Int.J.Curr.Microbiol.App.Sci (2023) *12*(02): 135-141

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 12 Number 2 (2023)**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2023.1202.013

Production and Application of Bioculture for Farm Waste Recycling

Kshama Deshpande*, Vijay Deshmukh, Ashok Kharde and Pratiksha Nagmode

Mahatma Gandhi Missions College of Agricultural Biotechnology, MGM Hills, Gandheli, Aurangabad, Maharashtra, India

**Corresponding author*

Keywords

Farm Waste, Bio-Culture, Degradation, Compost, fertilizers, pesticides

Article Info

Received:
01 January 203
Accepted:
30 January 2023
Available Online:
10 February 2023

A B S T R A C T

Bio degradation of organic waste such as agricultural waste is the most important and efficient way to remove these compounds from the environment. Bacteria, actinomycetes, fungi, algae and protozoa are the main microorganisms found in soil that decompose soil organic materials, of which bacteria are the most prominent and abundant. Microbes use these waste compounds for their own metabolism and produces some simple and useful compounds, important for soil health, plant growth and overall ecological balance. Microorganisms have the ability to chemically and physically interact with substances, leading to structural changes or complete degradation of target molecules. Therefore, this study was focused on the importance of isolation, characterization and identification of bacteria from farm waste and its application on farm waste for the production of compost. In this study the various bacteria from farm waste were isolated in the department of Microbial and environmental biotechnology at MGM College of Agricultural biotechnology, Gandheli, Aurangabad, Maharashtra. The bacteria were assessed by using various morphological biochemical and genetic characterization. The bio culture prepared from the isolated bacteria was applied for the degradation of farm compost. It was found that the bioculture degrade the compost in twenty one days with all the standard characters of compost. It was assessed by using detection tests. Thus by using the bio culture the farm waste decomposition can be carried out. With further molecular studied the exact species of the bacteria can be identified and used as a formulation for compost decomposition.

Introduction

Agricultural activity releases highly toxic organic compounds that have been synthesized and released into the environment directly or indirectly over a long period of time. Agricultural wastes include solids, liquids and gases. The production and improper disposal of these wastes has become a major pollution problem worldwide. In all developing and developed countries, a huge amount of waste is generated every day (Rao *et al.*, 2016).

Bio degradation of organic waste such as fertilizers, pesticides, and agricultural waste is the most important and efficient way to remove these compounds from the environment. Bacteria, actinomycetes, fungi, algae and protozoa are the main microorganisms found in soil that decompose

soil organic materials, of which bacteria are the most prominent and abundant. Microbes use the waste for their own metabolism and end up producing some simple and useful compounds that are important for soil health, plant growth and overall ecological balance. Microorganisms have the ability to chemically and physically interact with substances, leading to structural changes or complete degradation of target molecules. Therefore, this study was focused on the importance of isolation, characterization and identification of bacteria from waste soils (Wan Ishak *et al.,* 2011; Sarika *et al*., 2014).

Waste production and waste control play an important role in our environment. With the doubling of the population and the change in the lifestyle of the residents, the amount of municipal waste produced is increasing at an alarming rate. Most of this waste is landfilled in a designated collection yard. The biggest challenge for ecologists is the environmentally friendly management of this waste, and the application of microorganisms in this context has surpassed other available technologies. The organic waste is consumed by the bacteria, used by the bacteria as nutrients, and is no longer present to produce odor, sludge, pollution, or an unsightly mess. When bacteria consume waste, they convert the waste into safe by-products, and in the process of this conversion, they actually produce several metabolites that break down complex waste into simple compounds. Soil microorganisms are increasingly becoming an important resource in the search for industrially important molecules. The extent of microbial diversity in nature is still largely unknown, so there could be many other useful products yet to be identified from soil microorganisms.

In soil, 80 to 99% of microorganisms remain unidentified, and these biological communities are known to play a dominant role in maintaining a sustainable biosphere. Currently, both academic and industrial interest in soil bacteria (due to their several advantages over other microorganisms) is increasing in the search for these unique biologically active metabolites and new commercially important products from them. Bacteria are present in a variety of ecological habitats. They are considered very valuable because they are used in fermentation processes such as brewing, baking, cheese and butter production, chemical production such as ethanol, acetone, organic acids, enzymes, perfumes, etc., microbial mining and produce various antibiotics, vaccines, steroids as well as other therapeutically useful compounds with various biological activities. Thus, there is a huge opportunity to screen effective bacterial strains from landfills with valuable applications (Zaved *et al.,* 2008; Suganya *et al.*, 2012).

To cope with the demand for new organisms with the properties of producing unique enzymes/molecules in waste degradation, there has been a continuous effort to isolate new bacteria from diverse environments. Accordingly, this study was aimed at investigating bacterial strains from farm waste dumps with the ultimate goal of waste degradation and discovery of novel bioactive compounds for waste degradation.

Materials and Methods

Sample Collection

Soil Samples were Collected from location MGM campus, Aurangabad, Maharashtra. Sample were collected from upper Layer of soil and kept directly into Sterile beaker covered by sterile petri plate. The samples were Labeled and stored in dark until Analysis

Serial Dilution

Approximately, one gram of sample was taken into 9 ml sterile Distilled water in Beaker. the soil sample were shaken well with the help of Magnetic stirrer for 1 hour and Allowed to settled down for 10 minutes and supernatent were collected and add in test tube of 9:1 ratio (9 ml distilled water and 1 ml sample) and mix well. Again take 1ml sample from 1st tube and add in 2nd number tube having ratio same as 1st, this procedure can carry forward up to the 7th number tubes

Isolation of Bacteria

The suspensions were inoculated on solidified Nutrient agar medium with the help of sterilized inoculating needle by four way streaking method. The inoculated plates were incubated at 37℃

Identification of bacteria

Identification of isolated colonies was based on Morphological and Biochemical characteristics. The colonies were examined for cell Morphology, Gram staining, Motility. Among all the colonies grown on the nutrient medium at laboratory, total six isolates were taken for further investigation. Those are labeled as FWI 1 FWI 2 FWI 3 FWI 4 FWI 5 FWI 6 where FWI stands for farm waste isolates.

Biochemical characterization of bacterial isolates

Potential isolates were further characterized on the basis of their staining characteristics and further investigated for biochemical properties such as catalase, urease, cellulose degrading ability, nitrate reducing ability which helped in identification of the bacteria to genus level (Garcha *et al.*, 2016) Bergey's manual of determinative bacteriology (Holt *et al.*, 1994).

DNA isolation of bacterial isolates

Bacterial DNA of all the six isolates were carried out by using the standard methods. The purity of DNA was checked by using Nanodrop. This data will be useful for PCR amplication of the DNA and further studies.

Production of Bio culture

To prepare the microbial bioculture, loopfuls of each isolated species stored on nutrient agar slant were inoculated together The flask was shaken in an orbital shaker (120 rpm) at 37 ºC in the dark for 4 days to reach the phase of exponential growth.

Application of bio culture on farm waste

A total of 6 bacterial isolates were isolated from nutrient agar plates based on the colony morphology, color and shape. The bacterial population was found to be 4.5 cfu ml−1 at 10−7 Dilution. It is applied as 10 % on the fresh farm waste.

Characterization of compost produced by bio culture

After application of bio culture from isolated bacteria the farm waste was kept at room temperature which was in the range of 25^0 - 30^0. Daily observations were taken. After 21 days the compost was found to be prepared. The characterization of produced compost was done with respect to morphological characteristics, determination of volatile solid percentage and moisture percentage.

Results and Discussion

The Bio culture of bacteria was isolated using the basic nutrient medium in the laboratory. Among all the grown colonies total six bacterial colonies were selected which were found to have some unique appearance. These were further studied for their morphological characterization as shown in Table 1.

All the six bacteria were further subjected to the biochemical characterization for the identification up to genus level. They were tested for the presence of various enzymes such as catalase, cellulase, nitrate reductase, and urease.

The cellulase test was found to be positive indicating the presence of cellulase enzyme which degrades the lignocellulosic complexes into simple sugars. Cellulase of microbial origin have shown their potential application in various commercial sectors including textile, pulp and paper, laundry, brewing, agriculture and bio fuel.

Table.1 Morphological characteristics of the bacterial Isolates from farm waste

Characteristics	FWI1	FWI2	FWI3	FWI4	FWI5	FWI6
Size	6 mm	3mm	4mm	2mm	2mm	4mm
Shape	Circular	Circular	Circular	Circular	Circular	Circular
Colour	Milky white	Dull white	White	White	Off white	White
Margine	Entire	Entire	Entire	Entire	Entire	Entire
elevation	Raized	Flat	Raized	Raized	Flat	Flat
Surface	Smooth	Smooth	Smooth	Smooth	Smooth	Rough
Opacity	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque
Consistency	Buterous	Buterous	Vitrous	Buterous	Buterous	Buterous
Grams nature	-ve rods	+ve rods	+ ve rods	+ ve rods	+ ve rods	+ ve rods

FW - Farm waste isolate of bacteria

Table.2 Biochemical Characteristics of the isolated bacteria

Test name	FWI1	FWI2	FWI3	FWI4	FWI5	FWI6
Catalase test	+ve	+ve	+ve	+ve	+ve	+ve
Nitrate reductase test	+ve	+ve	+ve	+ve	+ve	+ve
Cellulase test	+ve	+ve	+ve	+ve	+ve	+ve
Urease test	+ve	+ve	+ve	+ve	+ve	+ve

Table.3 DNA quantification and puriety analysis

Colony no	ng/ml	Purity (260/280)
FWI1	856.7	1.47
FWI2	1140	1.8
FWI3	732	1.5
FWI4	1136	1.7
FWI5	549	1.4
FWI6	400	1.7

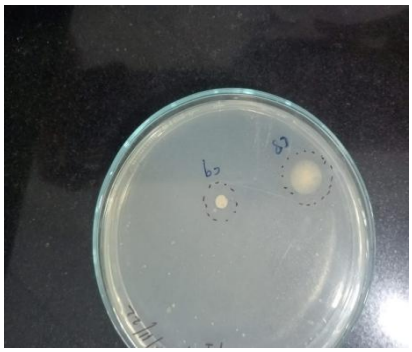
Table.4 Compost stability analysis

Parameter	Before decomposition of compost	After decomposition of compost
Colour	Yellow	Black
Weight	20 gm	15gm
Texture	fluffy	Moist
% of volatile solids	25 %	10.5%
% of moisture content	50%	64.7%

Fig.1

Farm Waste

Isolated bioculture colonies



Cellulase test

Nitrate Reductase test

Fig.2

Catalase Test

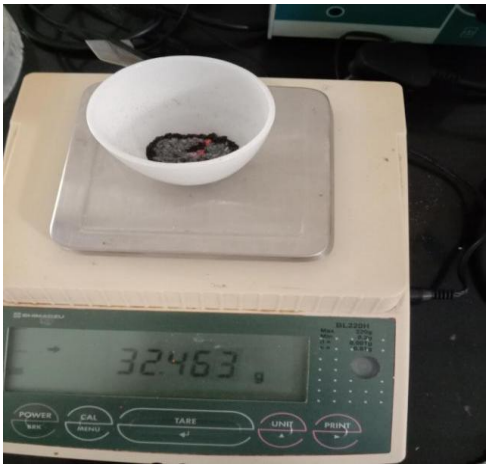
Urease Test

Farm waste

Fig.3

Decomposed farm Waste

Volatile soild determination



All the colonies showed the catalase test positive. It was essential for differentiating catalase-positive Micrococcaceae from catalase-negative Streptococcaceae. While it was primarily useful in differentiating between genera, it is also valuable in specification of certain gram positive organisms.

The nitrate reduction of all the isolates was shown to be positiveIt was used to determine the capability of isolated cultures to reduce nitrate (NO3-) to nitrite (NO2-) or other nitrogenous compounds via the action of the enzyme nitratase (also called nitrate reductase). This test was important in the identification of both Gram-positive and Gram-negative species.

The urease test identifies those organisms that are capable of hydrolyzing urea to produce ammonia and carbon dioxide. It was primarily used to distinguish urease-positive Protease from other Enterobacteriaceae.

All the six isolates were subjected for DNA isolation The use of DNA isolation technique led to efficient extraction with good quantity and quality of DNA, which showed the purity as shown in the table 3. Pure manual methods as well as commercially available kits were used for DNA extraction.

The prepared compost was subjected for volatile solid determination and moisture percentage determination. The volatile solids loss (VS% loss) is an indicator of how much organic matter conversion has occurred during composting. This value usually ranges from 30% for leaves to up to 90% for food wastes. It can be used as an indicator of compost stability. It assumes that ash is not lost during the composting process. The decrease in volatile solid percentage of the compost (15.95 %) indicating the presence of ash in the compost and compost stability.

The moisture percentage of the compost was found to be 64.7 % Composting proceeds best at a moisture content of 40-60% by weight. At lower moisture levels, microbial activity is limited. At higher levels, the process is likely to become anaerobic and foul-smelling. Thus with above findings in present research work, it can be concluded that by using the farm waste as a source of bacteria, the farm wastes can be transformed into the value added substance such as compost. This compost can be utilized for the improvement of soil texture in environmental friendly way. The bio culture that are isolated, characterised and assessed for its utility for waste degradation can be further subjected for identification on the genetic level by performing 16 S RNA sequence analysis.

References

Garcha, S., Verma, N., & Brar, S. K. (2016). Isolation, characterization and identification of microorganisms from unorganized dairy sector wastewater and sludge samples and evaluation of their biodegradability. *Water resources and industry*, *16*, 19-28.

Holt, J.G. (1994). Bergey's manual of determinative bacteriology. 9th Edition, Lippincott Williams and Wilkins, Baltimore.

Ishak, W. W., Jamek, S., Jalanni, N. A., & Jamaludin, N. M. (2011). Isolation and Identification of Bacteria from Activated Sludge and Compost for Municipal Solid Waste Treatment System. *Int. Proc. Chem. Biol. Environ. Eng*, *24*, 450-454.

Rao, M. R. K., Sivagnanam, S. K., & Syed, F. B. (2016). Isolation, characterization and identification of predominant microorganisms from agro-waste. *Der Pharmacia Lett*, *8*(5), 79-86.

Sarika, D., Singh, J., Prasad, R., Vishan, I., Varma, V. S., & Kalamdhad, A. S. (2014). Study of physico-chemical and biochemical parameters during rotary drum composting of water hyacinth. *International Journal of Recycling of Organic Waste in Agriculture*, *3*, 1-10.

Suganya, S., & Sumathy, V. (2012). Isolation and identification of bacteria from covered and uncovered mobile phones. *International Journal of Environmental Sciences*, *3*(1), 44-

54. https://doi.org/10.6088/ijes.2012030131006

Zaved, H. K., Rahman, M. M., Rahman, M. M., Rahman, A., Arafat, S. M. Y., & Rahman, M. S. (2008). Isolation and characterization of effective bacteria for solid waste degradation for organic manure. *Current Applied Science and Technology*, *8*(2), 44-55.

How to cite this article:

Kshama Deshpande, Vijay Deshmukh, Ashok Kharde and Pratiksha Nagmode. 2023. Production and Application of Bioculture for Farm Waste Recycling. *Int.J.Curr.Microbiol.App.Sci.* 12(02): 135-141.
doi: https://doi.org/10.20546/ijcmas.2023.1202.013