Screening and Characterization of Soil Bacteria which Improves Crop Yield

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ABSTRACT
Municipal solid waste has 500 tons of waste per day with a composition of 60% of domestic waste and 40% of commercial waste. The composition of domestic waste were organic waste, green waste, mixed paper, plastic, textile, ferrous, glass, rubber and leather, and others. The degradation of organic wastes by the bacterial consortia is highly significant. This results in loss of potentially valuable materials that can be processed as fertilizer, fuel and fodder. The main aim of this project was to develop some successful bacterial consortium that can concomitantly degrade different components of the organic municipal solid wastes with the help of their enzymes in less span of time under natural conditions without producing any foul odor. In addition to that, the other important aim was to characterize the bacteria and their roles in plant growth promotion activity were observed in pot experiments.

Keywords: Organic Waste, Green Waste, Bacterial Consortium, Characterize the Bacteria, Plant Growth Promotion Activity.

I. INTRODUCTION

1.1 Municipal Solid Waste (MSW)
Municipal solid waste (MSW) generation is an issue of worldwide concern. The generators of municipal solid waste are broadly classified as residential, industrial, commercial, institutional, construction, demolition, municipal and agricultural types (Sehker and Beukering, 1998). Municipal solid waste is also generated by human and animal activities that are discarded as useless or unwanted waste. Municipal solid waste commonly known as trash or garbage (US), refuse or rubbish (UK) is a waste type consisting of everyday items we consume and discard. It predominantly includes food wastes, yard wastes, containers and product packaging, and other miscellaneous inorganic wastes from residential, commercial, institutional, and industrial sources. Examples of inorganic wastes are appliances, newspapers, clothing, food scraps, boxes, disposable tableware, office and classroom paper, furniture, wood pallets, rubber tires, and cafeteria wastes. Municipal solid waste does not include industrial wastes, agricultural wastes, and sewage sludge.

Economic development, urbanization and improving living standard in cities of developing countries have lead to increase in the quantity and complex composition of municipal solid waste. Management of municipal solid waste resulting from rapid urbanization has become a serious concern for government departments, pollution control agencies, regulatory bodies and public in most of the developing countries (Glawe et al., 2005; Erdogan et al., 2008).

Several other factors like education standard and infrastructure of the country have significant effect on municipal solid waste generation. The estimation and prediction of municipal solid waste generation play an important role in municipal solid waste management. The quantity of municipal solid waste in developing countries has been consistently rising over the years (Kansal, 2002). The municipal solid waste composition varies from place to place and also bears a rather consistent correlation with the average standard of living (Visvanathan and Trankler, 2003). The waste generated in the developing countries is similar in composition, the variation between regions being dictated by the climatic, cultural and industrial, infrastructural and legal factors.

Inefficient management and disposal of municipal solid waste is an obvious cause for degradation of environment in the developing countries. Ecological impacts such as land degradation, water and air pollution are related with improper management of municipal solid waste (Khajuria et al., 2008). In Asian developing countries, most of the municipal solid waste is dumped on land in more or less uncontrolled manner. Lack of sufficient awareness at the grassroots level of the waste generators add to the problem of littering. As a result there is a serious threat to public health due to environmental pollution.

Limited landfill space and resistance to siting such facilities has spurred consideration of new approaches to increase the longevity of landfills. Such efforts have included exploring methods to enhance degradation rates of municipal solid waste (MSW) and subsequently, to recover materials and landfill space.

1.2 Qualitative and Quantitative Analysis of MSW
There are many categories of MSW such as food waste, rubbish, commercial waste, institutional waste, street sweeping waste, industrial waste, construction and demolition waste, and sanitation waste. MSW contains recyclables (paper, plastic, glass, metals, etc.), toxic substances (paints, pesticides, used batteries,
medicines), compostable organic matter (fruit and vegetable peels, food waste) and soiled waste (blood stained cotton, sanitary napkins, disposable syringes) (Jha et al., 2003; Reddy and Galab, 1998; Khan, 1994). The quantity of MSW generated depends on a number of factors such as food habits, standard of living, degree of commercial activities and seasons. Data on quantity variation and generation are useful in planning for collection and disposal systems. With increasing urbanization and changing life styles, Indian cities now generate eight times more MSW than they did in 1947. Presently, about 90 million tons of solid waste are generated annually as by-products of industrial, mining, municipal, agricultural and other processes. The amount of MSW generated per capita is estimated to increase at a rate of 1–1.33% annually (Pappu et al., 2007; Shekdar, 1999; Bhide and Shekdar, 1998). A host of researchers (Siddiqui et al., 2006; Sharholy et al., 2005; CPCB, 2004; Kansal, 2002; Singh and Singh, 1998; Kansal et al., 1998; Bhide and Shekdar, 1998; Dayal, 1994; Khan, 1994; Rao and Shantaram, 1993) have reported that the MSW generation rates in small towns are lower than those of metrocities, and the per capita generation rate of MSW in India ranges from 0.2 to 0.5 kg/day. The quantity of MSW generated (CPCB, 2000) and the per capita generation rate of MSW (CPCB, 2004) is shown in Table 1.1 and Figure 1.1, respectively.

| S. No. | Name of the state     | No of cities | Municipal population | Municipal solid waste (t/day) | Per capita Generates (Kg/day) |
|-------|-----------------------|--------------|----------------------|------------------------------|-------------------------------|
| 1     | Andhra Pradesh        | 32           | 10,845,907           | 3943                         | 0.364                         |
| 2     | Assam                 | 4            | 878,310              | 196                          | 0.223                         |
| 3     | Bihar                 | 17           | 5,278,361            | 1479                         | 0.280                         |
| 4     | Gujrat                | 21           | 8,443,962            | 3805                         | 0.451                         |
| 5     | Haryana               | 12           | 2,254,353            | 623                          | 0.276                         |
| 6     | Himachal Pradesh      | 1            | 80,054               | 35                           | 0.427                         |
| 7     | Karnataka             | 21           | 8,283,498            | 3118                         | 0.376                         |
| 8     | Kerala                | 146          | 3,107,358            | 1220                         | 0.393                         |
| 9     | Madhya Pradesh        | 23           | 7,225,833            | 2286                         | 0.316                         |
| 10    | Maharashtra           | 27           | 22,727,186           | 8589                         | 0.378                         |
| 11    | Manipur               | 1            | 198,535              | 40                           | 0.201                         |
| 12    | Meghalaya             | 1            | 223,366              | 35                           | 0.157                         |
| 13    | Mizoram               | 1            | 155,240              | 46                           | 0.296                         |
| 14    | Orissa                | 7            | 1,766,021            | 646                          | 0.366                         |
| 15    | Punjab                | 10           | 3,209,903            | 1001                         | 0.312                         |
| 16    | Rajasthan             | 14           | 4,979,301            | 1768                         | 0.355                         |
| 17    | Tamil Nadu            | 25           | 10,745,773           | 5021                         | 0.467                         |
| 18    | Tripura               | 1            | 157,358              | 33                           | 0.210                         |
| 19    | Uttar Pradesh         | 41           | 14,480,479           | 5515                         | 0.318                         |
| 20    | West Bengal           | 23           | 13,943,445           | 4475                         | 0.321                         |
| 21    | Chandigarh            | 1            | 504,094              | 200                          | 0.397                         |
| 22    | Delhi                 | 1            | 8,419,084            | 4000                         | 0.475                         |
| 23    | Pondicherry           | 1            | 203,065              | 60                           | 0.295                         |

Source: Status of MSW generation, collection, treatment and disposal in class-I cities (CPCB, 2000).
Figure 1.1: Per capita generation rate of MSW for Indian cities (CPCB, 2000)

It can be seen from Table 1 and Fig. 1 that the per capita generation rate is high in some states (Gujrat, Delhi and Tamil Nadu) and cities (Madras, Kanpur, Lucknow and Ahmedabad). This may be due to the high living standards, the rapid economic growth and the high level of urbanization in these states and cities. However, the per capita generation rate is observed to be low in other states (Meghalaya, Assam, Manipur and Tripura) and cities (Nagpur, Pune and Indore).

1.3. MSW Characteristics and Composition

The composition and the quantity of MSW generated form the basis on which the management system needs to be planned, designed and operated. In India, MSW differs greatly with regard to the composition and hazardous nature, when compared to MSW in the western countries (Gupta et al., 1998; Shannigrahi et al.,1997; Jalan and Srivastava, 1995). The composition of MSW at generation sources and collection points was determined on a wet weight basis and it consists mainly of a large organic fraction (40–60%), ash and fine earth (30–40%), paper (3–6%) and plastic, glass and metals (each less than 1%). The C/ N ratio ranges between 20 and 30, and the lower calorific value ranges between 800 and 1000 kcal/kg. The physical characteristics of MSW in metrocities are presented in Table 2. It has been noticed that the physical and chemical characteristics of MSW change with population density. From Table 2, it is observed that the differences in the MSW characteristics indicate the effect of urbanization and development. In urban areas, the major fraction of MSW is compostable materials (40–60%) and inerts (30–50%). The relative percentage of organic waste in MSW is generally increasing with the decreasing socio-economic status; so rural households generate more organic waste than urban households. For example, in south India the extensive use of banana leaves and stems in various functions results in a large organic content in the MSW.
Table 1.2: Physical characteristics of MSW in Indian metrocities

| Source | Status of solid waste generation, collection, treatment and disposal in metrocities, (CPCB, 2000). 
Also, it has been noticed that the percentage of recyclables (paper, glass, plastic and metals) is very low, because of rag pickers who segregate and collect the materials at generation sources, collection points and disposal sites.

Solid wastes may seem to be the most ordinary forms of wastes, but they could be responsible for many problems such as spread of diseases and emission of green house gases. All these years, solid waste disposal was a neglected issue as these wastes were simply dumped on land in the outskirts of the city. This gave rise to problems like odors, flies, mosquitoes, groundwater pollution, emission of landfill gases etc.

A world which is on the rapid path of development has led to an increasing waste generating World. It has also posed a challenge not only with respect to treating and disposing waste properly but also to see this as an opportunity to derive useful products from it. With the severe energy crises in the World today, an attempt has been initiated to produce energy from solid wastes. This started with the concept of ‘Gobar Gas’ production. Later, newer concepts like fuel alcohol production, bio-hydrogen have also started coming up. Micro-organisms are the agents which bring about the conversion of these wastes into useful products like fuel gases, fuel alcohol and also compost which can be used as manure. The gases produced in landfills due to decomposition by anaerobic organisms also can be used as a source of energy. The major problem today in producing energy from waste is the cost factor. Efforts are being made to produce genetically modified organisms which will produce energy from wastes more efficiently and at least cost.

We live in a world where Solid Waste is no more a waste, but a storehouse of precious potential products. We can look forward to major breakthroughs in the field of Solid waste management using microorganisms.

1.4. Applications of Microbiology in Solid Waste Management

Microorganisms are omnipresent and are responsible for many good as well as bad things in our biosphere. They are present even in waste materials. These microorganisms carry out various biochemical processes to degrade waste materials. This process may be aerobic or anaerobic. Solid waste decomposition is carried out by bacteria which decompose complex organic materials to simple water soluble organic compounds. These are then converted to CO2 and H2O aerobically, or to CH4 anaerobically. Fungi are mostly aerobic and feed on decaying organic matter. Soil fungi play a vital role in stabilizing solid wastes in composting and landfilling processes by decomposing plant tissues like cellulose and lignin. Protozoa are predators on bacteria. They are found wherever bacteria are prevalent. Thus they help to maintain the equilibria of microbial flora in solid waste disposal systems.

1.5. Bacteria in Decomposition of Solid Organic Waste

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 [Alexander, 1961] 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. In the due course of these decomposition bacteria produces an essential metabolite of great economic value, known as enzyme.

1.6. Application of Bacteria in Biocomposting

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. The municipal solid waste in the urban centers is generated by domestic, commercial and industrial sources It contains mostly organic wastes that can be decomposed by composting. The bacterial conversion of the organics present in MSW in the presence of air under hot and moist conditions is called composting, and the final product obtained after bacterial activity is called compost (humus), which has very high agricultural value. It is used as fertilizer, and it is non-odorous and free of pathogens (Ahsan, 1999; Khan, 1994). As a result of the composting process, the waste volume can be reduced to 50–85%. The composting methods may use either manual or mechanical means and are accordingly termed as a manual or mechanical process. Manual composting is carried out in smaller urban centers and mechanical composting plants have been set up in big Indian cities. (Bhide and Shekdar,1998; Chakrabarty et al.,1995). 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 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.

The scope of this thesis was to screen potent bacterial isolates from municipal solid waste, producing industrial enzymes with high titer value and to optimize the environmental conditions during fermentation, developing an industrial media formulation using response surface methodology and evaluating the characteristic properties of the crude alkaline protease enzyme produced from the isolates. The bacterial strains were also used for application in degradation of municipal organic waste.

1.7. Compost

Composting is a spontaneous, biological decomposition process of organic materials in a predominantly aerobic environment. During the process of composting, bacteria, fungi and other microorganisms, including micro arthropods, break down organic materials to stable, usable organic substances called compost. It is a useful way of transforming organic waste into valuable organic matter for use as an organic amendment for soils (Gajdos, 1992). The bioconversion process is gradually emerging as a natural, promising, environment friendly and potential microbial process to degrade environmental contaminants (Colwell, 1994). It is seen as an environmentally acceptable method of waste treatment which uses naturally occurring microorganisms, to convert biodegradable organic matter into humus - like product. The process destroys pathogens, converts N from unstable ammonia to stable organic forms, reduces the volume of waste and improves the nature of the waste (Georgacakis et al., 1996 and Sequi, 1996).

Samarta and Patro (1996) established that organic farming was the backbone of sustainable agriculture, improved the soil health and the crops grown in rich organic manure, resist pest and disease attack. It was an ecologically sound and sustainable way of growing more food. According to Keener et al.(2000), the composting process composed of three steps (i) an initial mesophilic phase which lasted for 1-3 days, where mesophilic bacteria and fungi degraded simple compounds such as sugars, amino acids, proteins, etc., and led to an increase in temperature (ii) thermophilic phase, where thermophilic microorganisms degraded fats, cellulose, hemicelluloses and some lignin,during this phase the maximum degradation of the organic matter occurred together with the destruction of pathogens (iii) cooling phase, characterised by a decrease in temperature due to the reduction of the microbial activity associated with the depletion of degradable organic substrates, the composting mass was colonised by mesophilic microorganisms which were able to degrade the remaining sugars, cellulose and hemicellullose. The addition of municipal solid waste compost to agricultural soils has beneficial effects on crop development and yields by improving soil physical and biological properties (Zheljazkov and Warman, 2004). At present, the municipal solid waste composting is being encouraged in many countries of the world and researchers have experienced the benefits of using municipal solid waste compost in the field (Pokhrel and Virraghavan, 2005). Gajalakshmi and Abbasi (2008) defined compost as the stabilized and sanitised product of composting, which had undergone a rapid stage of
decomposition, beneficial to plant growth and had certain humic characteristics, a key for sustainable agriculture and resource management.

1.8. Evaluation of Compost Maturity and Stability

The most important factors affecting the successful application of compost for agricultural purposes are its degree of stability and maturity. The terms stability and maturity are both commonly used to define the degree of decomposition of organic matter during the composting process even if they are conceptually different. Compost stability is strongly related to the rate of microbial activity in compost and is evaluated by different respirometric measurements and/or by studying the transformations in the chemical characteristics of compost organic matter (Lasaridi and Stentiford, 1998; Pichler and Knabner, 2000). Compost maturity refers to the degree of decomposition of phytotoxic organic substances produced during the active composting stage. Application of unstable or immature compost may inhibit seed germination, reduce plant growth and damage crops by competing for oxygen or causing phytotoxicity to plants due to insufficient biodegradation of organic matter (Wu et al., 2000; Brewer and Sullivan, 2003 and Cooperband et al., 2003). A large variety of techniques have been reported for the determination of compost stability. Chemical parameters such as pH, electrical conductivity (EC), cation exchange capacity, dissolved organic carbon (DOC), C:N ratio and NH4 + to NO3 - have been applied as indicators of stability (Wang et al., 2004). Stability indicators based on the study of microbial biomass and its activity has also been proposed. Mondini et al. (2006) reported that microbial biomass could be used as a stability parameter in lignocellulosic waste composts because it clearly reflect the transformation of organic matter during the composting process. Respiration (CO2 evolution rate and/or O2 uptake rate) is a general measure of microbial activity, and it has been widely used to evaluate the stability of compost (Gomez et al., 2006). The ATP content and enzyme activities were also useful as indicators of compost stability (Tiquia et al., 2002 and Bitzer et al., 2006). Biological methods involving seed germination tests and plant growth bioassays have been used to evaluate the maturity of compost (Cooperband et al., 2003). Major evaluation methods of compost maturity and stability are grouped into three categories in terms of testing method: physical testing methods, biochemical test methods and plant test methods.

Review of Literature

Modern times have plagued the humanity with new problems due to industrialization and simultaneous population explosion. Rapid industrialization and population explosion in India has led to the migration of people from villages to cities, which generate thousands of tons of Municipal Solid Waste (MSW) daily. Municipal solid waste consists of household waste, construction and demolition debris, sanitation residue and waste from streets. This garbage is generated mainly from residential and commercial complexes. The MSW amount is expected to increase significantly in the near future as the country strives to attain an industrialized nation status by the year 2020 (Sharma and Shah, 2005; CPCB, 2004; Shekdar et al., 1992). Poor collection and inadequate transportation are responsible for the accumulation of MSW at every nook and corner. Solid waste generation in India was 229 million tons in 2001 and solid waste generation per capita per day in India ranged from 100 to 500 grams (Arrifa and Jayalakshmi, 2005). According to a study by The Energy and Resources Institute (TERI), the annual per capita municipal solid waste generation in India is projected to grow from 1 to 1.33 per cent, which would lead to a generation of over 260 million tones of waste by 2047 - a five fold increase over 1997 levels. It is further projected that an additional 1400 km2 of land is needed to dispose this waste, most of it in urban areas. Modern urban living brings on the problem of waste, which increases in quantity, and changes in composition with each passing day (Singh and Shekhawat, 2000). It has been estimated that overall municipal waste generated in urban centers, anywhere between 45 to 75 per cent constituted organic matter. It is also important to note that waste consumption varied significantly across areas of different economic levels of residents. The per capita solid waste reaching disposal sites in Bombay, Calcutta, Chennai and New Delhi ranges from 0.45 to 0.6 kilo gram per person per day. While in other Indian cities it is from 0.15 to 0.53 kilo gram per person per day (Manimozhi et al., 2006). It is established that about 500 grams of biodegradable kitchen waste is generated per day in a family consisting of four members. Each household produces solid wastes, which can be broadly classified as biodegradable (vegetable and fruit peels, leftover food etc) and non biodegradable (plastic bags, metal containers and glass bottles and hazardous or toxic wastes) (Venkataratnam, 2001). The household wastes include seeds of fruits, fruit peels and remnants, waste vegetables, wasted flower, rotten food, used tea dust, remnants of eaten food, egg shells, bones, paper, garden waste, glass, metals, used cosmetics, medicine bottles, rubber, leather, plastics, textiles etc (Karpagam, 2005). When organic waste decomposes in landfills and uncontrolled dumps, it produces methane, one of the major greenhouse gases contributing to climate change. The management of MSW is going through a critical phase, due to the unavailability of suitable facilities to treat and dispose of the larger amount of MSW generated daily in metropolitan cities. Unscientific disposal causes an adverse impact on all components of the environment and human health (Rathi, 2006; Sharholy et al., 2005; Ray et al., 2005; Iha et al., 2003; Kansal 2002; Kansal et al., 1998; Singh and Singh 1998; Gupta et al., 1998). Domestic waste from urban areas, without proper planning, is turning to be a problem...
unconquered (Lakshmanan, 2009). Organic wastes, which are produced in large quantities all over the world, create major environmental and disposal problems. These materials cause major unpleasant odour problems and use of large quantities of land for disposal and are often a source of contamination of ground water (Edwards and Bater, 1992; Kannaiyan and Lilly, 1999). Road side garbage from houses remains uncleared because its volume is more than what the corporation can handle. Rag pickers, stray animals and birds scatter the garbage, looking for items useful to them. This results in the familiar site around the street corners of most towns and cities (Mani, 1996). The waste accumulation has increased simultaneously with the rapid increase in residential colonies, fast food outlets, vegetable vendors, fruit shops and other customer outlets in the respective areas. Garbage is also dumped in huge plastic bags that obstruct the traffic. Viswananathan (2005) says that the amount of large solid refuse has been gradually increasing and its treatment and disposal has become a major social and environmental problem as well as a challenge. Open dumping of garbage facilitates the breeding of disease vectors such as flies, mosquitoes, cockroaches, rats, and other pests. Further, the poorly maintained landfill sites are prone to groundwater contamination because of leachate production (Maheswari, 2005).

The waste generated is consequently released into the nearby environment. Consequently, the management of the MSW needs to be revamped to accommodate the changes in the quantity and quality to ensure the longevity of the environment. Due to several legislative, environmental, economic and social constraints, the identification of most sustainable disposal route for MSW management remains an important issue in almost all industrialized countries (Adani et al., 2000). Generally, MSW is disposed of in low lying areas without taking any precautions or operational controls. Therefore, MSW management is one of the major environmental problems of Indian megacities. It involves activities associated with generation, storage, collection, transfer and transport, processing and disposal of solid wastes. But, in most cities, the MSW management system comprises only four activities, i.e., waste generation, collection, transportation, and disposal. The management of MSW requires proper infrastructure, maintenance and upgrade for all activities. This becomes increasingly expensive and complex due to the continuous and unplanned growth of urban centers. The difficulties in providing the desired level of public service in the urban centers are often attributed to the poor financial status of the managing municipal corporations (Mor et al., 2006; Siddiqui et al., 2006; Raje et al., 2001; MoEF, 2000; Ahsan, 1999). Agricultural application of MSW, as nutrient source for plants and as soil conditioner, is the most cost effective MSW disposal option because of its advantages over traditional means such as landfilling or incineration. According to Canellas et al., (2001) the use of MSW in agricultural lands can be justified by the need of finding an appropriate destination for waste recycling. Organic waste is a potential resource of both nutrients and organic matter that can be used to replenish the soils under pressure from traditional agriculture.

However, direct use of waste is impractical. Composting offers a method of biological stabilization that eliminates odour and pathogens and renders a product that is safe and pleasant to use (Divya, 2001). Compost acts as a natural fertilizer by providing nutrients to the soil, increasing beneficial soil organisms, and suppressing certain plant diseases, thereby reducing the need for chemical fertilizers and pesticides in landscaping and agricultural activities. The chief objective to compost organic wastes should not be for the disposal of solid organic wastes but to produce superior quality manure to feed our “nutrient-organic-matter-hungry” soils. Composting of wastes controls the pollution of soil and water and ensures the survivability and growth of fish, prawns and other organisms (Setua et al., 2008).

Characterization of Municipal Solid Waste

Municipal solid waste contains organic materials such as paper, food and yard waste and plastics. To be specific, MSW can be divided into six major chemical compound classes: non-cellulosic carbohydrates (hemicellulose, starch, and mono- and oligosaccharides), cellulose, proteins, lipids, lignin, and plastics (Pichler and Kögel-Knabner, 2000). Paper and paperboard products make up the largest component of MSW of the total 229.9 millions of tons of MSW generated in the U.S. in 1999, 38.1% were paper products. Although 41.9% was recovered by recycling, there were still 50.8 million tons of papers that were deposited in landfills (US EPA, 1999). Newsprint is produced from mechanical pulp with some chemical wood pulp. Only 7% of the raw materials are lost during the production of newsprint. Therefore, most compounds of the wood, lignin, cellulose, hemicellulose are present in newsprint. Office paper is made from a 5chemical pulp of high purity. Most of the lignin is removed during chemical treatment, leaving cellulose as the major component. The percentage of the cellulose varies depending on specific chemicals used (Calkin, 1957). Food waste comprised 12.1% of MSW; the major organic components of food waste are carbohydrate, protein and lipids (US EPA, 1999).
Table 2.1: The sources of municipal solid waste

| Sources            | Examples                                                                 |
|--------------------|---------------------------------------------------------------------------|
| Residential        | Single family homes, duplexes, town houses, apartments                     |
| Commercial         | buildings, shopping malls, warehouses, hotels, airports, restaurants, Institutional Schools, medical facilities, prisons. |
| Industrial         | Packaging of components, office wastes, lunchroom and restroom wastes (but not industrial process wastes) |

**Types of Municipal Solid Waste Source:**
(Tchobanoglous G, Kreith F, 2002)

**a) Organic Waste**

Organic waste is produced wherever there is human habitation. The main forms of organic waste are household food waste, agricultural waste, human and animal waste. The organic waste component is broken down by microorganisms to form a liquid leachate. This leachate presents serious hazards if it reaches water course or enters water table. In developing countries there are different approaches to deal with organic waste. In fact the word waste is an inappropriate term for organic matter which is often put to good use. The economies of most developing countries dictate that materials and resources must be used to their full potential and this has propagated a culture of reuse, repair and recycling.

**b) Biodegradation of Organic Waste by Bacteria**

It is the easily biodegradable fraction of immediate interest. The breakdown of the complex organic matter is accompanied by production of intermediates or end products. These processes are performed by bacteria efficiently and in the due course they produce several enzymes having application in diverse fields. The importance of knowing the process of decomposition carried out by bacteria in sewage solids digestion cannot be overemphasized. It has been recognized for several years that knowledge of changes taking place during decomposition would aid considerably in understanding digestion and might even change or improve existing methods. Such information may easily be the basis for improved solids decomposition by regulating conditions to favour the most desirable process. Further study on the product produced during the degradation of sewage solids might lead to some valuable byproduct recovery. Heterotrophic bacteria play a fundamental role in the biodegradation process. In spite of the increasing interest in this type of organic waste processing, few, if any, reviews on this subject have been published. O’Shaughnessy, (1914) reported the presence of bacteria of the coli and proteus group as well as denitrifying, fat splitting and cellulose organisms in a skudge after active digestion. Hotchkiss and Murray, (1923) observed bacteria capable of attacking complex protein, yielding soluble compounds. Gaud, (1924) observed that under anaerobic conditions, coliform was the largest group with, Salmonell typhosa and B.subtilis present in great numbers. Decomposition of litter can be largely performed by bacteria (Kominkova et al., 2000; Kuehn et al., 2000; Findlay et al., 2002; Anesio et al., 2003). In the process of decomposition bacteria produce different enzymes which play dominant role in the process of degradation as well as these enzymes have varied application and huge commercial values.

**c) Activities of Bacterial Enzymes in Organic Litter Decomposition**

The microbes need to produce extracellular enzymes to convert polymeric compounds—such as cellulose, hemicellulose, and lignin—into smaller molecules that can be assimilated (Chro’st, 1991). The most relevant enzymes from this aspect involve those that break down the plant fibers (cellulases, hemicellulases, pectinases, phenol oxidases) as well as enzymes important for microbial acquisition of nitrogen and phosphorus (peptidases, ureases, and phosphatases) (Sinsabaugh et al., 2002). The enzymes enabling the degradation and utilization of chitin (b-glucosaminidases and chitinases) may also cause lysis of fungal cell walls (degradation of fungal cell wall for fungal growth and/or fungal lysis by bacterial action, Wohl and McArthur, 2001). Due to the close connection between enzyme activity and degradation of different fractions of organic matter, enzyme assays can be used to estimate degradation rates of particulate and dissolved organic carbon in freshwater systems (Sinsabaugh et al., 1994). Production of cellulolytic and xylanolytic (hemicellulolytic) enzymes has been reported to occur also in bacteria (Robb et al., 1979; Tanaka, 1993; Sala and Gu’dé, 2004). In a few cases, bacteria (particularly actinomycetes) have been shown to contribute to  degradation of lignin, either as primary decomposers (Benner et al., 1984), or through mineralization of intermediate products released through fungal activity (Ru’ttimann et al., 1991). However, it is generally assumed that bacteria mainly decompose polysaccharides and polymeric compounds after the previous decomposition of high molecular and/or lignified compounds by fungi. Microorganisms surviving under extreme environments as well as enzymes (secreted from them in the process of this decomposition) stable and active under extreme conditions are both scientifically interesting and industrially significant for several useful applications. The biodegradation ability of bacteria

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reduces the waste by their effective utilization to produce useful enzymes.

d) Microbial Degradation of Solid Waste

Fungi are known agents of decomposition of organic matter in general and of cellulosic substrates in particular. Being efficient consumers of carbon, fungi build up much higher biomass than other microorganisms. The most commonly observed species of celluolitic fungi in composting materials are Aspergillus, Penicillus, Rhizopus, Fusarium, Chaetomium, Trichoderma, Alternaria, and Cladosporium (Ashraf et al., 2007). Some of the species of Paecilomyces and Sporotrichum have also been named as efficient degraders of lignocellulosic waste (Kapoor et al., 1978). Fungal species were found to be numerous during both mesophilic and thermophilic phases of composting. Their importance along with actinomycetes has been reported in composting, especially during the late curing stage (Finstein and Morris, 1975). Actinomycetes have an important role in carbon cycle because they are well adapted to the penetration and degradation of organics such as lignocelluloses. They appear during the thermophilic phase as well as the cooling and maturation phase of composting and can occasionally become so numerous that they are visible as a white film on the surface of the compost. The genera of the thermophilic actinomycetes isolated from compost include Nocardia, Streptomyces, Thermoactinomycetes, and Micromonospora (Strom, 1985). The group, Thermomonomosporas is of particular interest because they have the ability to produce thermostable cellulytic enzymes (Ball and McCarthy, 1989). The thermophilic filamentous bacterium Thermobifida fusca (formerly Thermomonospora fusca) is a major cellulose degrader in soil (Irwin et al., 1993). Fungi that have received considerable study with respect to their cellulytic enzymes and/or wood-degrading capability include Cladosporium, Fusarium, Geotrichum, Myrothecium, and Trichoderma (Deuteromycetes); Aspergillus, Bulgaria, Chaetomium, Helotium Paecilomyces and Penicillus (Ascomycetes); Coriolus, Phanerochaeta, Poria, Schizophyllum and Serpula (Basidiomycetes) (Carlile and Watkinson, 1997). The conversion of cellulosic mass on to fermentable sugars through biocatalyst cellulase derived from cellulytic organisms has been suggested as a feasible process and offers potential to reduce the use of fossil fuels and also reduce environmental pollution (Dale, 1999; Lynd et al., 1999). Yau and Murphy (1998) reported that the biodegradation of coir waste (coco peat) was enhanced by the addition of nitrogen fertilizer and inoculation with soft-rot fungus, Chaetomium globosum and it resulted in the reduction of hemicelluloses. The active component mediating the biodegradation and conversion processes during composting was the resident microbial community, among which fungi played a very important role. Therefore, optimization of compost quality was directly linked to the composition and succession of microbial communities in the composting process (Peters et al., 2000; Taiwo and Oso, 2004).

Microbial degradation of cellulosic materials is the result of synergistic action of enzymes such as endo-β-1,4-glucanase, exo-β-1,4-glucanase and β-glucosidase, all of which attack β-1,4-glycosidic bonds. Endo-β-1,4-glucanase and exo-β-1,4-glucanase both act upon cellulose to produce cellobiose as final product. Endo-β-1,4-glucanase cleaves randomly β-glycosidic bonds in β-1,4-glucan chains to produce free chain ends and exo-β-1,4-glucanases acts at chain ends by removing cellobiose units from the free chain. On the other hand, β-glucosidase hydrolyses cellobiose to glucose, reducing the inhibition effect of cellobiose on endoglucanase and exo-cellulohydrolase (Fernandez et al., 2002; Brienzo et al., 2008). Xi et al. (2002) studied the effects of complex microorganisms (Bacillus casei, Lactobacillus buchneri, Candida rugopelliculosa, Trichoderma and Whiterot fungi) in composting process of the municipal solid waste (MSW) and sludge.

The parameters analyzed were biomass, temperature, oxygen consumption, organic matter, and C:N ratio. The experimental results showed that the complex microorganisms were effective in composting organic matter, speeding up the composting process and changing into humus. Singh and Sharma (2003) accelerated the process of composting, a mixture of municipal solid waste (MSW) and horticultural waste, by inoculating different microflora viz. Pleurotus sajor-caju (fungus), Trichoderma harzianum (fungus) and Azotobacter chroococcum (bacteria) in different combinations into mixed solid waste. The compost produced was evaluated for nutrient levels and effects on mung bean (Vigna radiata) growth. A significant difference was observed in the quality of compost produced with the bioinoculants over control treatments. The combination of P. sajor-caju, T. harzianum and A. chroococcum produced the highest quality compost. The crop growth was enhanced significantly with the combination of P. sajor-caju, T. harzianum and A. chroococcum over other treatments.

Xi et al. (2005) used a method to improve the composting efficiency by seeding withmicrobial consortia A (a blend of Bacillus azotofixans, B. megaterium and B. maclaginosus), B, a blend of effective cellulytic strains, such as, Trichoderma koningii, Streptomyces cellulosa, and White-rot fungi; and C, a mixture of A and B. There were four runs: the control run (not inoculated), Run A, Run B and Run C. During the runs, parameters such as temperature, O2, CO2 and H2S emissions, and microbial density were investigated to analyze the efficiencies of inoculation during composting. The maximum oxygen uptake rates in the control run, Run A, Run B and Run C were calculated as 0.22, 0.32; 0.28 and 0.34 mol/h/kg respectively while the corresponding total O2 quantities accumulated were 511.18, 684.57, 659.74 and 778.47...
g/hkg. In addition, odorous gases were highly reduced by inoculation. It was concluded that inoculation, stabilized the composting products efficiently and improved the efficiency of the composting process.

Abdulla (2007) used three cellulolytic actinomycete isolates, of the genera *Micromonospora*, *Streptomyces* and *Nocardides*, as inocula in combination with different organic amendments for rice straw composting and incorporation into soil. Results demonstrated that composting with thermally-treated municipal sludge amendment and actinomycete inocula under aerobic conditions accelerated the straw decomposition process and reduced its bulk volume by 38.6 - 64 per cent after three months, compared to 13.6 per cent in uninoculated controls. The nutritional characteristics of the incorporated soil improved, particularly, in case of *Micromonospora* inoculation, as indicated by increase in organic matter to 34.9% and nitrates content to 0.59 mg/g, while those in the control reached 20% and 0.21 mg/g, respectively after the same incorporation time. Application of municipal sludge and *Micromonospora* combination might represent a rapid and environmentally friendly approach for disposal of rice straw. According to Steger (2007), actinobacteria were believed to play a major role in organic matter degradation and humification processes in composts.

The effects of different temperature regimes on the succession of actinobacteria populations during composting were investigated in a laboratory reactor. Phospholipid fatty acid (PLFA) was used to investigate quantitative changes in the overall microbial biomass and community structure and in the size of actinobacteria populations. The peak in total microbial biomass was roughly twice as high and delayed in trials where the maximum temperature was 40 °C, compared to those, where it was 55 or 67 °C. There was a shift from members of *Corynebacterium*, *Rhodococcus* and *Streptomyces* at the onset to species of thermotolerant actinobacteria in the cooling phase, e.g. *Saccharomonospora viridis*, *Thermobifida fusca* and *Thermobispora bispora*. In conclusion, temperature was an important selective factor for the development of actinobacteria populations in composts and they constituted a substantial part of the community in the later compost stages.

Garcia et al. (2007) tested the efficacy of three microbial isolates, identified as *Bacillus* shackletonii, *Streptomyces* thermovulgaris and *Ureibacillus thermosphaericus* as inoculants in composting processes in relation to their capacity to improve lignocellulose degradation. Different waste from agricultural activities were used as raw material for the heaps: pepper plant waste (PPW) as the main component and olive-oil mill waste (OMW), almond shell (AS), pruning waste (PW) and rice straw (RS) as additives. Cellulase was more extensively degraded than hemicellulose and lignin, although the use of inoculants (*B. shackletonii* and *S. thermovulgaris*) improved the action of the autochthonous microbiota just in the AS heapsA higher efficiency was observed for lignin, since lower concentrations of this polymer were detected in the inoculated heaps in relation to control heaps. *U. thermosphaericus* was the most efficient microorganism. The inoculation of this strain decreased the final lignin content in a range between 17.23 per cent and 24.34 percent. *S. thermovulgaris* and *B. shackletonii* led to a higher reduction of the lignin levels in the OMW (13 per cent) and PW heaps (14.25 per cent) than the control heaps. The composting process could therefore be improved by means of inoculation, if the microorganisms used for this purpose were appropriate for the characteristic of raw material.

According to Wei et al. (2007), Municipal solid waste (MSW) compost contained a significant amount of humic substances. In order to enhance degradation processes and the degree of composting humification, complex microorganisms (*Bacillus* casei, *Lactobacillus buchneri* and *Candida rugopelliculosa*) and lignocellulolytic (*Trichoderma* and White-rot fungi) microorganisms respectively inoculated in the composting process. During the MSW composting, humic acid (HA) was extracted and purified. Elements (C, N, H, O) and spectroscopic characteristics of the HA were determined using elementary analyzer, UV, Fourier transform infrared (FT-IR), and fluorescence spectroscopy. The elements analysis, UV, FT-IR and fluorescence spectra, all led to the same conclusion that inoculations with microbes led to a greater degree of aromatization of HA than in the control process (CK) with no inoculated microbes. This indicated that inoculation with microbes in composting would improve the degree of humification and maturation processes, in the following order: ligno-cellulolytic > complex microorganisms > CK and mixed inoculation of MSW with complex microorganisms and ligno-cellulolytic during composting gave a greater degree of HA aromatization than inoculation with complex microorganisms or ligno-cellulolytic alone. But compared to HA of soil, the HA of MSW compost revealed a lower degree of aromatization. Ajnavi (2008) recycled agricultural waste comprising of garden waste (grass cutting) and leaf litter by different fungi (*Aspergillus niger* FS1 and *Trichoderma reseei* MTCC-164) and using cow dung and Di Ammonium Phosphate - 0.1% as activators. The results indicated that organic carbon was decreased from 28.6 % to 14.5% and cellulose from 534.4 ppm to 115.1 ppm, with concomitant increase in available nitrogen content from 74.6 ppm to 356.5 ppm over 90 days of incubation. In garden waste, cellulose was decreased by 77% and in leaf litter (comprising mainly with *Bambusa vulgaris* leaves) by 70 % when the biomass was treated with fungal consortia with the addition of 0.1% DAP as activator. The results indicated that activators like DAP enhanced the rate of decomposition when agricultural waste were either treated with fungal consortia or cow
dung. Use of the fungi and DAP in the biodegradation of the agricultural wastes was the best option for rapid conversion of agricultural waste and more suitable in biocycling.

Gaind et al. (2009) composted poultry droppings, neem cake, castor cake, jatropha cake and grass clippings in the presence of a fungal consortium which included Aspergillus awamori, Aspergillus nidulans, Trichoderma viride and Phanerochaete chrysosporium. Evaluation of compost maturity showed that mixture of wheat straw, poultry dropping and Jatropha cake had the lowest C:N ratio of 10:1, the highest humic acid fraction of 3.15%, the lowest dehydrogenase activity and a germination index exceeding 80% in 60 days of decomposition. Inoculated and grass clamping amended wheat straw-poultry dropping mixture resulted in compost with highest humus content of 11.8% and C:N ratio of 14:1, humic acid fraction of 2.84% and germination index of 59.66%. Fungal consortium was effective in improving the humus content of all the composted mixtures.

Nakasaki et al. (2009) observed that Thermobifida fusca, a thermophilic actinomycete produced enzymes for lignocellulose degradation during composting of sludge which contained a high concentration of lipids and fibers. Parveen and Padmaja (2009) carried out an experiment to assess the degrading efficiency of cellulytic fungi (Trichoderma viride, T. koningii and T. harzianum) on the biodegradation of Spent Mushroom Substrate (SMS).

The results revealed that, among fungal inoculated spent mushroom substrate samples (T1 - Raw SMS (uninoculated control), T2 - SMS + Trichoderma viride, T3 - SMS + Trichoderma koningii, T4 - SMS + Trichoderma harzianum), Trichoderma viride inoculated SMS (T2) was found to be an efficient degrader of spent mushroom substrate within 60 days of biodegradation as they showed drastic reduction in the biochemical parameters like organic carbon (23.86 per cent), cellulose (8.78 per cent), phenolic content (0.21mg g-1) and reducing sugars (0.14 mg g-1). C:N ratio was narrowed down drastically from 29:1 to 11:1 while nitrogen content increased to 2.7 per cent from 1.1 percent compared to uninoculated control T1.

According to Sanchez (2009), the ability of fungi to degrade lignocellulosic materials was due to their highly efficient enzymatic system. Fungi have two types of extracellular enzymatic systems; the hydrolytic system, which produces hydrolases that were responsible for polysaccharide degradation and a unique oxidative and extracellular ligninolytic system, which degrades lignin and opens phenyl rings. Lignocellulosic residues from wood, grass, agricultural, forestry waste and municipal solid waste are particularly abundant in nature and have a potential for bioconversion. Sengar et al. (2009) investigated the efficacy of Paecilomyces variotii and Chaetomium globosum in converting sugarcane bagasse into nutrient enriched organic manure. The results of the study revealed that, among fungal inoculated bagasse samples (T1 - Raw bagasse uninoculated control), T2 - bagasse + Paecilomyces variotii, T3 - bagasse Chaetomium globosum, T4 - bagasse + Paecilomyces variotii and Chaetomium globosum ), T4 treatment (bagasse + Paecilomyces variotii and Chaetomium globosum) was found to be an efficient degrader of bagasse within 60 days of biodegradation as they showed drastic reduction in the biochemical parameters like organic carbon (22.40 per cent), cellulose (13.40 per cent), phenolic content(0.43mgg-1) and non reducing sugars (0.38 mg g-1). C:N ratio was narrowed down from 49:1 to 17:1 while nitrogen content increased to 1.31 per cent from 0.65 percent compared to uninoculated control T1.

Zeng et al. (2009) studied the effect on compost maturity by inoculating the lignocellulolytic fungus, Phanerochaete chrysosporium during different phases of agricultural waste composting. In the three runs, a decrease in C/N ratio, increase in germination index and humification indices (humification ratio, humification index, percentage of humic acids and degree of polymerization) were reported. Furthermore, the different effects of inoculation during different phases on compost maturity was observed using ANOVA. P. chrysosporium induced significant changes in all parameters of compost maturity except C/N ratio during the second fermentation phase. Chaturvedi et al. (2010) conducted an experiment to analyze and evaluate the effect of microbial bio-augmentation on composting of Jatropha cake in terms of changes in physico-chemical and hydrolytic enzymatic parameters. The microbial inocula consisted of fungal strains, namely, Aspergillus awamori, Aspergillus nidulans, Trichoderma viride, and Phanerochaete chrysosporium which were further amended with beneficial soil microorganisms like Pseudomonas striata and Azotobacter chroococcum. Various physico-chemical and enzymatic activities were estimated at various stages of composting. It was observed that the bio-augmented compost resulted in most desirable phosphorus availability (3.25 mg g-1) at the time of maturity. The hydrolytic activities, cellulases (FPase, CMCase and cellobiase) and xylanase displayed a significant increase after 60 days, indicating their contribution in the process of rapid decomposition. Results indicated high germination index (80 per cent) of the bioaugmented compost, which was significantly correlated to key enzymatic parameters and phosphorus content of compost.

According to Gautam et al. (2010b), recycling of Municipal solid waste by composting was a time consuming process and to reduce the composting period, efficient decomposing microorganism were needed. Fourteen bacterial and fungi isolates were screened and characterized from MSW waste. The three isolates Pseudomonas sp., Trichoderma viride and Trichoderma sp. were found to be more efficient when compared to the other isolates and a microbial consortium was

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developed using these isolates. Further, to develop effective consortia, their efficiency to produce enzymes such as cellulases and pectinases at different temperature and pH were studied. The temperatures ranging from 40-60°C and pHe ranging from 6.0 to 7.0 were found to be favourable for enzyme production. Parveen and Padmaja (2010) converted cöir waste, an environmental pollutant into organic manure by inoculating a white ro¢ fungus, Phanerochaete chrysosporium and incubating for 60 days. The results of the study revealed that there was a significant reduction in lignin, cellulose and organic carbon, C:N ratio, total phenol, reducing and non-reducing sugar levels. An increasing trend was observed in total nitrogen from 0.26 to 1.01 per cent after 60 days of Phanerochaete chrysosporium inoculation.

Zeng et al. (2010) determined the effect of inoculation time points on the enzyme activities during agricultural waste composting. Four runs were used: without inoculation (Run A), inoculation with Phanerochaete chrysosporium (P. chrysosporium) during the first fermentation phase (Run B), inoculation during the second fermentation phase (Run C) and inoculation during both the first and the second fermentation phases (Run D). The results revealed that the effect of inoculation on carboxy methyl cellulase (CMCase) activities was negative during the first fermentation phase. The inoculation increased the activities of xylanase (almost 3000 U/g) during the first fermentation phase but no obvious difference among Runs A-D was observed during the second fermentation phase. The peak values of manganese peroxidase (MnP) in Runs C and D were three times higher than those of Runs A and B after 21 days. The inoculation positively affected the lignin peroxidase (LiP) activities during the first fermentation phase and had a significant negative effect on the laccase (Lac) activities during the second fermentation phase. Therefore, the inoculation during the second fermentation phase was more effective than that during the first fermentation phase.

II. MATERIALS AND METHODOLOGY

Sampling
Different soil samples are collected from municipal waste dumping places Alwal, Nallakunta, Secendrabad areas of Hyderabad, Telangana.

Serial Dilution
1. Taken 1 gram of soil sample in a test tube and added 10ml of distilled water.
2. Then serial dilution is done in this process 8 test tubes are taken with the first test tube containing 10ml of water and 1gm of sample.
3. 1ml of sample from the 1st test tube is transfer to next test tube and added 9ml of water using micro pipette respectively till the 7th one
4. The sixth and seventh test tubes are bacterial cultures and they are spread on agar slants then after 24 hours of inoculation antifungal activity is observed in the media.
5. The solution of the first test tube containing 1gm of sample is mixed using vortex shaker the sample containing the test tube 10.6 and 10.7 are taken and the others are discarded all the medium are shifted from autoclave to laminar air flow then u is switched on for sterilization for 10mins before that is done the laminar air flow is wiped with cotton dipped with ethyl alcohol the sterilized petri plates must be free from bubbles.

Preparation of Nutrient Agar
Agar medium is prepared by mixing nutrient medium with commercially available agar powder if nutrient medium is not available then it can be individually prepared by water, peptone, beef extract and agar. 1gm of agar is weighted and it is mixed with nutrient medium mixed with 100ml of water and poured into petri plates then it is kept for autoclave at 121°C at 15Pas for 15 min.

Sub-Culturing
The colonies are picked up from the 10.6 and 10-7 plates and are streaked upon agar slants Which when inoculated produces active cultures More active cultures are also prepare by inoculating the bacteria in nutrient broth without agar.

Characterization of Potential Isolates
Potential isolates were further characterized through morphological and biochemical tests.

Morphological Characterization

Gram's Staining
1. A smear was prepared with a bacterial culture.
2. 1-2 drops of crystal violet stain was added and the smear was left for 1 min.
3. Crystal violet stain was added and then rinsed off with water.
4. Grams’ stain was added on the smear and left for 1 min. Then rinse it off with water.
5. Then washed with 70% ethanol
6. 1-2 drops of safranin was added and rinsed off with water.
7. Observed under the microscope.

Capsule Staining
1. One loop full of Indian ink was placed at one end of the microscope slide. One loopful of sterile saline was mixed with the ink.
2. The bacteria was transferred and mixed in small amount of Indian ink.
3. A second slide was taken and was held at 45 degree angle so that the end of each slide touches the end of the other slide. Then the upper slide was pulled to meet the drop.
4. without raising the slide the top slide was pushed back to spread the stain.
5. The slide was allowed to air dry.
6. The smear was flooded with Ethylene blue for 3 minutes.

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7. Ethylene blue was removed and the slide was gently rinsed with the water.
8. The slide was allowed to air dry.
9. Observed under microscope.

**Spore Staining**
1. A smear was prepared with a bacterial culture.
2. Slide was covered with paper towel and kept it over a water bath.
3. Added drop by drop of Malakite green indicator on the paper towel for 3 min as the paper towel getting dry.
4. Slide was removed from the water bath and paper towel also removed from the slide.
5. Washed with few drops of distilled water.
6. Added few drops of safranin and waited for a minute
7. Flooded with distill water
8. Air dried the slide and observed under the microscope.

**Biochemical Tests Carbohydrates Fermentation**

**Principle**
Fermentation degradation of various carbohydrates such as glucose (a monosaccharide), sucrose (disaccharide), cellulose (polysaccharide) by microbes, under anaerobic condition is carried out in a fermentation tube. A fermentation tube is a culture tube that contains a Durham tube (i.e. a small tube placed in an inverted position in the culture tube) for the detection of gas production, as an end product of metabolism. The fermentation broth contains ingredients of nutrient broth, a specific carbohydrate (glucose, lactose, maltose, sucrose, or mannitol) and a pH indicator (phenol red), which is red at a neutral pH (7) and turns yellow at or below a pH of 6.8 due to the production of an organic acid.

**Materials Culture**
Sterile fermentation tubes along with Durham’s tubes of:
- Glucose broth
- Sucrose broth
- Lactose broth
- Inoculating loop

**Methodology**
Preparation of fermentation medium whose constituents are as follows:

| Ingredient                  | Amount       |
|-----------------------------|--------------|
| Tryptone /peptone           | 1.0 g        |
| Carbohydrate*               | 0.5 g        |
| Sodium chloride             | 1.5 g        |
| Phenol red                  | 0.0018 g     |
| Distilled water             | 100.0 ml     |

(*A specific carbohydrate--- such as glucose, sucrose and lactose is added)
1) Broth was taken into fermentation tubes and Durham’s tubes were inserted into the fermentation tube such a way that the broth enters into the Durham’s tube also. Autoclaved at 121°C for 15 minutes.
2) After autoclaving it was cooled at room temperature.
3) The culture was inoculated and all the 6 inoculated and 3 un inoculated tubes at 35°C for 24-48 hours were incubated

**Catalase Activity**

**Principle**
Some bacteria can reduce diatomic oxygen to hydrogen peroxide or super oxide. Both of these molecules are toxic to bacteria. Some bacteria, however, possess a defense mechanism, which can minimize the harm done by the two compounds. These resistant bacteria use two enzymes to catalyze the conversion of hydrogen peroxide and super oxide back into diatomic oxygen and water. One of these enzymes is catalase and its presence can be detected by a simple test. The catalase test involves adding hydrogen peroxide to a culture sample or agar slant. If the bacteria in question produce catalase, they will convert the hydrogen peroxide and oxygen gas will be evolved. The evolution of gas bubbles to form and is indicative of a positive test.
Catalase has one of the highest turnover rates of all enzymes; one molecule of catalase can convert millions of molecules of hydrogen peroxide to water and oxygen per second. Catalase is a tetramer of four polypeptide chains, each over 500 amino acids long. It contains four porphyrin heme(iron) groups which allow the enzyme to react with the hydrogen peroxide. The optimum pH for catalase is approximately neutral (pH 7.0).

Bacteria cannot protect themselves from the lethal effect of hydrogen peroxide, which is accumulated as a product of carbohydrate metabolism. Catalase is a hemoprotein. Catalytic decomposition of hydrogen peroxide involves the reduction of trivalent iron (Fe$^{3+}$) to (Fe$^{2+}$) and the reoxidation of the latter by oxygen. This reaction can be summarized by the following equation.

$$2H_2O_2\rightarrow 2H_2O + O_2$$

**Materials**
1. Culture
2. Tryptone soy agar media
3. Hydrogen peroxide (3%)(make HI-MEDIA)
4. Four sterile test tubes
5. Inoculating loop

**Methodology**
Preparation of Tryptone soy agar media of the following composition:

| Ingredient                  | Amount       |
|-----------------------------|--------------|
| Tryptone                   | 1.5 g        |
| Peptone                     | 0.5 g        |
| Sodium chloride             | 0.5 g        |
| Agar                        | 1.5 g        |
| Distilled water             | 100.0 ml     |

The contents were mixed thoroughly and the medium was sterilized by autoclaving at 15 lbs pressure for 15 minutes.
Hydrogen Sulphide Production Test

Principle

\[ \text{H}_2\text{S} \] is commonly called as “rotten egg” gas, because of the copious amounts liberated through reduction of sulphur containing amino acids or through the reduction of inorganic sulphur compounds like sulphates, sulphites or thiosulphates or \( \text{H}_2\text{S} \) production can be detected by incorporating a heavy metal salt containing iron or lead ion as \( \text{H}_2\text{S} \) indicator to the medium.

\( \text{H}_2\text{S} \) is a colorless gas when produced reacts with metal salt forms visible black insoluble ferrous sulphide precipitates causing blackening of the medium.

Tryptophanase

\[
\text{Tryptone} \quad \text{---------------------------} \quad \text{Indole + pyruvic acid + NH}_3
\]

\[
\text{Indole + Kovac’s reagent} \quad \text{---------------------------} \quad \text{Rosindole + H}_2\text{O}
\]

(Butonol) (Cherry red compound)

Materials

1. Culture.
2. Tryptone broth
3. Kovac’s reagent
4. Sterile test tubes
5. Inoculating needle

Methodology

1. Preparation of Tryptone broth;
   - Tryptone 1.0 g
   - Sodium chloride 0.1 g
   - Peptone 1.0 g
   - Distilled water 100.0 ml
   - It was sterilized in autoclave at 15 lbs (121°C) for 15 min.
2. The broth was cooled to room temperature and then pours into four sterile test tubes.
3. Three tubes were inoculated with culture, and third tube was kept as a un inoculated Comparative to control.
4. The tubes were incubated at 35°C for 48 hrs. After 48 hrs of incubation, 1 ml of Kovac’s reagent was added to each tube including control.
5. The tubes were gently shaken after intervals for 10-15 minutes.
6. The tubes were allowed to stand to permit the reagent to come to top.

Methyl-red and Voges-proskauer Tests Principle

The methyl-red (MR) and the Voges-Proskauer (V-P) tests are used to differentiate two major types of facultative anaerobic enteric bacteria that produce large amounts of acid and those that produce the neutral product acetoin as end product. Both these are performed simultaneously because they are physiologically related and are performed on the same medium MR-VP broth. Opposite results are usually obtained for the methyl-red and Voges-Proskauer tests, i.e. MR+, VP- or MR-, VP+. In these if an organism produces large amounts of organic acids: formic, acetic, lactic and succinic (end products) from glucose, the medium will remain red (a positive test) after the addition of methyl red a \( \text{P}^\text{H} \) indicator (i.e. \( \text{P}^\text{H} \) remaining below 4.4). In other organisms, methyl red will turn yellow (a negative test) due to the elevation of the \( \text{P}^\text{H} \) above 6.0 because of the enzymatic conversion of the organic acids (produced during the glucose fermentation) to non-acidic end products such as ethanol and acetone.

Materials

1. culture
2. MRVP broth
3. Methyl red \( \text{P}^\text{H} \) indicator (make HI-MEDIA)
4. V-P reagent I (naphthol solution)(make HI-MEDIA)
5. V-P reagent II (40% Potassium hydroxide)(make HI-MRedia)
6. Sterile test tube
7. Inoculating loop

**Methodology**

Preparation of MRVP broth (pH 6.9)
- Peptone 0.7 g
- Dextrose/Glucose 0.5 g
- Potassium phosphate 0.5 g
- Distilled water 100.0 ml

Sterilized by autoclaving at 15 lb pressure for 15 minutes.
1. After autoclaving it was cooled to 50°C and then poured into four sterile test tubes.
2. Three test tubes were inoculated with culture and keep one tube as un inoculated comparative control.
3. All four tubes were incubated at 35°C for 48 hours.
4. 5 drops of methyl red indicator was added to two tubes.

**Citrate Utilization Test**

Principle
Citrate is used to differentiate among enteric bacteria on the basis of their ability to utilize/ferment citrate as the sole carbon source. The utilization of citrate depends on the presence of an enzyme citrase produced by the organism, which breaks down the citrate to oxaloacetatic acid and acetic acid. These products are later converted to pyruvic acid and carbon dioxide enzymatically as shown below:

\[ \text{Citrate} \rightarrow \text{Citril acid} \rightarrow \text{Oxaloacetic acid} + \text{Acetic acid} \]

Enzymes

\[ \text{Oxaloacetic acid} + \text{Acetic acid} \rightarrow \text{Pyruvic acid + CO}_2 \]

Inoculating the microorganisms into an organic synthetic medium, Simmons’ citrate agar, where sodium citrate is the only source of carbon and energy, performs the citrate test. Bromothymol blue is used as an indicator. When the citric acid is metabolized, the CO₂ generated combines with sodium and water to form sodium carbonate an alkaline product, which changes the color of the indicator from green to blue and this constitutes a positive test:

\[ \text{CO}_2 + 2\text{Na}^+ + \text{H}_2\text{O} \rightarrow \text{Na}_2\text{CO}_3 + \text{H}^+ \]

(Produced during citric acid metabolism) (alkaline pH) (blue color)

Bromothymol blue is green when acidic (pH 6.8 and below) and blue when alkaline (7.6 and higher).

**Materials**
1. culture
2. Simmons citrate agar media (make HI-MEDIA)
3. Inoculating loop
4. Four sterile test tubes

**Methodology**

1. 3.4 gms of Simmons’ citrate agar (pH 6.9) media was weighed and dissolved in 100ml of distilled water and sterilized and autoclaved at 15 lb pressure for 15 minutes.
2. The medium was cooled and poured into four sterile test tubes, and was allowed to solidify in a slanting position.
3. Three Simmons’ citrate agar slants were inoculated with culture by means of a stab-and-streak inoculation. The fourth tube was kept as an uninoculated comparative control.
4. All the tubes were incubated at 37°C for 48 hours.

**Urease Test**

Principle
Urea is a major organic waste product of protein digestion in most vertebrates and is excreted in the urine. Some microorganisms have the ability to produce the enzyme urease. The urease is a hydrolytic enzyme, which attacks the carbon and nitrogen bond amide compounds (e.g. urea) with the liberation of ammonia as shown below:

\[ \text{H}_2\text{N} > \text{O} + \text{H}_2\text{O} \rightarrow \text{NH}_3 + \text{CO}_2 \]

Urease test is performed by growing the test organisms on urea broth or agar medium containing the pH indicator phenol red (pH 6.8). During incubation, microorganisms possessing urease will produce ammonia that raises the pH of the medium/broth. As the pH becomes higher, the phenol red changes from a yellow color (pH 6.8) to a red or deep pink (cerise) color. Failure of the development of a deep pink color due to no ammonia production is evidence of a lack of urease production by the microorganisms.

**Materials**
1. culture
2. Urea agar medium (make HI-MEDIA)
3. Urea solution 20% in 100 ml
4. Inoculating loop
5. Four sterile test tubes

**Methodology**

About 2.8 gms of urea agar medium was weighed and dissolved in 100 ml of Distilled water and autoclaved at 121°C for 15 minutes and cool to 50°C. Glucose 0.1 g Phenol red (0.02% solution) 3.0 ml Add to the molten base and steam for 1 hour, cool to 50°C.
Urea, 20% aqueous solution 100.0 ml
Sterilized by filtration and add aseptically to the basal medium.
1. The solution was mixed well and distributed into four sterile test tubes and the medium was allowed to Solidify in a slanting position to form slopes.
2. After the solidification, the three test tubes were inoculated with Culture, and one tube as un inoculated comparative control.
3. The slants were incubated for 24-48 hours at 37°C.

Principle
Many bacteria produce extracellular enzymes used to catalyze chemical reactions outside of the cell. In this manner, nutrient sources, such as starch, that are too large to be absorbed through the cell membrane can be broken down into smaller molecules and transported into the cell via diffusion.

In the starch hydrolysis test, the test bacteria are grown on agar plates containing starch. If the bacteria have the ability to hydrolyze starch, it does so in the medium, particularly in the areas surrounding their growth while the rest of the area of the plate still contain non-hydrolysed starch. Since no color change occurs in the medium when organisms hydrolyze starch, iodine solution is added as an indicator to the plate after incubation. While the non-hydrolysed starch forms dark blue color with iodine, its hydrolyzed end products do not acquire such dark blue color with iodine.

Consequently, transparent clear zones are formed around the colonies that hydrolyze starch while the rest of the plate show a dark blue coloration as iodine forms the colored complex with starch.

Media
Starch agar is a simple nutritive medium with starch added. Beef extract and pancreatic digest of gelatin provide nitrogen, vitamins, carbon and amino acids. Agar is the solidifying agent and starch is the carbohydrate.

Composition
Peptic digest of animal tissue 5.000, Sodium chloride 5.000, Yeast extract 1.500, Beef extract 1.500 Starch, soluble 2.000 Agar 15.000 Final pH ( at 25°C) 7.4±0.2

Method
1. Using a sterile technique, make a single streak inoculation of organism to be tested into the centre of labeled plate.
2. Incubate the bacterial inoculated plates for 48 hours at 37°C.
3. Following incubation, flood the surface of the plates with iodine solution with a dropper for 30 seconds.
4. Pour off the excess iodine.
5. Examine for the clear zone around the line of bacterial growth

Results
Serial Dilution

Isolation of Bacteria
Potential strains was and observed different colony morphology was streaked on nutrient agar slants to obtain pure culture and they were used for further study.

| Isolate | Shape   | Size   | Margin  | Elevation | Colour | Opacity |
|---------|---------|--------|---------|-----------|--------|---------|
| 1       | Circular| Small  | Entire  | Raised    | White  | Opaque  |
| 2       | Irregular| Medium| Serrate | Flat      | White  | Opaque  |
| 3       | Irregular| Medium| Serrate | Flat      | White  | Opaque  |
| 4       | Irregular| Medium| Serrate | Flat      | White  | Transparent |
| 5       | Circular| Small  | Entire  | Flat      | White  | Opaque  |
Morphological Tests

1. Gram’s Staining

*Observation*  
After the process of staining it is observed that, purple coloured bacterial cells in spherical shape.

*Result*  
From the above observation the given isolate was identified as Gram – negative Cocci(spherical) bacteria.

*Isolates 3 & 4*

*Observation*  
After the process of staining we observed purple coloured bacteria in rod shape.

*Result*  
From the above observation the given isolates 4 & 5 was identified as Gram – positive Bacillus bacteria.

2. Spore Staining

*Isolate 1 & 2*  
Isolate 3, 4 & 5 are non-spored.  

*Observation*  
After staining all bacterial isolates 1 & 2 are observed green in colour and 4, 5 & 5 are observed in pink in colour.

*Result*  
From the above observation isolate 1 & 2 is sporulated and 3, 4 & 5 are non sporulated.

4. Capsule Staining

*Isolate 3*  
Capsulated 1, 2, 4 & 5 are non-capsulated.

*Observation*  
After staining bacterial cells are observed in blue in colour for isolates 1 & 2 due to the absorption of CuSO4 by the capsule and isolate 3, 4 & 5 are observed in purple colour.

*Result*  
From the above observation the isolates 1, 2, are identified as Capsulated and isolate 3, 4 & 5 are non-capsulated.

Biochemical Tests

1. Indole Production Test

*Observation*  
There is no development of cherry (deep) red color in the top layered of the tube.

*Result*  
From above observation all the 5 test isolates shown negative results as there is no formation of cherry red colour was seen.

2. Methyl-red and voges-proskauer Tests

*Observation*  
After incubation period when methyl red indicator was added media colour changes to red. rest of test isolates shown yellow in colour.

*Result*  
From above observation test isolate shown positive results towards the test.
Red colored was observed in isolate 2 and isolate 1,3,4,5 was shown yellow colour.

**Observation**
After incubation period when vp reagent 1 & 2 were added to the test isolates 1,2,3,4 media colour changed to brick red in colour.

**Result**
Test isolates 1,2,3,4 are shown positive towards the test.

3. **Citrate Utilization Test**

**Observation**
After 48 hours of incubation it was observed that there is change in the medium color for all isolates from green to blue.

**Result**
From the above observation all the 5 test isolates are shown positive test results.

4. **Urease Test**

**Observation**
After 48 hours of incubation it was observed that there is color changed in the test isolates. Pink colour was observed in all the 5 isolates was shown positive results.

**Result**
From the above observation all the 5 test isolates shown positive result.

4. **Catalase Production Test**
Observation
After 24 hours of incubation when few drops of hydrogen peroxide was added to the test culture, appearance of gas bubbles was observed.

Result
From the above observation, all isolates are positive for catalase test.

5. Carbohydrate Fermentation Test

Observation
When phenol red indicator added to the test solution, color of the test solution changed to yellow. Formation of the gas bubbles in Durham tubes was observed.

Result
No colour change indicating that, no Ph change in the test media. Formation of gas indicates production of gas.

H2S Production test

Observation
By adding iodine solution on starch agar plate, it is observed that, formation of golden yellow color on Isolate-5.

Result
Isolate 5 showing positive result for Amylase test and rest all Isolates are negative for amylase test.

SEED GERMINATION TEST

A) Seed Germination by Paper Towel Method

Seed vigor index was studied using red gram as host plant and seed treatment was done using different treatments (Red grams seeds were treated with different isolates isolate 1: (T1), isolate 2: (T2), isolate 3: (T3), isolate 4: (T4), isolate 5: (T5) and control 6(T6) separately and seed vigor index was calculated. Highest seed vigor index was with isolate 1 followed by isolate 5 when compared with other isolates and control. Seed germination by paper towel method using Isolate 1 supernatant was 100 % compared with other treatments.
Seed vigor index by paper towel method

| Treatment | Germination % | Root length (cm) | shoot length (cm) | vigor index |
|-----------|---------------|------------------|-------------------|-------------|
| T1        | 100±(0.14)b   | 20±(0.24)b       | 27±(0.11)b        | 0.40±(0.12)b|
| T2        | 40±(0.02)a    | 22±(0.08)b       | 28±(0.02)b        | 0.40±(0.06)a|
| T3        | 100±(0.02)c   | 19±(0.02)f       | 11±(0.14)c        | 0.70±(0.44)c|
| T4        | 80±(0.44)c    | 15.5±(0.24)d     | 28±(0.24)c        | 0.48±(0.34)d|
| T5        | 60±(0.24)d    | 7±(0.14)d        | 23±(0.24)b        | 0.44±(0.24)  |
| T6        | 100±(0.02)c   | 2±(0.34)d        | 14±(0.24)d        | 0.20±(0.34)d|

Values superscribed by a-g are ranking highest to lowest of significant, same alphabet are insignificant according to Fischer’s least significance difference test (p<0.05). Values in the brackets are standard error; values in column are mean two independent experiments of 4 replications.

Efficacy of Isolate 1 on growth of Red gram (greenhouse study)

Greenhouse experiment was performed to evaluate growth of Red gram seeds using different treatments (isolates isolate 1: (T1), isolate 2: (T2), isolate 3: (T3), isolate 4: (T4), isolate 5 (T5), and control: (T6). Data revealed that there was an increase in shoot and root length when seeds were treated with isolate-1 and isolate 5 when compared to control. Similarly, plant biomass (shoot and root weight) increased with Isolate1 (0.40g and 53.6 %) followed by Isolate5 (0.28g and 37.9 %) treatment when compared to control. There was no toxicity effect by Isolate1 on seed germination.

| Treatment | Shoot length(cm) | root length(cm) | Bio mass weight (gm) |
|-----------|------------------|-----------------|----------------------|
| T1        | 14±(0.3)b        | 9.5±(0.3)b      | 0.46±(0.2)b          |
| T2        | 9.5±(0.2)a       | 9.6±(0.3)a      | 0.22±(0.3)a          |
| T3        | 9.1±(0.4)c       | 7.5±(0.4)c      | 0.19±(0.3)c          |
| T4        | 9.0 ±(0.2)c      | 2.5±(0.1)d      | 0.24±(0.5)d          |
| T5        | 12.3±(0.2)b      | 6.5±(0.3)c      | 0.42±(0.4)c          |
| T6        | 8.1±(0.2)a       | 7.0±(0.2)b      | 0.32±(0.2)a          |

Values superscripted by a-g are ranking highest to lowest of significant, same alphabet are insignificant according to Fischer’s least significance difference test (p<0.05). Values in the brackets are standard error; values in column are mean two independent experiments of 4 replications. Using Red gram as host plant, Isolate-1 and isolate-5 showed significant increase in plant defense enzymes when compared to pathogen control.
In seed germination, seed coated with test Isolate-1 showing 2times more growth compared with control.

### III. DISCUSSION

Every year, the level of waste generation continues to rise because of uncontrolled consumption due to increasing population, attitudes towards spending and high living Standards. Quantities of waste generated are growing in response to the rapid increase in population, accelerated urbanization and industrialization.

However, with the increasing population, the volume of waste generated remains abundant. Solid waste is generated from various domestic (schools, hospitals, universities, offices) and commercial (from restaurants, hotels, markets and industry) sources and consists of biodegradable matter, as well as inert non- biodegradable matter. The most dominant variable in the municipal solid waste (MSW) flow is food waste. Relatively
homogeneous residential waste, some differences in the waste depends on local factors and other demographics; most households dispose of the same type of waste (Agamuthu, 2013). The main household waste composition includes 71% organic waste, 12% plastic, 7.5% paper and paper products, 5% dirt and construction debris and 1% hazardous waste. The highest percentage is organic waste, although the composition of the waste varies depending on the source.

Economic development, urbanization and improving living standard in cities of developing countries have lead to increase in the quantity and complex composition of municipal solid waste. Management of municipal solid waste resulting from rapid urbanization has become a serious concern for government departments, pollution control agencies, regulatory bodies and public in most of the developing countries (Hoornweg et al., 1999). Several other factors like education standard and infrastructure of the country have significant effect on municipal solid waste generation. The estimation and prediction of municipal solid waste generate on play an important role in municipal solid waste management. The quantity of municipal solid waste in developing countries has been consistently rising over the years. The municipal solid waste composition varies from place to place and also bears a rather consistent correlation with the average standard of living. The waste generated in the developing countries is similar in composition, the variation between regions being dictated by the climatic, cultural and industrial, infrastructural and legal factors (Hoornweg et al., 2013).

Solid waste is mostly an urban phenomenon, and is generally an Urban Issue. Today, more than 50% of the World’s population lives in the cities and the rate of urbanization is increasing quickly. Solid Waste generation is the by-product of the Urbanization. It is highly related with Economic growth, degree of industrialization and consumption pattern. With the increase of urban population of the cities and towns all other activities associated with population also increases resulting in more and more generation of Municipal Solid Waste. And in the absence of technology and efficient and effective methods of disposing refuse worsen the quality of Air of the urban centers which have detrimental impacts on human health. The most common problems associated with improper management of solid waste include disease transmission, odor, nuisance, atmospheric, land & water pollution, fire hazards, aesthetical nuisance and economic losses. (Yeny and Yulinah, 2012) More or less every human activity creates some kind of waste. As countries develop economically, socially, and technologically waste generation also increases. Both developed and developing countries face the problems associated with solid waste generation and its management.

Rapid urbanization directs to the densification and an increase of large amounts of solid waste within a concentrated area. Global population rose to 6.9 billion in 2010 and the majority of people live in developing countries. A major challenge is how to manage the ever-increasing waste generated, especially in developing countries already lacking a sufficient public service infrastructure to manage municipal waste, and where poverty and unplanned settlements lead to unmanaged waste. (World Bank, 2012) Globally, we live in “throw-away” societies in which we consume packaged products that do not last past a single use or even a year, and we discard as waste what we no longer want. This wasteful lifestyle seriously impacts the environment, public health, and produces social and economic problems. Waste disposal can have serious environmental impacts: landfills consume land space, and cause air, water and soil pollution - including the emission of greenhouse gases, while incineration results in emissions of dangerous air pollutants. Our consumptive and wasteful behavior needs to be examined, and changed, so that we can live more sustainably. (World Bank, 2012) Solid waste generation is the common basis for activity data to estimate emissions from solid waste, disposal, biological treatment, and incineration and open burning of waste. Solid waste generation rates and composition vary from country to country depending on the economic situation, industrial structure, waste management regulations and life style. The availability and quality of data on solid waste generation as well as subsequent treatment also vary significantly from country to country. Statistics on waste generation and treatment have been improved substantially in many countries during the last decade, but at present only a small number of countries have comprehensive waste data covering all waste types and treatment techniques. Solid waste is generated from households, offices, shops, markets, restaurants, public institutions, industrial installations, water works and sewage facilities, construction and demolition sites, and agricultural activities (Hoornweg and Thomas, 1999). Solid waste management practices include: collection, recycling, solid waste disposal on land, biological and other treatments as well as incineration and open burning of waste. A new, far-reaching report on the state of municipal solid waste around the world predicts a sharp rise in the amount of garbage generated by urban residents between now and 2025 (Zang, 1998).

The report estimates the amount of municipal solid waste (MSW) will rise from the current 1.3 billion tons/year to 2.2 billion tons/year, with much of the increase coming in rapidly growing cities in developing countries (World Bank, 2012). Globally, waste volumes are increasing quickly even faster than the rate of urbanization. In general, as a country urbanizes and populations become wealthier, the consumption of inorganic materials (e.g. plastics, paper, glass, aluminum) increases, while the relative organic fraction decreases (UNEP, 2001).
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