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ISOLATION AND CHARACTERIZATION OF EFFECTIVE BACTERIA FOR SOLID WASTE DEGRADATION FOR ORGANIC MANURE

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ABSTRACT

The present investigation was conducted to find out the utilization of effective bacteria for solid kitchen waste degradation as organic manure or compost. Ten garbage samples were collected from different potential habitat of Kushtia and Jhenaidha regions of Bangladesh for isolating the effective garbage degrading bacteria. Three bacterial sample strains named as NAIUL, BCDump and BCSS (2) were isolated and cultured on Nutrient Agar and Czapek-Dox-Agar media. Characterizations of these strains were also studied by visual observation of colony, microscopic observation and biochemical tests identified the specific bacteria namely *Xanthomonas* spp., *Bacillus* sp., and *Pseudomonas* sp. from NAIUL, BCDump and BSS (2) strains, respectively. Changes of color, odor, weight loss, volume loss, temperature and pH of decomposing garbage were noted for selecting the most effective strain. The highest decomposition activity in term of weight loss and volume loss was shown by suspension of BCDump sample strain. To observe the effect of various additives on decomposition, various concentrations (5 %, 10 % and 15 %) of sucrose and molasses solution were used along with bacterial suspension. It was observed that 15% molasses treatment showed the best performance over the other treatments using BCDump sample strain. Decomposed garbage was used as compost or organic manure to observe their effects on biomass production (fresh weight basis) of potatoes (*Solanum tuberosum*). Garbage decomposed by all strains was used as organic manure. From the present investigation, it can be concluded that useful bacteria might be isolated from the surrounding environment for friendly bioconversion of solid organic waste.

KEYWORDS: bacteria, characterization, soil waste, organic compost

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1. INTRODUCTION

The word "waste" refers to useless, unwanted or discarded materials which are no longer considered of sufficient value and are thrown away by the possessor [1]. Wastes include solids, liquids and gases, among them solid or semi solid forms are called "solid waste". Everyday, a huge quantity of waste generate in all the developed and developing countries [2]. In rapidly growing cities of the developing world, urban solid waste management is currently regarded as one of the most immediate problem. Various types of waste are causing adverse effect on living organisms and environment. As a result, human and animal diseases occur, the air and soil environment are spoiled and the entire natural ecosystem balance is disturbed. Previous studies showed that on an average, each person in urban areas produces half a kilogram of garbage each day [3, 4]. According to EPA in 1989, only 10 % of this MSW (Municipal Solid Waste) is being recycled, while 80% goes into landfills, and 10 % is incinerated. In low-income countries, the solid waste generation rate is average only 0.4 to 0.6 kg/person/day, as opposed to 0.7 to 1.8 kg/person/day in fully industrialized countries [5-7]. Every day huge quantities of waste materials are generated in all cities and municipal areas of Bangladesh. In urban areas, solid waste has a very high organic content that ranged from 70- 85 %. The solid waste generation in urban areas in Bangladesh is growing with the growth of population as well as per capita GNP [8]. Only Dhaka City Corporation estimates around 3000 tons of garbage per day from its 10.07 million populations having an area of 325 square km. Per capita garbage generation per day varies from 0.35 kg to 0.4 kg in Dhaka [4]. The total generated waste is collected efficiency by formal system. About 10-15 % is recycled by formal system and 35-50 % is self disposable or illegal dumped (uncollected) [9]. Thus, under proper conditions this biodegradable fraction could be composed or co-composted for beneficial use as soil conditioners and biofertilizers [10].

The microbial population of soils is made up of five major groups including bacteria, actinomycetes, fungi, algae and protozoa, and among these groups, bacteria are the most abundant group [11] and the most important microbe for decomposing waste. Bacteria use wastes for their own metabolism and finally they produce some simple and useful compounds which are important for soil health, plant growing and over all to keep well balance of natural ecosystem. Composting is the controlled biodegradation or transformation of organic material, usually under aerobic conditions by which a material is transformed into an end product which is stable and soil like material called compost.

Number of microbes along with rodents and insects play a vital role for solid waste degradation. Among them, bacteria play the most important role and therefore, the effective bacteria can be employed for planned decomposition of solid organic waste. Considering tremendous importance of garbage decomposition, this investigation was undertaken in order to develop a reproducible protocol and to search for more active decomposer bacteria, which might be successfully and effectively degrade the organic wastes and produce useful components for plant nutrition.

2. MATERIALS AND METHODS

2.1 Sample collection

Ten samples (soil/humus) were collected during 26 May, 2005 from ten different areas of Kushtia and Jhenaidha regions of Bangladesh. Each sample (100 g) was collected in separate sterile screw cap tube, stored at 4ºC and marked according to their source and location as IUL (Garbage, in front of library, Islamic University Campus, Bangladesh), Dump (Garbage, behind Begum Khaleda Zia Hall, Islamic University Campus, Kushtia), SS(2) (Soil surface, pond side of Bangabandhu Sheikh Mujib Hall Islamic University Campus, Bangladesh), NPTQ (Wet soil, drain side of North Para Teachers Quarter's, Islamic University Campus, Bangladesh), DR (Drain side

garbage, Begum Fazilatunnessa Hall, Islamic University Campus, Bangladesh), SS (Soil surface, behind the Science Faculty of Islamic University Campus, Bangladesh), DRS (Wet soil of drain, right side of Commerce Faculty of Islamic University Campus, Bangladesh), ZD (Wet soil of drain, behind Ziaur Rahman Hall of Islamic university campus, Bangladesh), SG (Garbage, Sheikh Para, Jhenaidah, Bangladesh), and SLA (Refuse, slaughter house, Sheikpara, Bangladesh). Of these samples, three bacterial samples such as IUL, Dump and SS(2) were showed better growth on their favorable medium; IUL showed on Nutrient Agar medium (NA), Dump showed on Basic Czapek-Dox-Agar medium (BCDA) and BCSS(2) showed on Basic Czapek-Dox-Agar medium (BCDA) and therefore, these three samples were marked as NAIUL, BCDump and BCSS(2), respectively.

2.2 Culture method

Sterile dilution blanks were marked sequentially from 10^{-2} to 10^{-6}. Soil (1 g) was added to the 10^{-2} dilution blank and shaken vigorously for at least 1 minute. The dilute was then sedimented for a short period. One ml from this dilution was transferred to the 10^{-3} tube. Using a fresh sterile pipette, 1ml from the 10^{-3} dilution was transferred to the 10^{-4} dilution blank for each succeeding step, then from the 10^{-4} to the 10^{-5}, then from the 10^{-5} to the 10^{-6}. From each dilution tube, 1 ml of dilution fluid was transferred into different culture media (NA, BCDA and PDA) and incubated at 37°C for 24 h. After successful growth of microorganisms, the colonies were counted using a colony counter (Yc-2A, Prma optical works Ltd, Japan) and colony forming unit (g or ml) and the results per dilution count were recorded. All the plates were incubated at room temperature until next period (2 days) for successful growth observation. The viable titer was then calculated and made a circle on the back of each plate and a number was assigned. Each colony morphology e.g., size, shape, margin, elevation, consistency, color, transparency was determined. Gram stain was performed to observe the cellular morphology and Gram reaction of the bacteria.

2.3 Pure culture

One single colony was identified and re-streaked as a primary inoculant on the surface of a nutrient agar plate and Basic Czapek-dox agar (BCDA) medium. The plates were then incubated at 30°C or room temperature. Pure cultures were checked from nutrient agar plates and Basic Czapek-dox agar (BCDA). Gram stain was determined to re-check the identical cell morphology and gram reaction comparing to the original colony. At this point, another nutrient agar plate and Basic Czapek-dox agar (BCDA) were re-streaked with the correct colony, and pure cultures were re-checked using the above steps. After achieving a pure culture, the same colony was streaked onto a nutrient agar and BCDA slant. These cultures were incubated for 1 day in the refrigerator. The isolates from soil were used for further experiments.

2.4 Biochemical tests

2.4.1 Catalase test

To detect soil bacteria, 2-3 ml of the hydrogen peroxide solution was poured in to a test tube. Using a sterile wooden stick, several colonies of the test organisms were removed from nutrient agar plate and Basic Czapek-dox agar (BCDA) medium and immersed in the hydrogen peroxide solution. Bubbles formation was then observed.

2.4.2 Lactose and mannitol fermentation test

This test is used to differentiate the microorganisms fermenting carbohydrate (such as lactose and mannitol).

2.4.3 Voges Proskauer test

To detect the production of acetylmethylcarbinol acetoin, a natural product formed from pyruvic acid in the course of glucose fermentation. The buffered glucose broth together with the organism was inoculated and incubated at 37°C for 3 days. Approximately 3 ml of alpha naphthol was added followed by 1 ml of 40 % KOH and mixed well for 30 minutes. For the result, pink solution means VP (+) and no change means VP (-).

2.4.4 Urease test

To detect soil bacteria, a dense "milky" suspension of the test organism was prepared in 0.25 ml physiological saline in a small tube. A urease tablet was added into the tube and incubated at 35-37 ^{o}C for up to 4 h or overnight. Color change of the test organism was observed.

2.4.5 Indole test

Indole test was done for detecting the soil bacteria. The test organisms were cultured in tryptophan containing medium in a bijou bottle with 3 ml of sterile tryptone water. The medium was then added with 0.5 ml Kovac's reagent (4p-dimethylamino-benzaldehyde) with gentle shaking and the color was observed.

2.4.6 Citrate utilization test

Citrate utilization test was based on the ability of an organism to use citrate as its only carbon source and ammonia as its only nitrogen source. If citrate utilization test is positive, the media will turn from green to blue.

3. RESULTS AND DISCUSSION

The isolation and characterization of bacterial strains from different places of Kushtia and Jhenaidha regions were undertaken in this study. Bacterial growth depends upon various physiochemical conditions such as media, pH, temperature, incubation period, carbon source, etc. Bacteria can grow in a wide range of moisture level. In this study, it was found that the range of moisture content of collected samples varied from 25.09 to 78.19 %. Maximum moisture content (78.19 %) was obtained from NAIUL and BCDump samples, while minimum moisture content (25.09 %) was noted from BCSS (2) sample.

Bacterial population of various soils is closely correlated with their moisture content. The maximum bacterial density is found in regions of fairly high moisture content and the optimum level for the activities of aerobic bacteria often is a 50 to 75 % of the soil moisture holding capacity [11]. Numbers of the genera *Pseudomonas, Achromobacter* and *Bacillus* are found in most aerobic soils; where conditions are anaerobic and moist *Clostridium* will occur. *Actinomycetes* showed a similar quantitative increase under such conditions [12].

In the present investigation, the growth of isolated strains was observed in various growth media like potato dextrose agar (PDA), nutrient agar (NA) and Czapek-Dox- agar (both acidic and basic). It was observed that the basic Czapek-Dox-agar (BCDA) was suitable for massive growth of BCSS (2) and BCDump strains (Table 1) and nutrient agar (NA) medium was suitable for the massive growth of NAIUL strain (Table 2).

Table 1 Effect of different pH on the growth of isolated strains in BCDA[a] medium

pH of medium	Strains	Incubation period					
		6 h	12 h	24 h	36 h	48 h	72 h
7.1	BCSS(2)	-	+	+++	+++	+++	+++
	BCDump	-	+	+++	+++	+++	+++
	NAIUL	-	+	++	+++	+++	+++
7.6	BCSS(2)	-	+	++++	++++	++++	++++
	BCDump	-	+	++++	++++	++++	++++
	NAIUL	-	-	+	++	+++	+++
9.1	BCSS(2)	-	-	+	+	++	++
	BCDump	-	-	+	+	+	+
	NAIUL	-	-	-	-	+	++
10.6	BCSS(2)	-	-	-	-	-	-
	BCDump	-	-	-	-	-	-
	NAIUL	-	-	-	-	-	-
12.10	BCSS(2)	-	-	-	-	-	-
	BCDump	-	-	-	-	-	-
	NAIUL	-	-	-	-	-	-

[a]BCDA = Basic Czapek-Dox-Agar; - = No growth; + = Poor growth; ++ = Moderate growth; +++ = Good growth; ++++ = Massive growth

Table 2 Effect of different pH on the growth of isolated strains in NA[a] medium

pH of medium	**Strains**	**Incubation period**					
		6 h	12 h	24 h	36 h	48 h	72 h
4.2	BCSS(2)	-	-	-	-	-	-
	BCDump	-	-	-	-	-	-
	NAIUL	-	-	-	-	-	-
5.7	BCSS(2)	-	-	-	-	+	+
	BCDump	-	-	-	-	+	+
	NAIUL	-	-	-	-	+	+
7.2	BCSS(2)	-	+	++	++	+++	+++
	BCDump	-	+	++	++	+++	+++
	NAIUL	-	+	++	++++	++++	++++
8.7	BCSS(2)	-	-	++	++	++	++
	BCDump	-	-	+	+	++	++
	NAIUL	-	-	+	++	++	+++
10.2	BCSS(2)	-	-	-	+	+	+
	BCDump	-	-	-	+	+	+
	NAIUL	-	-	-	+	+	+

[a]NA = Nutrient Agar; - = No growth; + = Poor growth; ++ = Moderate growth; +++ = Good growth; ++++ = Massive growth

The pH is a key factor for growing bacteria in artificial media. In this study, the pH of two selected media (BCDA and NA) was optimized for culturing bacterial strains. The pH range of two media was found to be 7-8. NA and BCDA at pH 7.2 and 7.6, respectively were found to be suitable for the maximum growth of bacterial strains. From the results, it was found that the pH of the samples of BCDump, BCSS (2) and NAIUL strains were 7.79, 7.95 and 7.86, respectively and possibly for this reason, these strains were also found to grow well in *in vitro* condition at pH 7-8 in BCDA and NA. Bacteria can tolerate a soil reaction between pH levels 4 and 10, but the most favorable pH for the majority is just an alkaline side of neutrality. Bacteria such as *Thiobacillus thiooxidans* and *Acetobacter* sp. are capable of growing at the very low pH values between pH level 0 and 2 and some *Bacillus* sp. can grow at pH 11 [12]. *Thermoactinomycetes* grows only at temperatures between 50°C and 65°C and optimal growth occurs at pH 8 or 9, and is greatly depressed by reactions of around pH 5 [13]. *Mycobacterium tuberculosis* var. grows well but slowly (2-6 weeks) on a solid media such as coagulated egg, serum or blood or on glycerin agar at 37°C. *Vibrio*, *Streptococcus faecalis* and *Escherichia coli* also tolerate an alkaline reaction (pH 8-9) [13].

In this experiment, the bacterial cultures of three strains were incubated at different temperatures like 25, 29, 34, 37 and 40°C. The massive growth of all the strains was found in 37°C. The optimum temperature range for bacteria is from about 25-36°C. A great number of bacteria may grow quite well over the range of 10-40°C [14]. Sultana [18] observed that 33±4°C temperature was ideal for the growth of bacteria. Certain bacteria develop most vigorously at temperatures below 20°C. Thermophiles grow well at temperatures of 45-65°C and some thermophiles are in capable of multiplying below 40°C [11]. *Mycobacterium avium* grows well at 40°C whereas *M. chelonei* and *M. marinum* grow well at lower temperatures (18-30°C). *Lactobacillus* sp. grows best on tomato juice agar at 25 to 39°C. *Agrobacterium* sp. grows well on ordinary laboratory media at pH around 6.8 and at temperatures around 25 at 39°C [13]. *Streptococcus lactis* can be cultivated in sterile milk or on agar containing milk or whey or tomato juice at about 25°C. It grows best in the presence of glucose or lactose. It grows better at about 35°C than at 25°C [13].

The strains obtained in this study were incubated at different incubation periods (6, 12, 24, 36, 48 and 72 h). Incubation period of 24 h was suitable for well growth of BCSS (2) and BCDump strains, while NAIUL strain was found to be suitable with incubation period of 36 h. Coliform bacteria grow in the incubation period of 24±2 h and at 32°C and it shows good growth at 37°C for 48 h of incubation. Three methods were used (non-microscopic observation or visual observation, microscopic observation, and biochemical tests) to characterize the selected strains. In visual observation, it was found that after 24 h of incubation, the color of BCSS (2) was light orange, BCDump was white and NAIUL was light brown in their preferred medium (BCDA and NA). After 48-72 h of incubation, the color of BCSS (2) was orange, BCDump was yellow and NAIUL was brown. The colony type of BCSS (2) and BCDump strains were wet, and NAIUL was creamy. Staphylocoooi and Micrococci produce golden brown, yellow or white colony on ordinary media. Some enterococci, coryneforms and enterobacteria may produce black colonies on ordinary media. *Staphylococcus aureus* gives black, shiny, convex colonies on Baird-Parker medium [15].

Gram staining is an old and reliable method for observing the bacteria. Gram negative bacteria were decolorized by alcohol, losing the purple color of crystal violet. Gram positive bacteria did not decolorize and remained purple [16]. In the present investigation, all isolated strains were found to be Gram positive. All the isolated strains (BCSS (2), BCDump, NAIUL) were exhibited positive catalase test because they released oxygen gas from hydrogen peroxide (H_2O_2) by enzymatic degradation. Similar result was observed [17]. A number of biochemical tests have been performed to study the characteristics of bacterial strains (Table 3). Fermentation test is used to differentiate the microorganisms that ferment carbohydrate (such as lactose,

Table 3 Biochemical tests of some waste decomposing bacteria

Strains	Catalase	Lactose Fermentation Test	Mannitol Fermentation Test	Voges-Proskauer Test	Urease Test	Indole Test	Citrate Utilization Test	Identified Bacteria
NAIUL	Positive	Positive	Positive	Negative	Negative /Positive	Negative/ positive	Positive	*Xanthomonas* spp.
BCDump	Positive	Positive	Positive	Positive	Positive	Negative	Negative	*Bacillus* spp.
BCSS (2)	Positive	Positive	Positive	Positive	Negative	Negative	Negative	*Pseudomonas* spp.

mannitol). The isolated strains exhibited positive lactose and mannitol fermentation test because open and sealed tube produced yellow color.

In urease test, due to the formation of red pink color through the media, BCDump strain showed positive urease test and this strain was able to decompose urea to ammonia. BCSS (2) and NAIUL showed negative test because they did not produce red-pink color in this medium. Indole test demonstrates the ability of certain bacteria to split the amino acid tryptophan to indole which accumulates in the medium. NAIUL strain showed positive test and produced red color in media. BCDump and BCSS (2) strains showed negative test because they did not produce red color in media. Sultana [18] also observed the same result. BCDump and NAIUL strains showed positive test in hydrogen sulphite production test because they produced black color in media. However, BCSS(2) displayed negative test as this strain did not produce black color in media. The same result was observed by Sultana [18]. Citrate utilization test was based on the ability of an organism that uses citrate as its only carbon source and ammonia as its only source of nitrogen. NAIUL strain showed positive citrate utilization test because the medium turned from green to blue. BCSS (2) and BCDump did not turn the media to blue and therefore, they showed negative test (Table 4).

Table 4 Changes of temperature during garbage decomposition by bacterial suspension and different concentration of molasses solution at 3 day interval.

Days	Temperature (°C) of decomposition garbage by treating BCDump strain and molasses solution				Temperature (°C) of Decomposition garbage by treating BCSS(2) and molasses solution				Temperature (°C) of decomposition garbage by treating NAIUL strain and molasses solution			
	5%MS[a]	10%MS[a]	15%MS[a]	CT[b]	5%MS[a]	10%MS[a]	15%MS[a]	CT[b]	5%MS[a]	10%MS[a]	15%MS[a]	CT[b]
3	29	29	29	29	29	29	29	29	29	29	29	29
6	31	31	32	29	31	31	31	29	31	31	31	29
9	33	33	34	30	33	33	33	30	32	32	33	30
12	35	35	36	30	34	35	35	30	33	33	34	30
15	37	37	39	30	36	37	37	31	35	35	36	31
18	37	38	40	31	36	37	38	31	35	36	37	31
21	35	38	40	31	34	35	37	31	33	34	36	31
24	33	36	37	31	32	34	35	31	31	32	34	31
27	32	34	34	30	31	32	33	30	30	31	31	30

[a] MS, Molasses solution, [b] CT, Control.

Studies were conducted by various workers and the results showed the accumulation and separation of lower volatile fatty acids and VOC_s (Volatile Organic Compounds) during domestic waste decomposition [19]. As the waste is broken down by microorganisms (bacteria), the weight and volume of the matter decreases. In the present study of decomposition, we also observed that the weight and volume of treated garbage decreased because bacteria broke down the garbage and converted to simple molecules. For solid waste degradation (without additive), the highest weight loss in suspension treatment was recorded at 50.70 % after 30 days by treating with BCDump strain. In similar study, Hoitink [20] found the highest weight loss (25.76 %) using *Trichoderma* strain after 30 days. The highest volume loss in suspension treatment was found at 38.67 % for BCDump strain after 30 days. In case of pellet treatment, the highest volume loss was at 37.53 % in BCDump strain after 30 days.

Additives such sucrose can be used with garbage for decomposition which can increase the growth of bacteria [21]. It was observed that by treating with sucrose solution, the growth of bacteria was high and decomposition rate was also high. It seems that addition of sucrose solution to the garbage enhanced decomposition rate through increasing fermentation. In 5 %, 10 % and 15 % sucrose solution treatment (along with bacterial strain), the highest weight losses percentages were found 52.73 %, 55.32 % and 65.12 %, respectively in BCDump strain after 30 days of inoculation. Among those treatments, the highest weight loss was 65.12 % in BCDump strain along with 15 % sucrose solution. Similar trend was found for volume loss. The highest volume loss was found 51.34 % after 30 days of inoculation in BCDump strain along with 15 % sucrose solution. So it can be inferred that in sucrose treatment, the most effective strain was BCDump along with 15 % sucrose solution (in term of weight loss and volume loss). As sucrose treatment was effective for garbage decomposition, in later in order to reduce the cost, molasses were used as the alternative and cheap source of carbohydrate for decomposition of solid waste. In 5 %, 10 % and 15% molasses solution treatment, the highest weight loss percentages 56 %, 62.70 % and 80.24 %, respectively were found in BCDump strain after 30 days inoculation. Among those treatments, the highest weight loss 80.24 % was found in 15 % molasses solution in BCDump strain. Similar trend was found for volume losses. The highest volume loss was found 64.27 % after 30 days of inoculation in BCDump strain along with 15% molasses solution (in term of weight loss and volume loss). So it can be inferred that in molasses treatments, the most effect strain was BCDump along with 15 % molasses solution (in terms of weight loss and volume loss).

Again comparing the effect between different concentrations of sucrose and molasses treatment for degradation of solid waste with bacterial suspension, the highest weight loss was 80.24 % in 15 % molasses solution in BCDump strain, whereas the highest weight loss 65.12 % was found in 15 % sucrose solution in BCDump. Similar trend was found for volume losses. Therefore, it can be concluded that the garbage decomposition by 15% molasses was the most effective followed by15 % sucrose solution in term of volume losses, whereas bacterial suspension (without any additive) was able to lower volume change than additives treatments. In 15 % sucrose treatment, the highest volume loss (%) observed 51.34 % after 30 days of inoculation in BCDump strain and in 15% molasses solution, the highest volume loss (%) was 64.27% after 30 days of inoculation in BCDump strain. In similar study, Bari (2006) found 66 % weight loss for *Trichoderma* strain in 4 % concentration of glucose level. Harper *et al.* [22] reported that in composting pile, the final weight of 26.1 kg (58 lb) of afterbirth and dead piglets after composting for two weeks was only 3.1 kg (6.9 lb), and the remaining tissue was easily crumbled in the sawdust medium. In this experiment, the average daily weight loss was more than 6 % of the original animal mass. In the present investigation, it was observed that in term of weight loss (%) and volume loss (%), molasses treatment was better than sucrose treatment to decompose organic solid waste with bacterial suspension. It can be explained in this way that probably in molasses, there were some extra components (which were absent in sucrose) that could give extra nutrient to rapid growth of bacteria and to increase the number of bacterium cell. So the degradation process

with molasses solution was higher with higher number of bacteria than sucrose solution treatment. So, the growth of bacterial strain was the main factor to decompose garbage.

It was observed that the relationship between weight loss and volume loss was linear and no interaction was found between the two parameters. Both volume loss (%) and weight loss (%) were increased gradually with the progression of decomposition process and the result was same for each treatment of each strain. The weight loss (%) was higher than volume loss (%) in garbage decomposition in each treatment of each strain as shown in Figure 1.

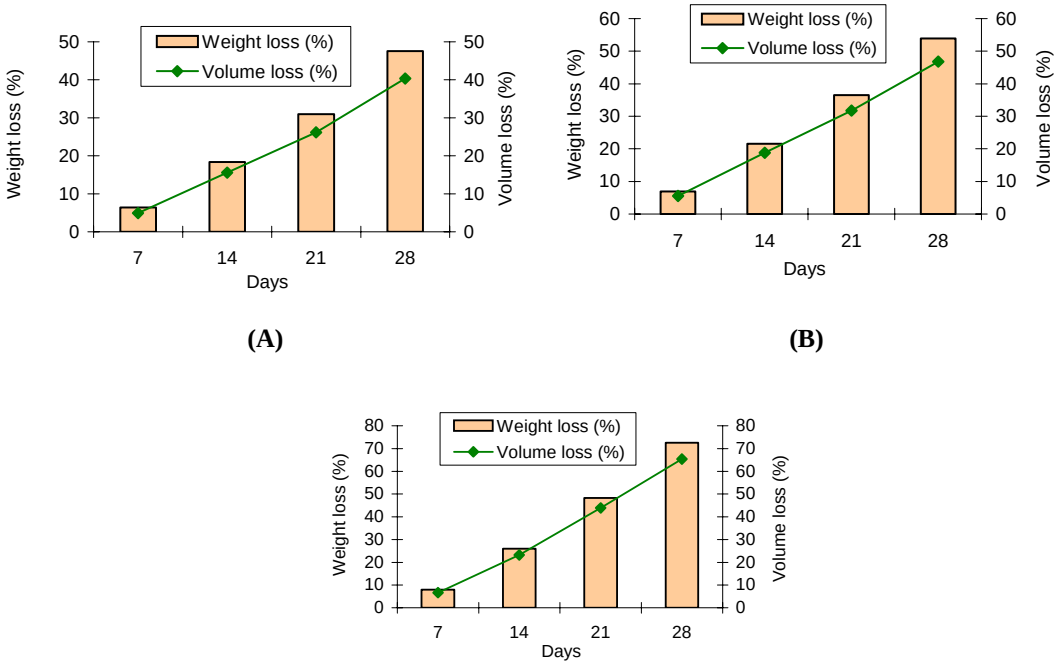

Figure 1 Weight loss (%) and volume loss (%) of decomposed garbage at 7 day interval using BCDump suspension and different concentrations of molasses solution. (A) BCDump + 5% molasses solution; (B) BCDump +10% molasses solution; (C) BCDump +15% molasses solution

When microbes degrade waste, heat is produced and temperature is increased with decomposition process [23]. In this study, we observed that in all cases temperature gradually increased after 4-6 days, and the temperature reached at the peak after 15-24 days and declined gradually after 28-30 days, and reduced to a level as it was in its initial stage. For decomposition (in composting method) of solid waste in *in vitro* condition by bacterial suspension and culture pellet, the highest temperature was 37°C after 15-18 days in suspension treatment of BCDump and BCSS(2) (Figures 2 and 3).

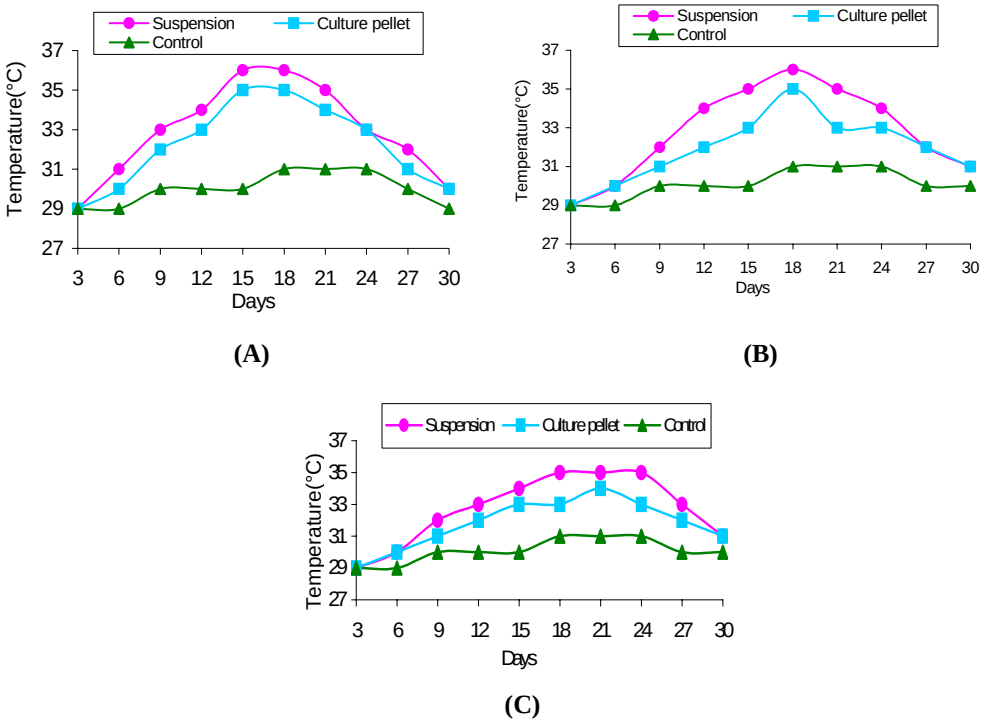

Figure 2 Changes of temperature (°C) during garbage decomposition at 3 days interval using suspension and culture pellet of the three strains. (A), BCDump strain; (B), BCSS (2) strain; (C), NAIUL strain.

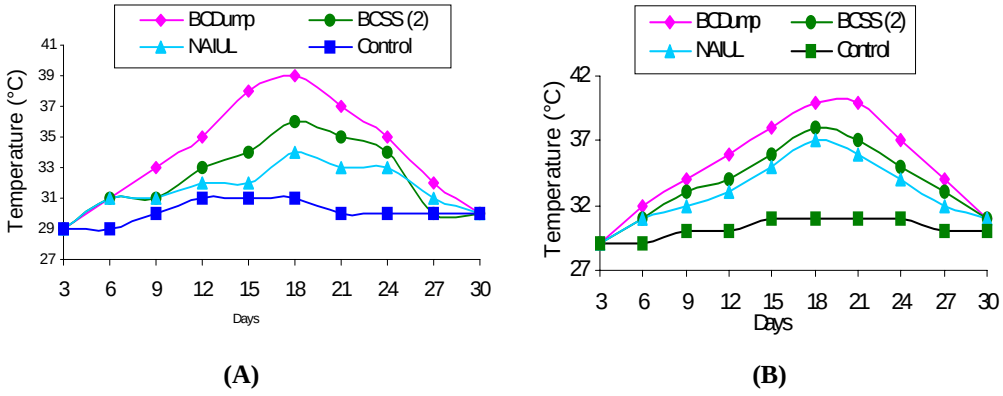

Figure 3 Changes of temperature (°C) during garbage decomposition at 3 days interval using the three strains separately along with the most effective concentration of additives. (A),Three strains +15% sucrose solution; (B),Three strains +15% molasses solution.

Using sucrose solution, the highest temperature was 39°C after 18 days in BCDump strain along with 15 % sucrose solution. For molasses solution, the highest temperature was 40°C after 18 days in 15 % molasses solution in BCDump strain. So it can be inferred that high decomposition rate is possibly associated with temperature. In this investigation, the weight and volume loss were the highest in which treatment the temperature was found to be the highest. In all cases, the temperature was high in BCDump strain and the weight loss and volume loss were also the highest in this strain. In this investigation, the initial pH of fresh garbage was 4.11 (acidic). In all cases, the pH of decomposed garbage (in suspension and pellet treatments, sucrose and molasses treatment) became alkaline from its initial acidic state, ranged from 7.31-11.06. The highest pH 11.06 was noted using 15% molasses solution decomposed by BCDump strain.

When bacteria are cultivated in a medium, pH 7 is very likely that this pH will change as a result of substance produced by the organism, which may be either acidic or basic [24]. It was reported that the pH varies with time during the composting process. The optimum pH range for most bacteria is between 6.0 and 7.5. During the initial period (first 2 to 7 days) pH drops to 6 or less and then begins to rise to about 7.5-8.5 for the remainder of the composting. In the initial stage of composting, organic acids are produced. For this acidic condition the pH value reduces in the initial days. But after some days, the temperature rises and the more decompositions of organic waste cause the rise in pH value. For decomposition of organic kitchen wastes by bacteria culture pellet, the odor was bad smell after 10, 20 and 30 days that indicate possible slow degradation of organic matters. In decomposition of solid waste by bacterial suspension, it was found that after 10 days the odor was bad smell and after 30 days there was no smell that indicates the possible complete degradation of organic kitchen wastes. In similar study, Rahman [25] reported that, in decomposition of solid waste by composting the odor was reduced. The study suggests that the useful bacteria might be isolated from the surrounding environment for friendly bioconversion of solid organic waste. To protect human health and natural environment from waste problems, the developed method of decomposition of waste as shown in this study is found to be very effective and useful. The study was conducted to observe the effect of fresh garbage, decomposed garbage and bacterial suspension on biomass production (fresh weight basis) of potato (*Solanum tuberosum*). For conducting the experiment, decomposed garbage (using bacterial suspension and 15 % molasses), fresh garbage, bacterial suspensions were used separately in the soil pot of potato. Control treatment (only garden soil) was also performed for each treatment. It was observed that decomposed garbage treatment (decomposed by BCDump suspension and 15% molasses) showed highest biomass production (65.61g), whereas fresh garbage and bacterial suspension exhibited 50.98 and 39.31 g, respectively. Decomposed garbage treatment (decomposed by BCSS (2) suspension and 15 % molasses) showed the highest biomass production (58.41g), whereas fresh garbage and bacterial suspension exhibited 48.45 and 37.07 g, respectively. It was observed that decomposed garbage treatment (decomposed by NAIUL suspension and 15% molasses) showed the highest biomass production (52.18 g), while fresh garbage and bacterial suspension exhibited 47.69 and 34.19 g, respectively. It was found that in all cases, addition of decomposed garbage in soil enhanced biomass production when compared with fresh garbage or bacterial suspension treatment. The highest biomass in term of fresh weight achieved using decomposed garbage by BCDump suspension along with 15 % molasses solution followed by BCSS (2) and NAIUL. In all cases, fresh garbage and bacterial suspension performed better than control (garden soil only).

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