"Microbial Biodegradation of Pre and Post Consumed Food Waste using Thermophilic Amylolytic and Cellulolytic Bacterial Consortium"

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Abstract:

Food waste management is a worldwide environmental problem because of rising production and inappropriate disposal of food waste, which leads to resource depletion and greenhouse gas emissions. A thermophilic microbial culture was isolated and used to check the biodegradation of food waste with cellulase and amylase producing bacteria at their optimum conditions. Microorganisms were enriched on starch and carboxymethyl cellulose as selective substrates from food waste compost and cow manure compost in modified basal salt medium. Some selected isolates exhibited mean hydrolytic activity with the capability to degrade starch and cellulose.

The food waste was subjected to an anaerobic process at thermophilic temperature (60 ± 2°C) for 15 days using different proportions of food waste. Comparatively, the 1:1 ratio performed the best with a total organic matter removal of 85.64%. The decreases in TOC (Total organic carbon) and Carbon-Nitrogen ratio at all sites demonstrate a substantial mineralization process occurred, meanwhile the increase of total Kjeldahl nitrogen due to concentration effects. Germination index was 85% which indicates compost maturity and non-phytotoxicity.

In this study, we underline the promise of using thermophilic microbial consortia as an efficient and ecological step for food waste valorization and composting.

Introduction:

Food Waste appears as an important global problem with population growth, hyper-urbanisation, dynamic lifestyles, and inefficient food supply chain management in recent decades. Food waste leads to a significant loss of resources, including water, energy, nutrients, and agricultural elements up to the preparation stage at every stage of the creation of so-called feedback. At every stage of the food pathway—production, processing, transportation, retail, and food preparation for consumption—a sizeable amount of food is lost or wasted. India generates around 74 million tons of food waste annually from a range of sources, including homes, eateries, establishments, and the food processing industry. This poses a significant challenge to sustainable waste management (Pandey, 2019). Food waste management is a significant global challenge, especially in developing countries where poor collection systems and limited processing capabilities can place additional environmental pressure due to fast accumulation of waste (Gatto & Chepeliev, 2024). This poses a significant challenge to sustainable waste management (Pandey, 2019). Food waste management is a significant global challenge, especially in developing countries where poor

collection systems and limited processing capabilities can place additional environmental pressure due to fast accumulation of waste (Thi et al., 2015).

Improper disposal of food waste is one the major cause for environment pollution and climate change. Reactive systems typically dispose of organic food waste in landfills where anaerobic microbial decomposition occurs, resulting in the generation of methane, a greenhouse gas that has 25 times higher global warming potentials than carbon dioxide. Moreover, the disposal in landfill will create leachate which contaminates soil and groundwater around. Current disposal processes for these waste types are through incineration, which lowers the volume of invasive species but can also cause emissions from toxic pollutants including dioxins and other damaging gases. While it converts organic wastes into nutrient-rich compost and is therefore regarded as an environmentally sustainable option, traditional composting is also linked with long processing duration, odour production, nitrogen loss and inconsistent compost quality (Cerda et al., 2018), (Pham et al., 2022). As a result, there is a need for innovative biological approaches to enhance the efficacy of food waste treatment and resource recovery. (Pham et al., 2022).

Food waste are heterogeneous organic substrates predominated by carbohydrates, proteins, lipids, minerals and other bioactive compounds. More than 70% of food waste is carbohydrates, mainly starch and cellulose (30–60%), with proteins not accounting more than 5-10% and lipids from 10% to 40% (M. K. Awasthi et al., 2018). The Composition of food waste depends on the source of waste; for example, animal-based wastes such as meat and eggs have high levels of protein and lipids, while vegetable residues, rice waste, and cereal-based wastes are rich in starch and cellulose. They are complex organic polymers that can slowly be degraded only after enzymatic hydrolysis, it is a key process which involves microorganisms i.e. the microorganisms secrete extracellular enzymes capable of converting macromolecules to simple compound so they will be assimilate by cells as nutrient. (M. K. Awasthi et al., 2018).

Microbial based biodegradation has been revered as a promising eco-sustainable and effective food waste management strategy. Microorganisms use organic waste as a source of nutrients, and produce several hydrolytic enzymes involved in the biodegradation of carbohydrates, proteins, and lipids (S. K. Awasthi et al., 2025). The production of enzymes from microorganisms offers many advantages such as high catalytic activity, stability, low cost and easy scale-up compared to plant- and animal-derived enzymes (Nigam, 2013), (Adrio & Demain, 2014). Although many types of microbial enzymes are reported, it is

reasonable to assume that amylase and cellulase are necessary for food waste decomposition since starch and cellulose constitute the almost all structures found in organic wastes.

Amylase is most important starch-hydrolyzing enzymes, which catalyzes α-1,4-glycosidic bonds in starch molecules to produce dextrin, oligosaccharides and glucose. Microbial amylases are used widely for food processing, textile, paper detergent, pharmaceuticals and waste treatment industries as these are stable catalysts with efficient catalytic properties (Gupta et al., 2003), (Nigam, 2013). Another crucial enzyme of cellulose-decomposition is cellulase. Hydrolysis of cellulose is a stepwise process mediated by the action of endoglucanase, exoglycanase, and β-glucosidase enzymes. These enzymes work together to sequentially hydrolyze cellulose polymers into the disaccharide cellobiose and the monosaccharide glucose, which enables biomass conversion and organic waste degradation (Lynd et al., 2002), (Ibrahim et al., 2021).

Microbial biodegradation is influenced by environmental factors such as temperature, pH, oxygen supply, moisture content and substrate composition. This is mainly because thermophilic microorganisms are able to thrive at higher levels of temperatures and hence produce thermostable enzymes, making them extremely beneficial for food waste treatment. The organic matter of waste is much better decomposed by high-temperature degradation, and pathogen survival is also significantly reduced in this way. Thermophilic microorganisms can resist the effects of inhibitory compounds in food waste, including high concentrations of salts and fatty acids and thus would be more appropriate for complex waste degradation process. (M. K. Awasthi et al., 2018).

Bacillus species are notable food waste degrading microorganisms because of their ability to grow easily in the environment, high growth rate, and propensity for enzyme production. Genus Bacillus produces different types of extracellular enzymes, such as amylases, cellulases, proteases and lipases and is responsible for efficient degradation of the organic matter. Isolation of amylolytic and cellulolytic aerobic bacterial strains such as Brevibacillus borstelensis, Bacillus licheniformis, and Bacillus thuringiensis. The strains shown potential for enhancing biodegradation efficiency and promoting sustainable composting. (M. K. Awasthi et al., 2018).

Microbial cultures applied for food waste degradation are potential tools that allow nutrient recovery and decreased environmental emissions. Microbial-based treatment approaches enable the recycling of nutrients, biofertilizer (through compost), enzyme recovery and even bioenergy generation—features that support a circular economy concept (Cerda et al., 2018). Still, many aspects need to be studied before the industrial applications

of enzyme systems such as microbial community optimization, enzyme purification approaches by category including stability during processing such as freeze-drying/drying and storage, process scale-up both at laboratory level but also in a continuous operation point of view and adaptation to different compositions of food waste (Pham et al., 2022).

Thus, this study aims to isolate and screen microbial cultures for amylase and cellulase production potential, such that they can be applied to biodegrade pre-consumer and post-consumer food waste. Establishing efficient microbial-based degradation systems would supply a sustainable strategy for food waste disposal and environmental remediation.

Material and Methods:

1) Collection and Preparation of Compost Samples:

Food Waste compost (FWsC) samples were obtained from a windrow composting experiment initially established in Samples of cow manure compost (CMC) were collected from Bardoli, Surat, Gujarat, India. The composite samples were then taken out of the incubating bags, placed in sterile plastic bags and brought to lab under aseptic conditions. The compost samples were well-mixed and then maintained at 4°C until subsequent analyses and microbial isolation experiments. Compost is a well-known source of hydrolytic enzyme-producing microorganisms due to the presence of diverse microbial populations adapted to degrade complex organic substrates in compost environments (Bernal et al., 2009).

2) Seed Samples:

The germination index assay was performed using seeds of mung bean (Vigna radiata) and chickpea (Cicer arietinum) for the evaluation of maturity and potential phytotoxicity of the treated food waste compost. Seed germination and root elongation are among the most common bioassays used to assess compost and other organic soil amendments quality as well as their phytotoxic effects. Seed germination and root elongation are among the most common bioassays used to assess compost and other organic soil amendments quality as well as their phytotoxic effects (Oktiawan & Zaman, 2018).

3) Enrichment and isolation of thermophilic starch- and cellulose-degrading bacteria:

Enrichment based on initial isolation was performed from the compost samples in order to identify thermophilic organisms that had the ability to degrade both starch and cellulose. Selective enrichment of starch- and cellulose-utilizing bacteria was performed using modified basal salt medium (MBSM) (M. K. Awasthi et al., 2018).

Maltose-containing starch-degrading microorganism isolation MBSM formulate with a sole carbon source was made for each 100 mL distilled water containing the following (prepared using sterile instruments): $NaNO_3$ (0.2 g), KH_2PO_4 (0.2 g), $Na_2HPO_4 \cdot 12H_2O$ (0.2 g), $MgSO_4 \cdot 7H_2O$ (0.02 g), $CaCl_2 \cdot 2H_2O$ (0.01 g), $FeSO_4 \cdot 7H_2O$ (0.0001%) and NaCl: 80 mg/L

KCl: 80 mg/L 10 mL of the trace metal solution ($FeCl_3$, $ZnSO_4 \cdot 7H_2O$, $CoCl_2 \cdot H_2O$, $CuSO_4 \cdot 5H_2O$, $MnSO_4 \cdot 7H_2O$, H_3BO_3 and $Na_2MoO4 \cdot 2H_20$) was also added.

MBSM equilibrated with carboxymethyl cellulose (CMC) as a selective carbon source was used to enrich cellulose-degrading bacteria containing K_2HPO_4 (0.165 g), NH_4SO_4 (0.160 g), NaCl (0.096 g), yeast extract (0.5 g), cysteine (0.05 g), $CaCl_2$ (0.0096 g), $MgSO_4 \cdot 7H_2O$ (0.096 g), resazurin solution. 1 mL and trace element solutions 1 mL in 100 mL distilled water and CMC in 2%. 2.4% KamaBioA036 medium was further supplemented with 0.02% ketoconazole to balance the rate of fungal contamination and an appropriate volume of E-Gel additive (available in eGeltab Hs0053, Life Technologies, USA).

To determine the microbial composition of the compost isolates, about 1 g of compost sample was inoculated in a flask containing MBSM with either 2% starch or CMC dissolved to prepare 100 mL or different amount ratio. Cultures were incubated for 3 days under shaking conditions at 200 rpm. After the enrichment, 1 mL of culture broth was serially diluted and 0.1 mL aliquots were spread onto starch agar plates or CMC agar plates. After incubation for 24 h at 37°C, the starch- and cellulose-degrading bacterial colonies were isolated. Enrichment and isolation strategies were used to obtain amylolytic for food-waste biodegradation from compost and cellulolytic bacteria. Cellulose in microbial systems is a substrate for extracellular cellulolytic enzyme systems that hydrolyze cellulose into soluble sugars that can then be used by microorganisms. (Lynd et al., 2002).

4) Plate Assay for Screening of Amylolytic and Cellulolytic Bacteria

Starch agar and CMC agar plates were consecutively utilized for screening purified bacterial isolates against amylase and cellulase production, respectively. The carbon source used was either starch or CMC and individual colonies were point-inoculated on the respective agar media and incubated at 37°C for 24 h.

Following 10 days of incubation starch agar plates were flooded with Lugol's iodine solution for 5 min. The residue starch that remains in the medium reacts with iodine creating dark pigmentation while, starch hydrolysis produces a clear area around the bacterial colony. Thus, formation of a distinct hydrolytic zone was regarded as a positive test for amylase production.

Cellulolytic activity on CMC agar plates was assessed by flooding with 0.1% Congo red solution for 5 min and washing with 1 M NaCl solution for 20 min. A clear zone was formed in the surrounding area of a bacterial colony, which implies cellulose degradation and constant cellulase production. We use in plate-based hydrolysis assays to screen for extracellular amylase- and cellulose-producing microorganisms. Microorganisms possessing

cellulose hydrolysis capacity consist of extracellular cellulase systems with diverse catalytic functions which synergistically work together on cellulose degradation (Lynd et al., 2002).

5) Establishment of the Microbial Consortium and Biodegradation Experiment with the Food Waste:

A microbial consortium had been developed using the selected bacterial isolates with high amylolytic and cellulolytic potential. An optical density of about 1 (6.8×10^6 CFU/mL) of the bacterial consortium was prepared and inoculated with approximately 10 mL of the bacterial suspension into a mass of food waste equal to 100 g.

These inoculated food waste samples were incubated in aerobic at 60 ± 2°C for a period of 15 days. Each treatment had three independent replicates. The aerobic microbiome community was established by aerating the incubated food waste samples with a sterile spatula under aseptic conditions during incubation.

The application of mixed microbial cultures which include amylolytic and cellulolytic bacteria is also relevant to the biodegradation of food-waste as this material primarily consists of complex carbohydrate polymers requiring complementary enzymatic activities. Consortium with amylase- and cellulase-producing strains was able to enhance the degradation of pre- and post-consumer food waste–with a 1:1 mixture exhibiting particularly effective degradation.

Higher incubating temperatures have been purposely selected to promote thermotolerant or even thermophilic microorganisms and their thermostable hydrolytic enzymes. They could be potentially advantageous for organic waste treatment because thermophilic temperatures favor rapid biodegradation and kill undesirable microorganisms (M. K. Awasthi et al., 2018).

6) Physico-chemical Determinations on Naturally Biodegraded Food Waste

The biodegraded food waste as characterized in terms of pH, electrical conductivity (EC), total Kjeldahl nitrogen (TKN), total organic matter (TOM), total organic carbon (TOC) and moisture content according to a standard method for compost and organic waste characterization. Variation of these physicochemical parameters indicates organic matter degradation, nutrient transformation/stabilization and quality of end compost material. (Cerda et al., 2018), (Qasim et al., 2019).

6.1 pH and Electrical Conductivity:

A sample of 7 g in a conical flask was mixed with 100 mL distilled water on a reciprocating shaker for 30 min. The suspension was kept for 10 min and then filtered using filter paper. The values of pH and EC of the filtrate were measured by calibrated pH meter,

conductivity meters respectively. Electrodes were rinsed with deionized water after measuring.

The pH during biodegradation correlates with the production and further utilization of organic acids, while EC reflects the accumulation of soluble ionic constituents released during organic matter decomposition. (Qasim et al., 2019).

6.2 Total Kjeldahl Nitrogen (TKN):

To measure TKN, 1 g powdered sample was added in a digestion tube to which 10 mL distilled water was added. To this, 3 g of catalyst mixture approximately and (10 mL) concentrated sulfuric acid were then added. The sample was digested at elevated temperature until complete digestion occured, as monitored by a clear green digest. The cooled digested sample was analyzed by the correct distillation/titration method, and nitrogen content was determined as per standard TKN method. The cooled digested sample was analyzed by the correct distillation/titration method, and nitrogen content was determined as per standard TKN method (Hicks et al., 2022).

7) Total Organic Matter (TOM):

The sample weight before and after drying for 24 h in a hot-air oven is determined followed by the weighing of dried sample which was then put inside crucible and heated (2 h) with stepwise temperature increment in the furnace, cooled down in a desiccator and weighed.

The TOM was calculated using the following formula:

$$TOM\ (\%) = [(W_2 - W_3) / (W_2 - W_1)] \times 100$$

Where W_1 is the weight of blank container, W_2 means the weight of container and dried sample, and W_3 is what we have weighed after furnish treat. where W_1 is the weight of blank container, W_2 means the weight of container and dried sample, and W_3 is what we have weighed after furnish treat (Navarro et al., 1993).

Microbial fermentation treatment increases the biodegradability by reducing the organic matter which can be used as an index for biodegradability performance. Microbial fermentation treatment increases the biodegradability by reducing the organic matter which can be used as an index for biodegradability performance (M. K. Awasthi et al., 2018).

8) Total Organic Carbon

TOC was estimated from organic matter using following formula:

$$TOC\ (\%) = Organic\ Matter\ (\%) / 2$$

The reduction in TOC during biodegradation is indicative of the mineralization and microbial utilization of organic substrates that contain carbon. (Navarro et al., 1993).

9) Moisture Content

Moisture content was determined by weighing the sample before and after subjecting it to oven drying for 24 h. The moisture content was estimated using following formula:

$$\text{Moisture Content (\%)} = [(W_2 - W_3) / (W_2 - W_1)] \times 100$$

where W_1 is empty vessel weight; W_2 is the vessel with initial sample; and W_3 is dry weight.

For microbial development and the mineralization of organic waste during composting and biodegradation, moisture is essential. (LANG & STEINBERG, 1980).

10) Germination Index Assay

Method to Assess Maturity and Phytotoxicity of Biodegraded Food Waste Using a Germination Index (GI) Assay with Mung Bean Seeds A total of 7 grams of sample was stirred with 100 mL distilled water for 30 min. The suspension was filtered after standing for 10 min.

In sterile Petri plates layered with filter paper 10 ml of the extract was added. Ten mung bean seeds were distributed uniformly onto the filter paper and kept in the dark at 25 °C for 72 h, after that numbers of germinated seed and root length were measured.

Germination index was calculated using following formula:

$$\text{GI Index (\%)} = \frac{[\textit{Seed germination (\%) (Treatment)} \times \textit{Root length (Treatment)}]}{[\textit{Seed germination (\%) (Control)} \times \textit{Root length (Control)}]} \text{ X } 100$$

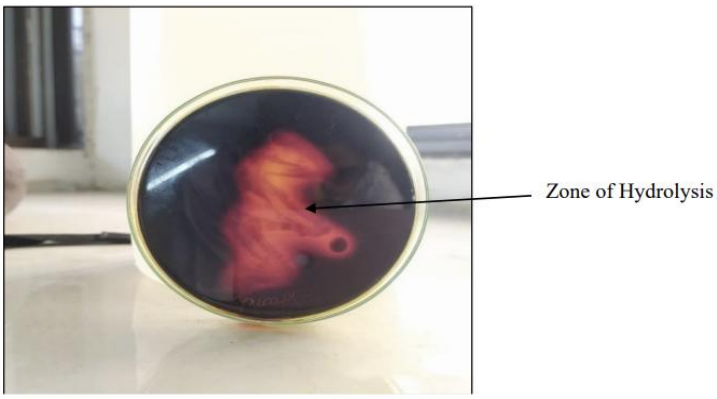
Germination Index is known as biological indexes which can be used to evaluate maturity and phytotoxicity of compost, because residual organic acids, salts, ammonia and other phytotoxic compounds can prevent or inhibit seed germination and rooting (Cerda et al., 2018)

Result:

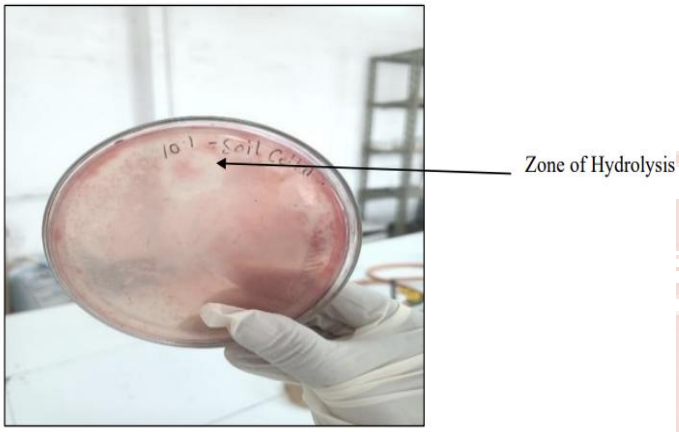
1. Isolation of starch- and cellulose-degrading bacteria from compost samples

Bacterial isolates were obtained from collected food waste compost (FWsC), and cow manure compost (CMC) samples The isolates exhibited clear hydrolytic zones on starch and CMC agar media, indicating starch and cellulose degrading ability, respectively.

On starch agar that had been treated with iodine, the starch-hydrolyzing amylase-producing organisms formed clear zones around their colonies. Regions in which starch persisted (was resisted hydrolysis) reacted positively with iodine and was colored black; whilst those regions in which this substrate was hydrolyzed remained clear. Likewise, isolates degrading cellulose caused turbid (or clear) hydrolytic zones on CMC agar showing cellulase activity.



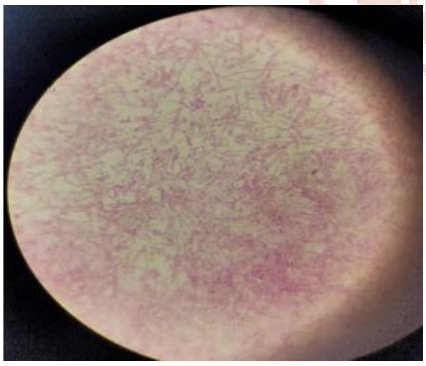
[Figure 1: Zone of hydrolysis produced by bacteria on agar plates containing Starch]

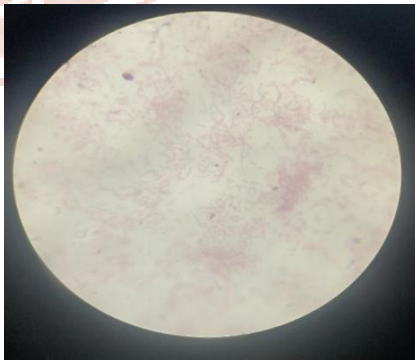


[Figure 2: Zone of hydrolysis produced by bacteria on agar plates containing CMC]

2. Gram staining and morphological characterization:

Gram staining was used to determine the morphological characteristics of isolation for selected isolates. All starch- and cellulose-degrading isolates were Gram-stained, tested for shape through microscopy and recorded maintaining a pink colour. On the basis of these features, both isolates were classified as Gram negative, rod-shaped bacteria.





[A] Starch Degrading Bacteria **[B] Cellulose Degrading Bacteria**

[Figure 3: Gram's staining results]

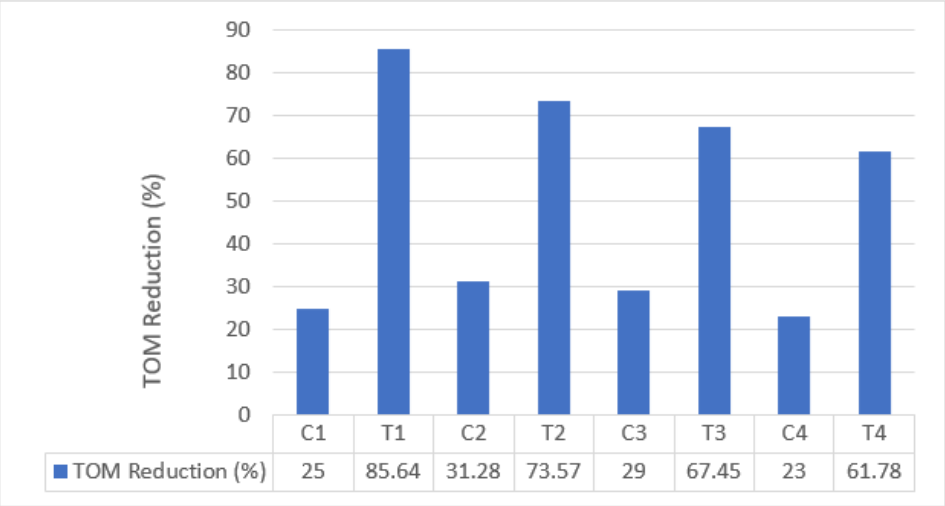
3. Bacterial Consortium for Biodegradation of Food Waste:

Two starch- and cellulose-degrading bacteria were used in a mixed consortium to conduct a small-scale flask experiment and screen rapidly biodegradable food-waste compositions. Four inoculated treatments (T1–T4) and their respective controls were included in the experimental design.

[Table 1: Treatment design to study biodegradation of food waste]

Treatment	Mixing Ratio	Food Waste Composition
Control-C1	1:1	Pre-consumed + post-consumed food waste
Treatment-1 (T1)	1:1	Pre-consumed + post-consumed food waste
Control-C2	1:2	Pre-consumed + post-consumed food waste
Treatment-2 (T2)	1:2	Pre-consumed + post-consumed food waste
Control-C3	—	Pre-consumed food waste
Treatment-3 (T3)	—	Pre-consumed food waste
Control-C4	—	Post-consumed food waste
Treatment-4 (T4)	—	Post-consumed food waste

About 10 mL of the bacterial cell suspension was inoculated into the 100 g food waste. Fifteen days of thermophilic incubation with equal proportions mixed amylolytic and cellulolytic bacteria. The bacterial consortium was effective for the degradation of food waste, where the total organic matter (TOM) degradation was significantly higher in T1 (85.54%) than T2 (73.57%), T3(67.45%) and T4(61.78%). The greater degradation in T1 could be related to the balanced availability of substrates present in the 1:25 mixture, which provided good microbial growth and supported enzymatic activity. Microbial Community and Mechanisms Differences among Treatments may Also be related to Food Waste Composition, Accumulated Short chain Fatty Acids (VFA).

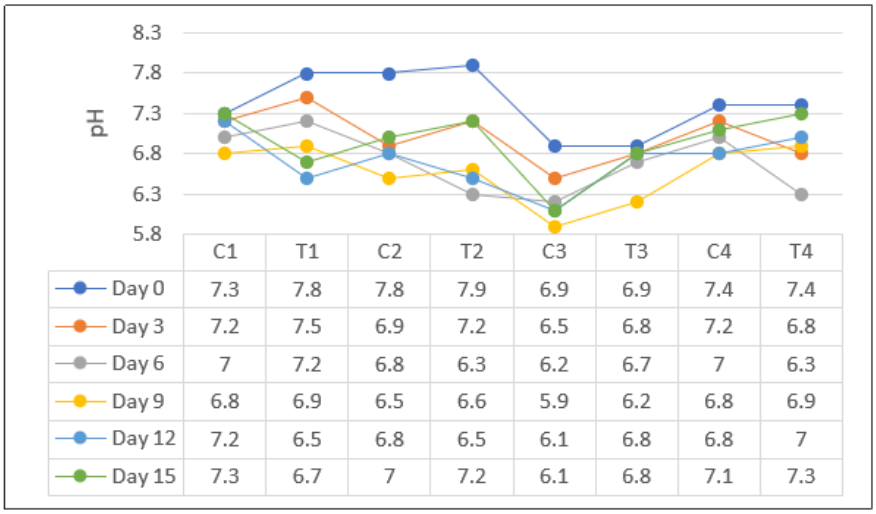


[Figure 4: The reduction in Total Organic Matter (TOM) after 15 days]

The T1 degradation was the most reduced which demonstrates that a balanced substrate promotes microbial growth and enzyme disintegration activity.

3.1 pH and Electrical Conductivity:

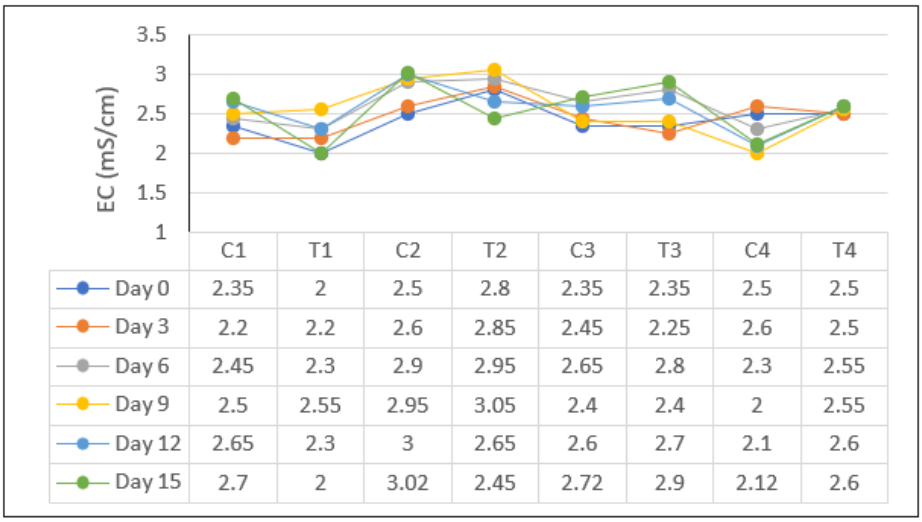
During the 15 days of biodegradation, the pH in all treatments varied. The original pH values were in the ranges of between 6.9 and 7.9. In the early stage of the degradation process, pH often decreased, which might be due to the production of organic acids and volatile fatty acids through microbial metabolism. After about 9 days, a progressive increase in pH was observed for some treatments inoculated. In contrast, some control treatments had lower pH values due presumably to slower microbial degradation and persistent accumulation of organic acids.



	C1	T1	C2	T2	C3	T3	C4	T4
Day 0	7.3	7.8	7.8	7.9	6.9	6.9	7.4	7.4
Day 3	7.2	7.5	6.9	7.2	6.5	6.8	7.2	6.8
Day 6	7	7.2	6.8	6.3	6.2	6.7	7	6.3
Day 9	6.8	6.9	6.5	6.6	5.9	6.2	6.8	6.9
Day 12	7.2	6.5	6.8	6.5	6.1	6.8	6.8	7
Day 15	7.3	6.7	7	7.2	6.1	6.8	7.1	7.3

[Figure 5: Changes in pH during food waste biodegradation]

During the entire 15-day period of biodegradation monitoring, electrical conductivity (EC) was tracked to investigate changes in soluble ion concentration released during microbial degradation. The EC parameters were different at the treatments confirming that organic matter was transformed and mineralized differently.



	C1	T1	C2	T2	C3	T3	C4	T4
Day 0	2.35	2	2.5	2.8	2.35	2.35	2.5	2.5
Day 3	2.2	2.2	2.6	2.85	2.45	2.25	2.6	2.5
Day 6	2.45	2.3	2.9	2.95	2.65	2.8	2.3	2.55
Day 9	2.5	2.55	2.95	3.05	2.4	2.4	2	2.55
Day 12	2.65	2.3	3	2.65	2.6	2.7	2.1	2.6
Day 15	2.7	2	3.02	2.45	2.72	2.9	2.12	2.6

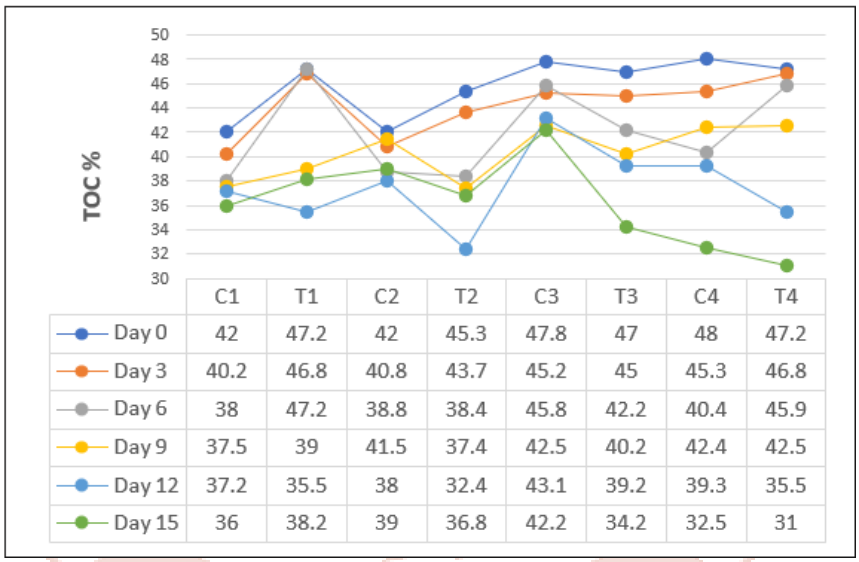
[Figure 6: Changes in Electrical Conductivity during Biodegradation]

4. Total organic carbon, Total nitrogen and C/N ratio:

Total organic carbon (TOC), Total Kjeldahl nitrogen (TKN) and C/N ratio during the 15-day of biodegradation period are presented in table. TOC in all treatments demonstrated a structural change over the time of incubation. 31.28% in T3, 37.53 % in T1, 39.63% in T4 and 42.29% in T2 were the reported TOC reduction values obtained.

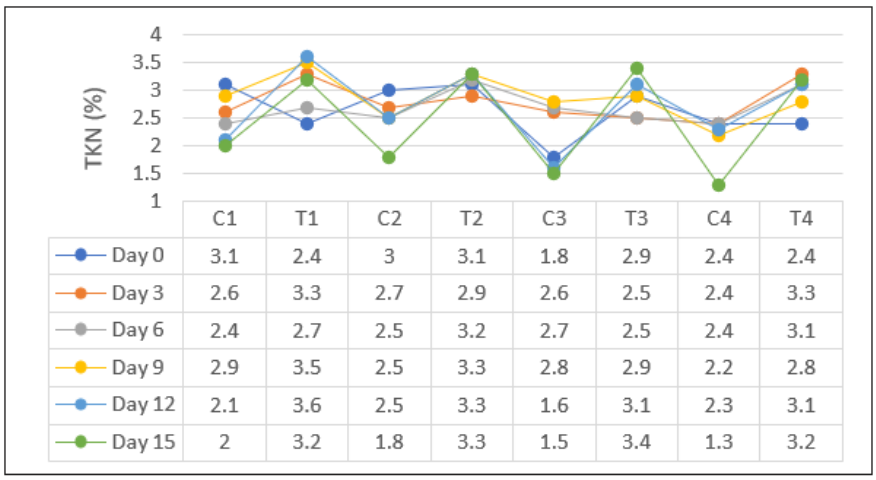
During the incubation, a wide range of TKN values was observed. The total Kjeldahl nitrogen (TKN) was higher in the inoculated treatments, around 3.6% for T1, 3.3% for T2, 3.4% for T3 and 3.2% for T4 at Day15. TKN values at the end of incubation for control treatments were comparatively different.

During the degradation period, the C/N ratio varied. C/N ratios at day 15 were about 16.4, 12.1, 21.8, 19.6, 28.0, 18.0, 24.1 and 18.8 for C1, T1, C2, T2, C3, T3, C4 and T4 Respectively.



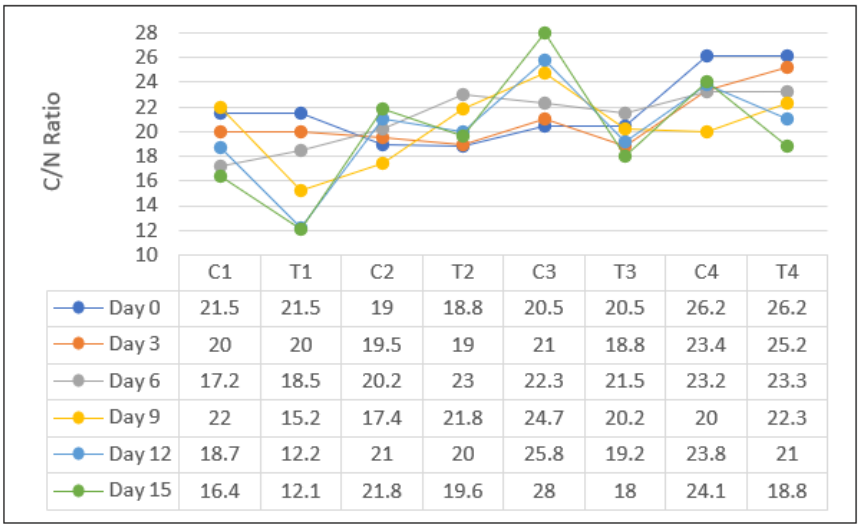
	C1	T1	C2	T2	C3	T3	C4	T4
Day 0	42	47.2	42	45.3	47.8	47	48	47.2
Day 3	40.2	46.8	40.8	43.7	45.2	45	45.3	46.8
Day 6	38	47.2	38.8	38.4	45.8	42.2	40.4	45.9
Day 9	37.5	39	41.5	37.4	42.5	40.2	42.4	42.5
Day 12	37.2	35.5	38	32.4	43.1	39.2	39.3	35.5
Day 15	36	38.2	39	36.8	42.2	34.2	32.5	31

[Figure 7: Reduction in Total Organic Carbon (TOC)]



	C1	T1	C2	T2	C3	T3	C4	T4
Day 0	3.1	2.4	3	3.1	1.8	2.9	2.4	2.4
Day 3	2.6	3.3	2.7	2.9	2.6	2.5	2.4	3.3
Day 6	2.4	2.7	2.5	3.2	2.7	2.5	2.4	3.1
Day 9	2.9	3.5	2.5	3.3	2.8	2.9	2.2	2.8
Day 12	2.1	3.6	2.5	3.3	1.6	3.1	2.3	3.1
Day 15	2	3.2	1.8	3.3	1.5	3.4	1.3	3.2

[Figure 8: Total Kjeldahl nitrogen (TKN) Value During Biodegradation]



	C1	T1	C2	T2	C3	T3	C4	T4
Day 0	21.5	21.5	19	18.8	20.5	20.5	26.2	26.2
Day 3	20	20	19.5	19	21	18.8	23.4	25.2
Day 6	17.2	18.5	20.2	23	22.3	21.5	23.2	23.3
Day 9	22	15.2	17.4	21.8	24.7	20.2	20	22.3
Day 12	18.7	12.2	21	20	25.8	19.2	23.8	21
Day 15	16.4	12.1	21.8	19.6	28	18	24.1	18.8

[Figure 9: C/N Ratio During Biodegradation]

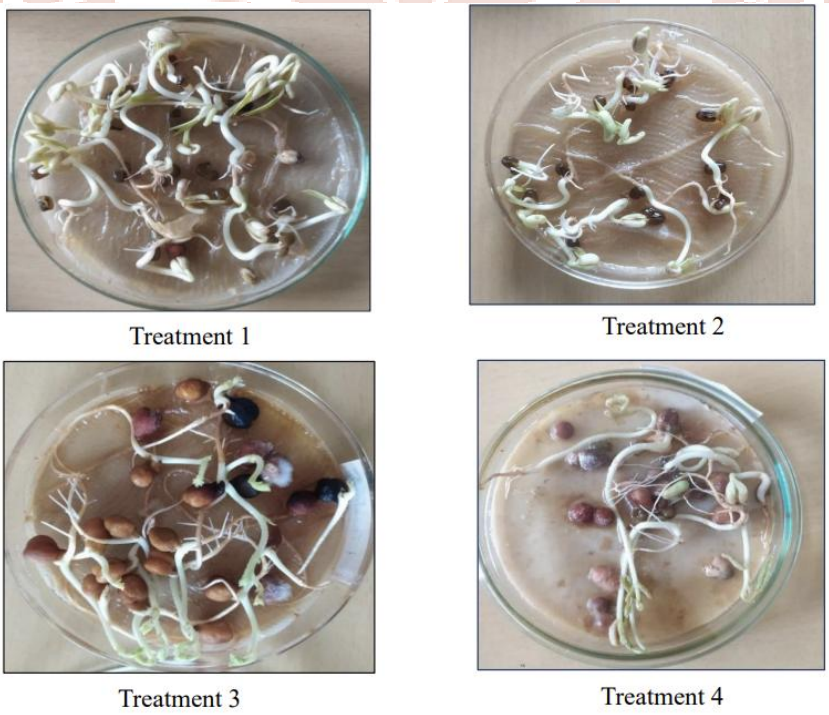
5. Evaluations of Germination Index and Phytotoxicity:

Phytotoxicity of the compost samples was evaluated by GI, which were measured after a 15-day incubation period. There were differences in GI between the tested treatments, being greater for T1 (85%), followed by T3 (76.7%), T2 (67.4%) and T4 (55.8%). T1 vigor differences versus the other treatments.

[Table 2: Germination Index (GI) for Compost Maturity]

Treatment	Germination Index (%)	Compost Quality
T1	85.0	Non-Phytotoxic
T3	76.7	Moderately Mature
T2	67.4	Partially Mature
T4	55.8	Immature/Phytotoxic

The GI from T1 was more than 80% while GI values in other treatments were less than 80%.



[Figure 10: Germination index of all compost]

Discussion:

The results of this study suggested the potential of compost-derived bacterial cultures to biodegrade food waste via amylolytic and cellulolytic activities. Confirmation of Hydrolytic Activity towards Complex Polysaccharides on Starch and CMC Agar. Clear hydrolytic zones observed with the selected isolates on both starch and CMC agar indicate their ability to break down polysaccharides.

A thermophilic mixed bacterial consortium improves the degradation of food waste. Of the treatments tested, T1 showed the highest TOM degradation at 85.64%, followed by T2 (73.57%), T3 (67.45%) and T4 (61.78%). The better degradation of starch in PA seems to be linked to a balanced availability of substrates and a more active microbial biomass.

Initially, pH decreased during degradation probably due to the generation of volatile fatty acids and increased after the ninth day in inoculated treatments, signalling continually stabilization. Driven by the solubility of mineral nutrients released during organic matter decomposition, EC generally increased in those treatments and then slowly declined for T1 and T2 when going toward the later stage.

While TKN rises show microbial mineralization and a concurrent concentration of nitrogen through the breakdown of organic matter, TOC reductions by the treatments show a tendency of decline. The decline in the C/N ratio then provided further assistance for the stability of food waste.

The GI values were 85.0% (T1), 67.4% (T2), 76.7% (T3) and 55.8% (T4) for final time points, respectively. Low phytotoxicity is evidence of compost maturity, and T1 presented a GI > 80%.

In conclusion, the selected bacterial consortium could accelerate food waste degradation and promote organic matter stabilization. Overall, microbial consortia were able to reduce food waste in a sustainable manner, with the T1 treatment performing most favourably across all measured parameters.

Conclusion:

The findings of this study demonstrated the viability of employing thermophilic bacterial cultures for effective food waste biodegradation. With a total decrease of 85.64% at a 1:1 ratio of pre-consumed and post-consumed food waste, the results showed that the mixed microbial consortium (T1) significantly aided in the decomposition of organic matter under thermophilic circumstances. Microbial decomposition and stabilization of organic matter were validated by changes in pH, EC, TOC, TKN, and the C/N ratio. Additionally, T1's germination index was higher (85%), which offered more proof of reduced phytotoxicity and

more fully developed compost. In summary, the results offer evidence-based support for nutrient recovery via microbial consortia and environmentally friendly and sustainable food waste management.

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