

# Eco-Friendly Microbial Growth Media From Sugarcane Bagasse And Fruit Peels: Optimizing Natural Substrates For Sustainable Cultivation

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## Abstract:

Fruit and vegetable waste (FVW) poses a significant challenge in waste management due to its rapid biodegradability and the substantial quantities generated daily, often leading to environmental issues. Fruit processing residues contain high levels of phenolic compounds and have a wide range of industrial applications in food, cosmetic and pharmaceutical industries. This study aims to formulate media for growth of various microbes using fruits peel such as banana peels, papaya peels and orange peels with sugarcane bagasse. Formulating media for growth of different microbes such as *Staphylococcus aureus*, *Bacillus subtilis*, *E. coli*, *Actinomyces*. This study helps to formulate a good potential media from these agricultural byproducts as alternative substrates for cultivating various microbes, focused on optimizing the nutrient content of agricultural waste to support microbial growth and activity. This study also explores that with adequate amount of agar, bagasse and fruit peels can successfully promote the growth of various microbial species, potentially decreasing the need for synthetic and expensive media components. It proves to be an eco-friendly approach for resource utilization and waste management as the mixture of sugarcane bagasse and fruit peels (citrus, banana, papaya, and sugarcane bagasse) shows best results for growing microbes rather than other combination of peels (bagasse and banana peel, bagasse and papaya peels, bagasse and orange peel) and the growth of microbes was analysed by spectrophotometer at 600nm wavelength at the different intervals of time, were *Staphylococcus aureus* showed maximum growth of  $1.081(\text{cfu}/\text{mL}10^8\text{-}10^9)$ , *Actinomyces* showed  $1.326(\text{cfu}/\text{mL}10^8\text{-}10^9)$  growth, *E.coli*  $1.168(\text{cfu}/\text{mL}10^8\text{-}10^9)$ . This method will not only support sustainable bioprocesses but will also provide a valuable use for agricultural waste.

**Key words:** Agriculture; Bagasse; Eco friendly; Microbes; Media; Peels

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## 1. INTRODUCTION

Fruit and vegetable waste (FVW) represents a significant challenge in waste management due to its high biodegradability and the vast quantities generated daily. At municipal landfills, this waste often leads to problems such as unpleasant odours, attraction of pests, and accelerated decomposition, which can further strain waste management systems (Misi et al., 2002). One of the primary sources of FVW is central food distribution markets, which handle large volumes of perishable goods. These markets, which include sections for meat, fish, fruits, and vegetables, contribute substantially to the overall waste stream. For instance, in a typical central food distribution market, the total waste generated, encompassing all categories, amounts to approximately 90 tonnes per day. (Mata et al., 1992). On a broader scale, the situation in India is even more alarming. It is estimated that around 5.6 million tonnes of fruit and vegetable waste are produced annually across the country. This enormous volume of waste poses significant environmental, economic, and logistical challenges, especially since most of it is improperly managed (Bouallagui et al., 2003). Currently, a large proportion of this waste is either left to decay in open dumps outside city limits or sent to already overburdened municipal landfills. Such practices lead to multiple problems: Environmental pollution through foul odours, leachate generation, and methane emissions contributing to greenhouse gases, Health hazards due to pest infestation, flies, and rodents, causing the spread of diseases and Wastage of potential resources, as these organic wastes are rich in nutrients and energy-yielding compounds that can be converted into valuable products. If managed scientifically, this waste could be utilised for sustainable applications such as composting, vermicomposting, animal feed, bioethanol production, or biogas generation, promoting a circular economy approach.

For instance, papaya is one of the major fruits contributing to waste volumes. The top producers of papaya in 2018 were Mexico (1.04 million tonnes), Brazil (1.06 million tonnes), and India (5.99 million tonnes) (Vazquez et al., 1999). Papaya fruit is used in various forms such as fresh consumption, juice production, ice cream, dried papaya products, sweets, jams, and bakery formulations, which increases its demand globally and within domestic markets (Zhou et al., 2021). However, due to its high perishability and poor post-harvest management, large quantities of papaya are discarded during handling, transportation, and market distribution. This not only results in economic loss to farmers and traders but also adds to the environmental burden when improperly disposed of (Zhou et al., 2021). Citrus (*Citrus limetta*, family Rutaceae) is the most produced fruit globally, accounting for 23% of global fruit production. Citrus peel (CP) produces 0.5–3 kg of essential oil per tonne of fruit. These oils are used in medicines, cosmetics, food preservation, and beverages. Citrus Peels are also rich in pectin (Zhou et al., 2021).

Fruit processing companies frequently dump large amounts of fruit waste into rivers or landfills, posing environmental risks. As a result, there is an urgent need for disposal techniques that recycle this waste, produce resources and develop products with added value (Srilatha et al., 1995). Improper handling of these wastes has far-reaching implications, including the release of greenhouse gases like methane during decomposition, contamination of soil and water resources, and an increased burden on municipal waste management systems. Addressing this challenge requires a shift toward innovative and sustainable waste management strategies, such as converting FVW into valuable products like biofertilizers, animal feed, or renewable energy sources (Abbas, 2021). Efforts to tackle FVW must also focus on reducing waste generation at the source, promoting awareness about waste segregation, and implementing policy measures to encourage the adoption of circular economy practices. By leveraging the biodegradable nature of FVW, there is significant potential to transform what is currently seen as an annoyance into an opportunity for resource recovery and environmental conservation (Harvey et al., 2007).

Agricultural wastes (such as crop residues, peels, husks, and bagasse) and their industrial by-products (like molasses from sugar industries or oilseed cakes) are often discarded without any significant value. However, these wastes are rich in organic compounds, fibres, sugars, and bioactive components, making them attractive sources for developing value-added products. For example: Lignocellulosic wastes (straw, bagasse, peels) can be converted into bioethanol, biogas, eco-friendly media for microbes and bioplastics (Abdelraof et al., 2019).

Bagasse, the fibrous by-product from sugarcane processing, has traditionally been employed primarily as a fuel in sugar factory boilers and to a lesser extent in manufacturing paper and board (Huang et al., 2012). Despite its widespread use for energy generation, bagasse has a low calorific value, making it an inefficient fuel source compared to alternatives (Verma et al., 2012). Currently, over 85% of bagasse is burned as fuel, leaving a considerable surplus of this material. This creates an urgent need to identify new applications for bagasse to reduce waste and prevent its accumulation in landfills. There are nearly 571 sugar mills in India, producing about 19.2 million tons of sugarcane. One alternative use is ethanol production, where only about 9% of bagasse is utilized (Parameswaran, 2009). Fruit and vegetable wastes contain bioactive compounds including natural pigments, antioxidants, and antimicrobials that can be extracted for applications in food safety, pharmaceuticals, cosmetics, and textiles. Papaya waste, in particular, has been used as a substrate in microbial fuel cells (MFCs), offering an inexpensive and sustainable option for generating renewable energy (Joymak et al., 2021). These approaches align with circular economy principles by addressing waste management, energy generation, and bioactive compound recovery (Pathak et al., 2017). Agro-industrial waste, especially from fruits and vegetables, is highly diverse and holds promise across industries. These wastes contain valuable fibres, polyphenols, and bioactives that serve as antioxidants and functional ingredients in health products (Kebaili et al., 2021). Fruit and vegetable waste is increasingly being studied as a component in microbial media. Their natural composition, including carbohydrates, proteins, lipids, and minerals, makes them ideal alternatives to synthetic media in microbial industries (Balaji et al., 2014). Microorganisms play crucial roles in industries such as pharmaceuticals, food, agriculture, and biotechnology. They are essential for producing fermented foods, antibiotics, vaccines, and other medicinal products (Ferreira et al., 2018). For optimal growth and reproduction, microbes require sources of energy, carbon, nitrogen, phosphorus, sulfur, and other minerals (Adesemoye et al., 2005). Culture media, designed to support microbial growth, can be expensive. Therefore, there is a need for low-cost, eco-friendly alternatives derived from agricultural and kitchen waste (Tenore et al., 2022; Berde et al., 2018).

Fruit and vegetable residues such as peels, seeds, and cores are rich in phenolic compounds and sugars, making them suitable for microbial cultivation and the production of bioethanol, biogas, and animal feed (Cruz et al., 2020)

This study is based on the formulation of media for cultivating microbes such as *Staphylococcus aureus*, *Bacillus subtilis*, *E. coli*, and *Actinomyces* using sugarcane bagasse, citrus, papaya, and banana peels. These raw materials are selected for their availability and nutrient-rich composition (Roshan et al., 2014). Sugarcane bagasse comprises approximately 42% cellulose, 28% hemicellulose, 20% lignin, 4.6% other polysaccharides, 3% sucrose, and 2.4% ash. It also contains 48% fibres and 2% soluble sugars (Sun et al., 2024; Dmitriev et al., 2004).

**Table 1: Comparison of Various Lignocellulosic Materials for Bioethanol Production (%)**

Lignocellulosic Material	Hemicellulose	Cellulose	Lignin	References
Banana peels	14.8	13.2	14	(Tyagi et al., 2009)
Papaya peels	12	13.2	31	(Alonso et al., 2025)
Orange peels	29	40	20	(Rosado et al., 2023)
Sugarcane bagasse	28	42	40	(Sun et al., 2024; Dmitriev et al., 2004).

Citrus peels are rich in phenolics, flavanones, carotenoids, and ascorbic acid. Banana peels contain high levels of dietary fibre and phenolic compounds (Basavaraju et al., 2022). Papaya peels are abundant in alkaloids, saponins, tannins, anthraquinones, carotenoids, phenolics, and flavonoids (Kalakoutsii et al., 1976).

*Staphylococcus aureus* is Gram-positive, with a thick peptidoglycan wall comprising wall-associated proteins, teichoic acids, and murein (Mondal et al., 2012). *E. coli* is a Gram-negative, non-sporulating, rod-shaped bacterium commonly found in food and the intestines of warm-blooded animals (McBirney et al., 2016). *Actinomyces* are Gram-positive, filamentous bacteria belonging to the phylum Actinobacteria, known for their complex life cycles and ecological significance (Gorinstein et al., 2001).

This formulation of media is eco-friendly, cost-effective alternative to synthetic culture media and offering sustainable solutions to food waste management and microbial cultivation.

Therefore, the following four materials were selected as ideal raw materials for the present study: banana peels, papaya peels, orange peels, and sugarcane bagasse.

## 2. MATERIAL AND METHODS:

Sugarcane bagasse was collected from local villagers of Bijhari and fruit peels were collected from juice shops of Bijhari, District Hamirpur (H.P.) and all the microbes **a)** *Staphylococcus aureus* **b)** *Bacillus subtilis* **c)** *Actinomyces* **d)** *Escherichia* were procured from the department of Microbiology, Career Point University Hamirpur (H.P.) Fruit peels i.e. citrus peels, banana peels, papaya peels and sugarcane bagasse were dried for 2-3 days in sunlight, separately. All the peels were grounded into the peels to powder and above powders was used in appropriate proportion to prepare media.

### 2.1 Materials and Ingredients:

Citrus peel powder, Banana peel powder, Papaya peel powder, Sugarcane bagasse powder each component was taken in equal proportion. Additionally, distilled water and agar were added.

#### 2.1.2 Measurement of Ingredients:

Four different combinations of the powders were prepared, with each containing specific ingredients measured precisely:

Sample A: 0.5 g sugarcane bagasse powder and 0.5 g banana peel powder, Sample B: 0.5 g sugarcane bagasse powder and 0.5 g citrus peel powder, Sample C: 0.5 g sugarcane bagasse powder and 0.5 g papaya peel powder and Sample D: A mixture of all four powders, with 0.5 g of each (citrus peel, banana peel, papaya peel and sugarcane bagasse).

**Table 2: Composition of different media**

Sr.no	Sugarcane bagasse	Citrus peel	Papaya peel	Banana peel	Agar	Distilled water
a)	0.5g	0.5g	-	-	2 g	100 ml
b)	0.5g	-	-	0.5g	2 g	100 ml
c)	0.5g	-	0.5g	-	2 g	100 ml
d)	0.5g	0.5g	0.5g	0.5g	2 g	100 ml

### 2.1.3 Sample Preparation:

100 ml of distilled water was added to each combination of powders. The mixtures were stirred briefly to ensure uniform wetting of the powders and the hydrated mixtures were left undisturbed at room temperature for 30 minutes to allow for extraction of nutrients into the water.

After that, each mixture was filtered through filter paper to remove insoluble residue and obtain a clear filtrate containing dissolved nutrients. The filtrate served as the liquid base for media preparation. To each filtrate, 2 g of agar was added as a solidifying agent. The mixtures were heated gently while stirring until the agar dissolved completely. The media preparations were transferred into sterilization vessels (e.g., flasks or bottles). Sterilization was carried out in an autoclave at 121 °C and 15 PSI for 15 minutes to eliminate any contaminants. Post sterilization, the molten media was carefully poured into sterile petri dishes under aseptic conditions. The petri dishes were left undisturbed at room temperature until the media cooled and solidified (Rakholiya et al., 2014).

The prepared media, enriched with nutrients extracted from fruit and sugarcane waste powders, was ready for microbial inoculation and growth studies. The use of agro-waste materials as nutrient sources promotes sustainability and resource efficiency in microbiological research.

### 2.1.4 Microbial Growth Assessment:

Bacterial cultures, including *Bacillus sp.*, *Escherichia coli* (*E. coli*), *Staphylococcus aureus*, and *Actinomyces*, were selected for analysis. Each bacterial species was streaked individually onto prepared media plates. The media plates were prepared using various fruit peel powders (citrus, banana, papaya) combined with sugarcane bagasse, as well as on control nutrient agar plates.

The inoculated plates were then incubated under ambient conditions, specifically at temperatures between 30-32°C, for an incubation period of 48 to 72 hours. After the interval of 72 the colony morphology and characteristics of each bacterial species were observed and recorded. Colony morphology analysis was performed using the streak plate method on both the fruit peel powder-based agar media and nutrient agar to compare growth characteristics across media types (Rakholiya et al., 2014).

Additionally, to evaluate bacterial growth in liquid culture, a broth medium was prepared, and bacterial cultures were grown in this liquid broth. Bacterial growth in the broth was measured spectrophotometrically at an optical density (OD) of 600 nm, which provided an indication of the bacterial cell concentration in the medium over time. Comparative optical density of microbes at different time intervals

$$\text{CFU/ml} = \text{No. of colony} \times \text{Dilution factor} \times \text{Volume of culture plated}$$

This formula is used to determine the colony-forming units (CFU) per millilitre of a microbial culture.

### Relationship Between Absorbance and CFU/ml:

The absorbance measured using a spectrophotometer provides an indirect estimate of microbial growth. For example, an absorbance between 0.1 and 0.5 corresponds approximately to a cell concentration of  $10^6$  to  $10^7$  CFU/ml. An absorbance between 0.5 and 1.0 corresponds to roughly  $10^7$  to  $10^8$  CFU/ml, while an absorbance between 1.0 and 2.0 indicates a cell density of approximately  $10^8$  to  $10^9$  CFU/ml. These values represent general estimates and may vary depending on the microbial species and experimental conditions.

## 3. RESULTS AND DISCUSSION:

Banana peels are nutritionally dense, containing approximately 6–9% protein, 3.8–11% crude fat, and a high dietary fibre content of 43.2–49.7%. (Fujikawa et al., 1989). The nutrients present in the peels are the essential requirements in a suitable microbial medium growth.

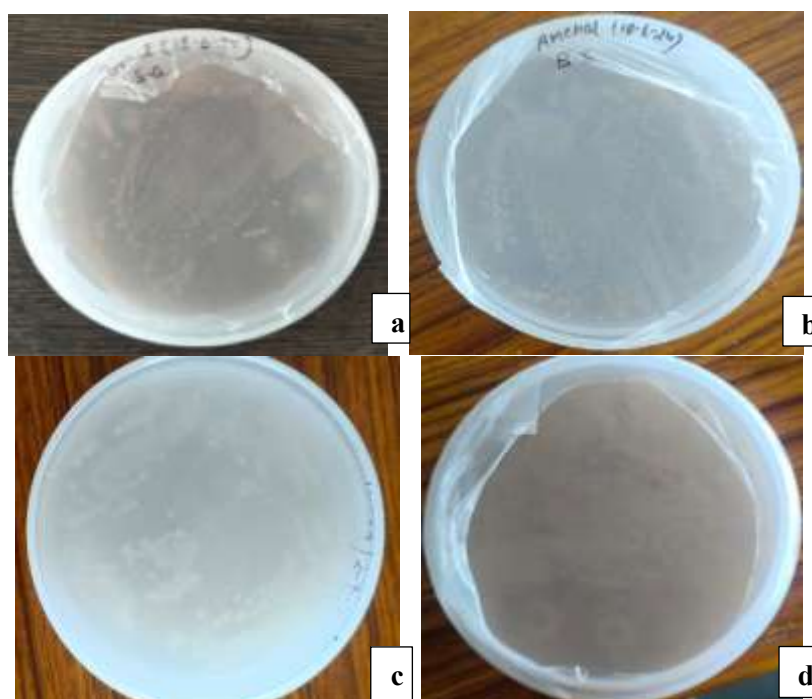
Protein is essential for the growth of microbes and it makes up a significant portion of microbial cells (Gabrielet al., 2012).

They also provide free sugars and a variety of essential minerals, making them a valuable component in nutrient media formulations for microbial cultivation. Fruit peel wastes are rich in carbohydrates, which include both simple sugars (like glucose, fructose, sucrose) and complex sugars (like starch, cellulose, hemicellulose, and pectin). Simple sugars are directly utilized by microorganisms as an immediate energy source for growth and metabolism. Complex sugars need to be broken down by microbial enzymes (e.g., cellulases, amylases, pectinases) into simpler sugars before they can be absorbed and metabolized (Jamal et al., 2013).

Papaya peel powder, has a nutrient source, and protein content of around 5.31%, contributing to the overall protein availability in the media. Proteins get broken down into amino acids and peptides, which microorganisms use to create their own enzymes, structural proteins, and metabolites. This boosts microbial growth and makes papaya peel powder a great nutrient supplement for media formulation (Jamal et al., 2013).

Orange peels, known for their high levels of natural flavonoids, are rich in phenolic compounds, which offer antioxidant properties and support microbial growth (Aburesha et al., 2018). Additionally, sugarcane bagasse contains about 18.4% protein, enhancing the protein content of the medium. Together, these components provide a balanced mix of proteins, fats, fibre, sugars, and bioactive compounds, creating a nutrient-rich environment ideal for microbial growth in the formulated media.

**Sample A:** Media prepared for a) *Staphylococcus aureus* b) *Bacillus subtilis* c) *Actinomycte* d) *Escherichia coli*. Media was prepared by mixture of all fruit peels (citrus, banana, papaya, and sugarcane bagasse) after 48-72 hrs growth was seen. In *Staphylococcus aureus* smooth and light golden-yellow-coloured colonies were formed. *Bacillus subtilis* were slightly raised, irregular edges, wrinkled and off-white to cream-colored. *Actinomycte* were quite powdery and whitish in colour. *Escherichia coli* Circular, smooth white or off-white. This media was proved best with highest rate of growth.



**Fig 1:** Growth of a) *Staphylococcus aureus* b) *Bacillus subtilis* c) *Actinomycte* d) *Escherichia coli* on media prepared by mixture of all fruit peels (citrus, banana, papaya, and sugarcane bagasse) after 48-72 hrs.

**Sample B:** Media prepared for a) *Staphylococcus aureus* b) *Bacillus subtilis* c) *Actinomycte* d) *Escherichia coli*. Media was prepared by mixture of sugarcane bagasse and orange peel after 48-72 hrs very minute growth of *Staphylococcus aureus* and *Bacillus subtilis* were found and there was no growth in *Actinomycte* and *Escherichia coli*. It might be because *Actinomycte* and *Escherichia coli* required highly nutrient media. They also required good amount nitrogen source to grow but fruit peels lack nitrogen source.



**Fig 2:** Microbial growth on media prepared by mixture of sugarcane bagasse and orange peel  
**Sample C:** Media prepared for a) *Staphylococcus aureus* b) *Bacillus subtilis* c) *Actinomycte* d) *Escherichia coli*. Media was prepared by mixture of sugarcane bagasse and papaya peel after 48-72 hrs very less growth was observed on *Bacillus subtilis*, *Staphylococcus aureus* and *Escherichia coli*. The observed effects may be due to limited nutrient contents of the peels (every bacteria needs specific nutrients), pH, temperature, over heating in autoclave, heat sometimes destroy the sensitive nutrients.



**Fig 3:** Microbial growth on media prepared by mixture of sugarcane bagasse and papaya peel  
**Sample D:** Media prepared for a) *Staphylococcus aureus* b) *Bacillus subtilis* c) *Actinomycte* d) *Escherichia coli*. Media was prepared by mixture of sugarcane bagasse and banana peel failed to show the growth of bacteria that might be due to of lack of nutrients present in the peels of banana like sucrose, carbon and nitrogen source. There was no any other supplement (e.g., peptone, yeast extract) present in the media for the growth of bacteria.



**Fig 4:** Microbial growth on media prepared by mixture of sugarcane bagasse and banana peel

**Table 2: Comparison between growth of microbes on different media**

S.NO.	Type media of	<i>Staphylococcus aureus</i>	<i>Bacillus subtilis</i>	<i>Escherichia coli</i>	<i>Actinomycete</i>
1	Media prepared by sugarcane bagasse, citrus peel, banana peel and papaya	+	+	+	+
2	Media prepared by mixture of sugarcane bagasse and orange peel	+/-	+/-	-	-
3	Media prepared by mixture of sugarcane bagasse and papaya peel	+/-	+/-	+/-	-
4	Media prepared by mixture of sugarcane bagasse and banana peel	-	-	-	-

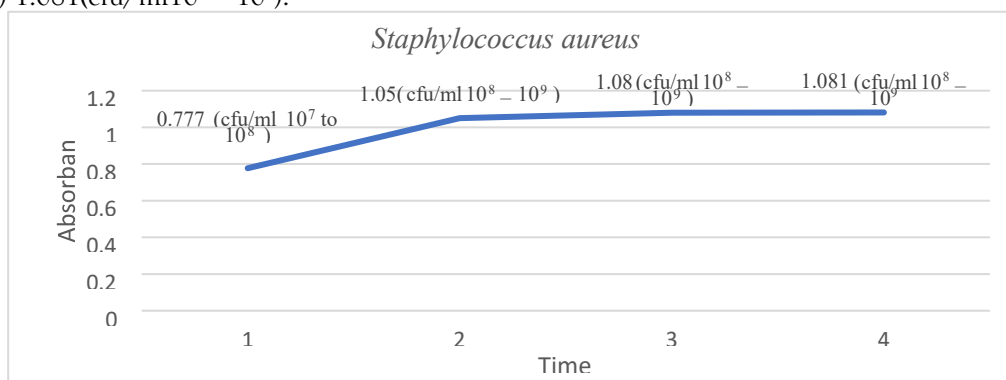
- (+) Indicates presence of bacteria
- (-) Indicates Absence of bacteria
- (+/-) Indicates minute presence of bacteria

### 3.2 Analysis of bacterial growth by Spectrophotometer:

Bacterial growth was checked at 37°C after the duration of 48 hrs. Time interval selected for growth curve was one hour. Fresh culture of test organisms was inoculated in Nutrient broth and formulated medium. Growth was measured at 600nm. The spectrophotometer quantifies optical density or absorbance at a designated wavelength—typically 600 nm (OD600) for bacterial cultures. OD 600 is utilized since bacterial cells scatter light at this wavelength, facilitating an indirect estimation of cell density. (Bachmann et al., 2020)

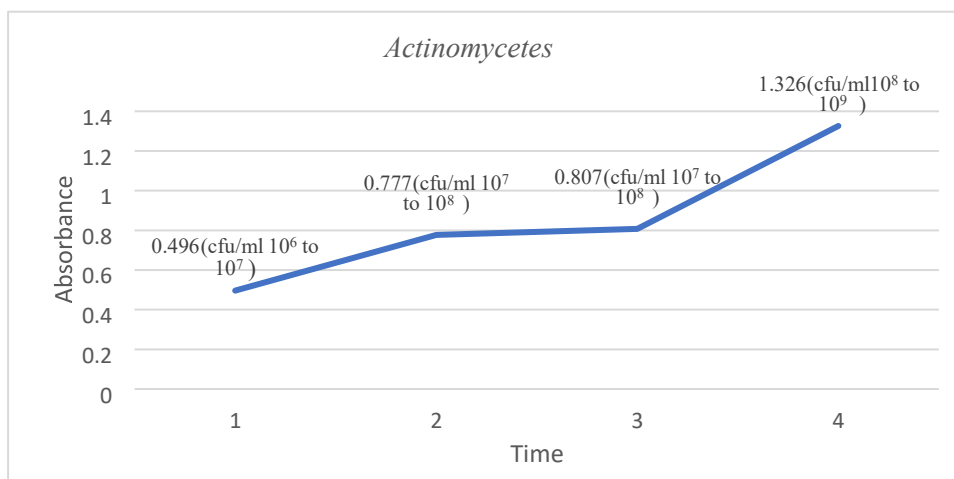
#### Microbial growth of different microbes:

Each bacteria growth rate was checked by using spectrophotometer at 600nm. In a) *Staphylococcus aureus* growth was observed at the intervals of one-hour, maximum growth was observed after four hours at (600nm)  $1.081(\text{cfu/ml } 10^8 - 10^9)$ .



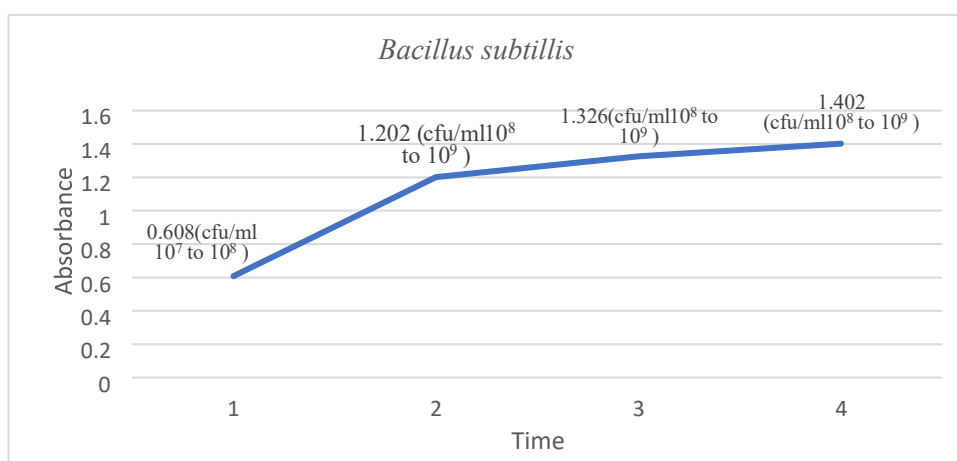
a) *Staphylococcus aureus* growth curve at different intervals of time at 600nm

*Actinomycetes* growth was observed four times at the intervals of one-hour and the readings were  $0.496(\text{cfu/ml } 10^8 - 10^9)$ ,  $0.777(\text{cfu/ml } 10^8 - 10^9)$ ,  $0.807(\text{cfu/ml } 10^8 - 10^9)$ , maximum growth was observed after four hour (at 600nm)  $1.326(\text{cfu/ml } 10^8 - 10^9)$



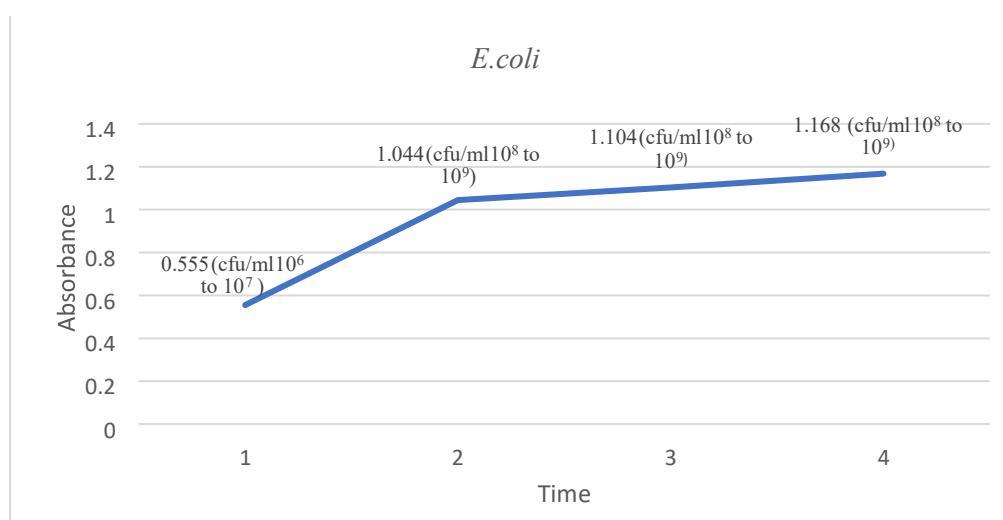
b) *Actinomyces* growth curve at different intervals of time at 600nm

The growth of *Bacillus subtilis* was also observed four times and curve started from 0.608(cfu/ml $10^8 - 10^9$ ) at first hour, second hour 1.202(cfu/ml $10^8 - 10^9$ ), third hour 1.326(cfu/ml $10^8 - 10^9$ ) and maximum growth was observed after four hour (at 600nm) 1.402(cfu/ml $10^8 - 10^9$ ).



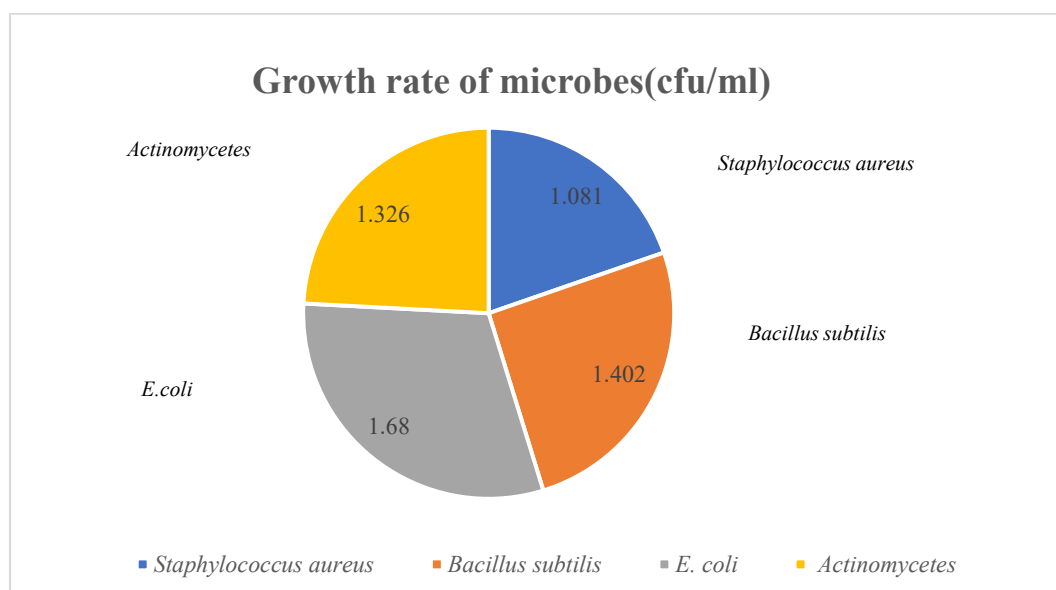
c) *Bacillus subtilis* growth curve at different intervals of time at 600nm

*E.coli* growth was observed four times at the intervals of one-hour and the readings were 0.555 (cfu/ml $10^8 - 10^9$ ), 1.044(cfu/ml $10^8 - 10^9$ ), 1.104(cfu/ml $10^8 - 10^9$ ) and maximum growth was observed after four hour (at 600nm) 1.168(cfu/ml $10^8 - 10^9$ )



d) *E. coli* growth curve at different intervals of time at 600nm





**e) Microbial highest growth rate:** This chart represents the growth rate of various microbes (in colony-forming units per millilitres, cfu/ml) where, 20% of the total, or 1.081 cfu/ml, were *Staphylococcus aureus*. 26%, or 1.402 cfu/ml, of *Bacillus subtilis*. *E. Coli*: 31%, the highest percentage, 1.68 cfu/ml. 24%, or 1.326 cfu/ml, where *Actinomyces*.

The cultivation of microorganisms in laboratory settings requires a nutrient-rich environment to support their growth and metabolic activity. Traditionally, synthetic media are used to provide these essential nutrients. The environmental and cultural factors are essential for cell growth and the synthesis of bioactive compounds. Factors such as initial pH, temperature, and others significantly influence the production of bioactive metabolites (Oskay et al., 2011).

To assess the impact of these nutrient alternatives, various mixtures of sugarcane bagasse and fruit peels were prepared, and their ability to support microbial growth was tested. In Figure 1, the media was formulated by combining sugarcane bagasse with citrus peel, banana peel, and papaya peel. In Figure 2, the media was prepared with a mixture of sugarcane bagasse and orange peel, while Figure 3, used a combination of sugarcane bagasse and papaya peel. Lastly, in Figure 4, sugarcane bagasse was mixed with banana peel to create the media. Among these four media formulations, the media prepared with the complete mixture of sugarcane bagasse, citrus peel, banana peel, and papaya peel (Figure 1) produced the most favourable results. This formulation demonstrated enhanced microbial growth compared to the other mixtures. The superior performance of the combined media can be attributed to the nutritional diversity provided by each peel type. Together, these peels are rich in essential nutrients such as natural sugars, fibres, organic acids, and vitamins, which are crucial for microbial growth.

The inclusion of multiple fruit peels in the media provided a broader range of nutrients, thereby creating a balanced and nutrient-dense environment that supported the growth of a wider variety of microbes. This diverse nutrient profile is likely to meet the varying dietary requirements of different microbial species, making the mixture of sugarcane bagasse with citrus, banana, and papaya peels a robust and versatile alternative to synthetic media. This study highlights the potential of using organic waste materials as effective, eco-friendly, and economical sources for nutrient media in microbial cultivation.

The growth of *Staphylococcus aureus*, *Bacillus subtilis*, *E. coli*, *Actinomyces* was successfully observed on media prepared by the mixture of sugarcane bagasse, citrus peel, banana peel and papaya peel collectively. The growth of microbes was observed within 48 - 72 hours at the temperature 30-32 °C. According to the existing literature it was observed that the cells of the majority of known bacterial species like *B. subtilis* exclusively establish their colonies in two dimension and do not penetrate the interior of solid substrates (Scherer et al., 1966). Whereas *Staphylococcus aureus* produces yellow pigment staphyloxanthin or cream - coloured and distinctive gold-coloured colonies in the growing area which is made up of several layers of densely packed cells (Kiefer et al., 2010). In the present study *Staphylococcus aureus* was observed having dense colonies with smooth, raised and round appearance this with off white, wrinkled spores like appearance because the nutrient media had different nutrients and components. *E. coli* has rough-type colonies with uneven edges, a sharp, cut-glass appearance, are flat, dry, and spreading (Krasilnikov et al., 1960). In the present study colonies of *E. coli* were observed with large circular dense growth and *Bacillus*

*subtilis* was found with off white, wrinkled spores like appearance and *Actinomyces* gives the culture a peculiar look.

Aerial hyphae create distinctive, thick, braided masses of mycelial threads on colonies' surfaces (McBirney et al., 2016). In present study *Actinomyces* colonies were white, filamentous and irregular in shape. The present study may differ in colour of colonies formed by microbes, shape due to the use of different type of nutrient material, environmental and temperature conditions.

The optical density (OD) of various microbial cultures was measured using a UV-Vis spectrophotometer to assess cell concentration and growth. Spectral measurements were taken across a range of wavelengths, with 600 nm being a common reference wavelength. For *E. coli*, an optimal wavelength of approximately 420 nm was identified for accurate quantitative measurements of cell density, as this wavelength allows for a precise assessment of bacterial concentration in the culture.

In the case of *Staphylococcus aureus*, wavelengths below 600 nm, particularly those showing significant changes in OD, were suitable for determining growth. For *Bacillus subtilis*, a range of wavelengths, including those below 600 nm as well as 600 nm itself, effectively measured cell density. *Actinomyces* cultures demonstrated optimal OD readings at wavelengths between 650 nm and 660 nm, indicating this range as ideal for monitoring its growth (McBirney et al., 2016). According to (Lang et al., 1994) the maximum growth observed in *Actinomyces* were  $0.0864 \text{ (cfu/ml)} 10^8 - 10^9$ .

In present study the results indicated that the nutrient media formulated with the combined fruit peels effectively supported the collective growth of the microbial cultures, showing measurable growth across various species. However, when different fruit peel powders were paired individually with sugarcane bagasse (in specific combinations such as sugarcane bagasse with orange peel, banana peel, or papaya peel), these media formulations did not universally support microbial growth. In these single-pair combinations, either no growth or only minimal growth was observed, suggesting that the nutrient requirements for all microbes were not sufficiently met with these pairings. Consequently, it was concluded that the complete mixture of fruit peels in the media provided a more balanced nutrient profile that supported a broader spectrum of microbial growth, while individual pairings with sugarcane bagasse alone did not achieve similar growth results.

#### 4. CONCLUSION

A significant portion of household organic waste, particularly vegetable and fruit peels, is often discarded as refuse. However, these peels contain valuable organic compounds and essential nutrients. In this study, it was found that fruit peel waste materials are rich in nutrients and minerals that can meet the dietary requirements of microbes important to various industries and laboratory settings. By providing a nutrient-rich substrate for microbial growth, fruit peels offer a sustainable resource for cultivating microbes that can be used in a range of applications, from industrial processes to scientific research.

Using fruit peel waste as a medium for microbial cultivation not only supports microbial growth but also promotes waste recycling and environmental conservation. This approach helps reduce reliance on synthetic chemicals and costly raw materials typically required for microbial media preparation, thereby minimizing the environmental impact of traditional waste disposal methods. Fruit peels, particularly when combined with sugarcane bagasse, can serve as an effective nutrient source in media formulation, representing a cost-effective and eco-friendly alternative to conventional microbial growth media.

This strategy is especially valuable in regions like India, where fruit peels are abundantly available and can be easily sourced from agricultural or domestic waste. The widespread availability of fruit peels in India makes them an ideal resource for creating nutrient media. This method also addresses the common issue of culture media shortages in laboratory settings by offering a readily available, sustainable solution. The results of this research have the potential to significantly alleviate the problem of media scarcity for laboratory practices, while also contributing to a circular economy model through the repurposing of organic waste.

In conclusion, media prepared from sugarcane bagasse and fruit peels not only provides a viable alternative for traditional microbial media but also presents a practical solution to environmental concerns associated with organic waste disposal. This research underscores the importance of integrating waste-derived materials into industrial and laboratory applications, highlighting the dual benefits of resource conservation and waste minimization.

## Authors' contribution

A.T.: Performed all the experiments, collected the data, and wrote the paper, Co-authors: P.S. and A.S.: Supervised the study and involved in planning of paper, experiments, selection and collection of raw material they were also involved in preparation of samples P.S.: Examined all the experiments, N.K.: supervised the research paper and gave required suggestions, A.S.: Worked on editing the manuscripts.

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