbes from different organic wastes**

Bagasse			Mushroom Waste		
Fungi	<i>Actinomyces</i>	Bacteria	Fungi	<i>Actinomyces</i>	Bacteria
BF-1 BF-2	BA-1 BA-2	BB-1 BB-2	MF-1 MF-2 MF-3 MF-4 MF-5	MA-1 MA-2 MA-3	MB-1 MB-2
Press Mud			Farm Yard Waste		
Fungi	<i>Actinomyces</i>	Bacteria	Fungi	<i>Actinomyces</i>	Bacteria
PF-1 PF-2	PA-1 PA-2	PB-1 PB-2 PB-3 PB-4 PB-5	FF-1 FF-2 FF-3 FF-4	FA-1 FA-2 FA-3 FA-4 FA-5	FB-1 FB-2 FB-3 FB-4 FB-5

Table 3 shows the physio-chemical parameters and chemical composition of different organic wastes

**Table 3 Physiochemical parameters and chemical components of different organic wastes**

S.No	Different Organic Wastes	Physiochemical parameters					Contents of Chemical Components(%)			
		pH	Moisture (%)	Org. Carbon (%)	N <sub>2</sub> Content	C:N Ratio	Cellu	Hemi-Cellu.	Lign	Starch
1.	Bagasse	5.5	32.22	54.6	0.9	60:1	41	36	21.5	86.4
2.	Press Mud	5.1	48.6	46.8	1.2	39:1	30.2	25	10.4	52.8
3.	Mushroom Waste	5.6	54.86	23.4	0.9	27:1	32.2	86	7.9	28.8
4.	Farm yard Waste	7.3	64.08	35	0.4	88:1	60.4	66	9.1	52.8

The pH level was 7.3 and the C: N ratio was found to be high 88:1 in farm yard waste.

There was not much variation in the cellulose content in all the four waste samples.

High hemicelluloses content was observed in mushroom waste whereas lignin and starch content was high in bagasse.

Table 4 shows the result of morphological characteristics and biochemical tests of five bacterial isolates from the farm yard waste. All the isolates were gram negative rods.

**Table 4 Morphological and Biochemical Characterisation of farmyard waste isolates**

Isolate No.	Morphological Characterisation		Biochemical Characterisation									
			Citrate Test	MR Test	VP Test	Indole Test	Triple Sugar Iron Test				Catalase Test	Oxidase Test
	Culture Characterisation of Colonies	Gram Staining					H <sub>2</sub> S	Gas	Butt	Slant		
F1	Transparent	Gram(-)	+	+	+	-	-	-	+	-	+	+
F2	Transparent White	Gram(-)	-	+	-	+	-	+	+	-	+	-
F3	White	Gram(-)	+	-	+	-	-	+	+	-	-	-
F4	Transparent	Gram(-)	+	-	-	-	-	-	+	-	+	+
F5	White	Gram(-)	+	-	+	-	+	-	+	-	+	+

“+” = Present. “-” = Absent

When citrate test was conducted among the five isolates four shown positive by the appearance of blue colour in the medium and the remaining one isolate showed no change in colour indicating negative results.

In the MR test 2 isolates shown positive and the remaining three showed negative results. The VP test showed that three isolates showed positive the rest two negative results. The indole test showed only one isolate positive and the rest four showed negative results. The TSI showed that one isolate produced H<sub>2</sub>S which was identified by the

formation of black colour in the medium and remaining did not, the gas formation was observed in two isolates, all the five isolates changed their butt to yellow colour indicating acid formation, the slant of one isolate turned to yellow and the all the remaining isolate slants turned pink indicating alkaline formation. It was then compared with standards. The catalase test indicated four positive and one negative where as in the oxidase test two were positive and two negative.

Usually the agricultural residues contain cellulose, hemicelluloses and lignin in the ratio of 4:3:3.

Table 5 shows the morphological and biochemical results of the five bacterial isolates from press mud

**Table 5 Morphological and Biochemical Characterisation of farmyard waste isolates**

Isolate No.	Morphological Characterisation		Biochemical Characterisation									
	Culture Characterisation of Colonies	Gram Staining	Citrate Test	MR Test	VP Test	Indole Test	Triple Sugar Iron Test				Catalase Test	Oxidase Test
							H <sub>2</sub> S	Gas	Butt	Slant		
P1	Red	Gram(-)	+	+	-	-	-	-	+	+	+	-
P2	Transparent White	Gram(-)	+	-	-	+	-	-	-	-	+	+
P3	Transparent White	Gram(-)	-	-	+	-	-	-	-	-	+	+
P4	Lemon Yellow	Gram(+)	-	+	-	-	-	-	+	+	+	+
P5	Yellow	Gram(+)	-	+	+	-	-	-	+	-	+	+

"+" = Present. "-" = Absent

Three isolates were gram negative and remaining two were gram positive. The citrate test three isolates were negative and two was positive. Three isolates shown positive and remaining two were negative for M R Test and V P test indicated two to be positive and three negative. Indole test gave only one positive result by the appearance of red ring while others showed negative results. The TSI results shown negative for all the five isolates thus indicating no gas formation. Among the five isolates three changed their butt to yellow colour indicating acid formation. The slants of two isolates turned to yellow while all other turned pink indicating alkaline formation. All the five isolates were positive for catalase test whereas four were positive and one negative in oxidase test.

Table 6 shows the morphological characteristics and biochemical test results of the two bacterial isolates from bagasse

**Table 6 Morphological and Biochemical Characterisation of farmyard waste isolates**

Isolate No.	Morphological Characterisation		Biochemical Characterisation									
	Culture Characterisation of Colonies	Gram Staining	Citrate Test	MR Test	VP Test	Indole Test	Triple Sugar Iron Test				Catalase Test	Oxidase Test
							H <sub>2</sub> S	Gas	Butt	Slant		
B1	Transparent	Gram(-)	+	-	-	-	-	-	-	-	+	+
B2	Transparent White	Gram(-)	+	+	+	-	-	-	+	-	+	-

"+" = Present. "-" = Absent

All the bacteria isolated were gram negative. The citrate test indicated both the cultures to be positive. The MR test show one positive and one negative. The VP test also showed one positive and one negative. No positive results were observed in Indole test. No isolates produced H<sub>2</sub>S and the gas formation was not observed in TSI test. The butt of one of the isolates changed to yellow colour and other was pink. The slants of both isolates were negative. The catalase test gave positive results for both isolates whereas one was positive and one negative of oxidase test.

Table 7 shows the morphological and biochemical characteristics of two bacterial isolates from mushroom waste, Gram staining results in one being positive and the other being negative

**Table 7 Morphological and Biochemical Characterisation of farmyard waste isolates**

Isolate No.	Morphological Characterisation		Biochemical Characterisation									
	Culture Characterisation of Colonies	Gram Staining	Citrate Test	MR Test	VP Test	Indole Test	Triple Sugar Iron Test				Catalase Test	Oxidase Test
							H <sub>2</sub> S	Gas	Butt	Slant		
M1	Transparent	Gram(-)	-	+	-	-	-	-	+	-	+	+
M2	Transparent White	Gram(+)	+	-	-	-	-	-	-	-	+	+

"+" = Present. "-" = Absent

The gram negative isolate showed negative to citrate test whereas the gram positive isolate showed positive. MR test results also gave one positive and one negative result. VP test also show negative results for both the bacteria. Indole test showed both as negative. TSI test resulted in lack of H<sub>2</sub>S production and lack of gas production in both bacterial isolates. Both isolates changed their butt to yellow and slants changed to pink colour. Both isolates showed positive results in catalase and oxidase test.

**Table 8. Culture characteristics of fungal isolates**

S.No.	Sample isolates	Cultural characteristics
1.	B1	Dull green colour colonies
2.	B2	Dark green colour Colonies
3.	P1	Green Colour colonies
4.	P2	White colour Colonies
5.	F1	White Colour colonies turning green
6.	F2	Yellow colour colonies
7.	F3	Greenish grey colour colony
8.	F4	Green colour colonies
9.	M1	Green colour colonies
10.	M2	Dark green colour colonies
11.	M3	Dark green colour colonies
12.	M4	Green colour colonies
13.	M5	Green colour colonies

Thirteen fungal isolates were identified, among them five were green coloured colonies, one was dull green, three were dark green, one white, one white turning into green, one yellow colony and the other was greenish grey colour colony.

The results of *Actinomycetes* species isolated from different organic wastes were based on their morphological characteristics as shown in Table 9.

**Table 9 Culture characteristics of Actinomycetes Isolates**

S.No.	Sample isolates	Cultural characteristics
1.	B1	Transparent colonies
2.	B2	Transparent Colonies
3.	P1	White with light orange colour colonies
4.	P2	Greyish black colour Colonies
5.	F1	Transparent white Colour colonies
6.	F2	Transparent dull white colour colonies
7.	F3	Transparent white colour colonies
8.	F4	Dull white colour colonies
9.	F5	Pure transparent colonies
10.	M1	Greyish white colour colonies
11.	M2	Isolated grey with black amorphous colonies

**Table 10 The different microbial isolates from different organic wastes**

S.No.	Name of the Sample Isolates	Name of the Bacterial Culture	Name of the Fungal Culture	Name of the Actinomycetes culture
1.	B1	<i>Pseudomonas Sp.</i>	<i>Penicillium sp.</i>	<i>Thermoactenomyces sp.</i>
2.	B2	<i>Bacillus sp.</i>	<i>Trichoderma sp.</i>	<i>Thermoactenmycetes sp.</i>
3.	M1	<i>Bacillus sp.</i>	<i>Trichoderma sp.</i>	<i>Streptomyces sp.</i>
4.	M2	<i>Pseudomonas sp.</i>	<i>Rhizopus sp.</i>	<i>Nocardia sp.</i>
5.	M3	-	<i>Mucor sp.</i>	<i>Streptomyces sp.</i>
6.	M4	-	<i>Trichoderma</i>	-
7.	M5	-	<i>Trichoderma</i>	-
8.	F1	<i>Bacillus sp.</i>	<i>Alternaria sp.</i>	<i>Thermoactenomyces sp.</i>
9.	F2	<i>E.coli</i>	<i>Aspergillus sp.</i>	<i>Streptomyces sp.</i>
10.	F3	<i>Enterobacter sp.</i>	<i>Aspergillus sp.</i>	<i>Streptomyces sp.</i>
11.	F4	<i>Pseudomonas sp.</i>	<i>Aspergillus sp.</i>	<i>Thermo actenomyces sp.</i>
12.	F5	<i>Klebsiella sp.</i>	-	<i>Thermo actinomycetes sp</i>
13.	P1	<i>Serratia sp.</i>	<i>Aspergillus sp.</i>	<i>Micro monospora sp.</i>
14.	P2	<i>Pseudomonas sp.</i>	<i>Alternaria sp.</i>	<i>Streptomyces sp.</i>
15.	P3	<i>Pseudomonas sp.</i>	-	-
16.	P4	<i>Cellulomonas sp.</i>	-	-
17.	P5	<i>Microspora sp.</i>	-	-

A total of 12 *Actinomycetes* isolates were identified. Among them two were transparent, three were transparent white and the remaining were one each of white with light orange colour, greyish black, dull white, pure transparent, greyish white, grey with black amorphous and greyish white colonies.

Table 10 shows the microorganisms that were isolated from the different organic waste.

The following are the bacterial isolates from the organic waste of *E.coli*, *Klebsilla*, *Pseudomonas*, *Bacillus*, *Micrococcus* and *Serratia* and also shows the fungal isolates such as *Mucor*, *Rhizopus*, *Trichoderma*, *Penicillium* and *Aspergillus*. The *Actinomycetes* species such as *Micromonospora*, *Thermoactinomycetes*, *Streptomyces* and *Nocardia* were isolated. In our study we have isolated *Penicillin* species and *Trichoderma* species from the bagasse. Dorothy *et al* (1985) stated that cellulolytic bacteria include aerobic species such as *Pseudomonas* and *Actinomycetes* are more effective in the degradation of cellulose rich organic waste such as mushroom waste and farm yard waste [6]. In the present study we have isolated *Pseudomonas* species from naturally, partially decomposed cellulose rich materials such as bagasse, press mud, farm yard waste and mushroom waste. In the present study the occurrence of *Pseudomonas* species and *Enterobacter* in the farm yard, mushroom waste and press mud was observed. This investigation was done in order to isolate, characterise and identify the decomposing microorganisms such as bacteria, fungi, *Actinomycetes* from the various organic waste and to understand their capacity so that these organisms could be utilised as effective biodegrading agents in the agro and organic wastes.

## DISCUSSION

Organic waste samples were collected from agro-based industries and from the agriculture fields consisted of both biotic and abiotic components with different physio-chemical parameters such as pH, moisture, carbon content, nitrogen content etc. and the chemical components such as cellulose, hemicelluloses, lignin and starch etc. These samples were found to have various species of organisms which have degrading properties. Bagasse, Press mud, Mushroom waste and farm yard waste samples, were collected and cultured to isolate bacteria, fungi, and *Actinomycetes* and by the pure cultures organisms and colonies are separated. From our study it was understood that bacteria, *Actinomycetes* and fungi spp. were present in degrading organic waste and are considered to be responsible for degradation. Hence it is of interest to investigate the physicochemical and morphological characteristics of the different isolates obtained from agro-industrial waste materials.

Gold and Alic, 1993, have reported the ability of white rot fungi *Phanerochaete chrysosporium* and *Trametes versicolor* in degrading lignin, and other wood polymers like cellulose, hemicelluloses and lignin due to the presence of Lignin peroxidase, Manganese peroxide and laccase (Mougin *et al*, 1994) [7, 8]. White rot fungi and brown rot fungi are efficient in degradation of lignin (Kirk, 1971) [9]. *Thermonospora fusca* can degrade cellulose (Crawford, 1978) and the combination of *Pleurotus sp.* and *Pseudomonas sp.* were able to degrade cellulose faster (Crawford 1978; Thilagavati *et al*, 2006) [10, 11]. Inoculation of cellulolytic microorganisms such as *Actinomycetes* and fungal strains rapidly decrease the hemicelluloses and cellulose content in paddy straw composting. *Streptomyces* strains decompose lignocelluloses [12]. Kurt and Buyukalaca, 2010, have studied the yield performances and changes in enzyme activities of *Pleurotus spp.* on different agricultural wastes [13]. The laccase enzyme productivity by *Pycnoporus sanguineus* on selected agro waste by solid state fermentation was reported by Vikneswary *et al*, 2006 [14]. Similarly Erden *et al*, 2009, have reported a new and different lignocellulolytic material from Turkey for laccase and manganese peroxide production by *Trametes versicolor* [15]. Our study indicated as shown in Table 2, the fungal and *Actinomycetes* colonies were found to be more in mushroom and farmyard waste whereas the bacterial colonies were more in press mud and farm yard waste. But in all the four samples fungi, *Actinomycetes* and bacteria were invariably present in various concentrations. It was observed that the physiochemical parameters and chemical components of different organic wastes are shown in Table 3, which determined the number of microorganisms. It was observed that the farm yard waste which had an alkaline pH of 7.3, high moisture content, high carbon content and high hemicelluloses content supported the growth of maximum number of all the three types of microbes. Further the moderate number of colonies in bagasse and mushroom wastes could be attributed to equal nitrogen content, very high or low hemi cellulose and starch content.

## CONCLUSION

Further analysis of the individual activities of each of three microbial species and on the different agro-based wastes are being carried out in our laboratory to specifically identify their role in the biodegradation and its environmental impact.

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