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(Original Research paper)

Sustainable bioplastic production from banana peel waste: Unveiling antibacterial potential and environmental impact

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Abstract: A variety of synthetic and semi-synthetic substances make up the polymer material known as plastic. Plastics are visually appealing, but their breakdown and disposal have created major problems. In order to address the contamination of ecosystem by plastics, it is necessary to create a "eco-friendly" solution. The goal of this research is to demonstrate that the starch in banana peels (*Musa acuminata*) can be utilized to create biodegradable plastic as an alternative to traditional plastic. The successful

incorporation of glycerol into the starch matrix was confirmed by structural analysis using Fourier Transform Infra-Red (FTIR) analysis, which also revealed hydroxyl, carbonyl, and C-H stretching vibrations. Scanning Electron Microscopy (SEM) showed a fibrous microstructure, which improved the bioplastic material's mechanical properties.The elongation test was used to compare the biodegradable film with a synthetic plastic and a control film in order to ascertain the film's strength.The degree of deterioration was assessed for each of the three types of film in the soil burial degradation test. The biodegradable film broke down more quickly. Banana peels' antibacterial properties were assessed for use in subsequent bioplastic-based product manufacturing. The findings show that bioplastic made from banana peels can solve significant industrial issues, food packaging, boosting productivity such as bags, toys, and water bottles.

1. Introduction

Plastics, or synthetic organic polymers, are an essential part of modern life, yet their widespread use and production didn't start until about 1950. Early synthetic plastics, Similar to Bakelite, plastics were first employed in the early 1900s, but their widespread use outside of the military didn't occur until the Second World War (Kumari *et al.*2023). Since then, plastics have been produced at a rate that has surpassed that of most other materials made by humans, with the exception of those that are widely used in building, such steel and cement. Since most plastics are not biodegradable, they are made primarily of fossil fuels leading to accumulation in landfills and the environment. This pervasive plastic waste is now so widespread that it is considered a marker of the proposed Anthropocene era (Geyer *et al.* 2017).

In response to the environmental impact of traditional plastics, biodegradable and biocompatible polymers are gaining importance. These biopolymers are used in various fields, including pharmacology, biomedicine, and environmental science. They possess unique properties such as high solubility in various solvents, low melt viscosity, and less entanglement in the solid state, which distinguish them from conventional plastics. Bioplastics, made from biomass like corn, banana peels, and sugarcane, can decompose naturally in aerobic (composting) or anaerobic (landfill) environments. Microbes in the soil break down these polymers, releasing monomers that have less harmful effects on the environment. These bioplastics can be derived from natural sources (e.g., PHA or PHB) or from petroleum-based plastics with added biodegradability enhancers (Jayachandra Yaradoddi *et al.* 2020).

Starch is a significant source for developing bioplastics. It is abundant, low-cost, renewable, and biodegradable. Starch can behave like a thermoplastic when combined with a plasticizer and heated or mechanically treated. But because films made from starch are frequently fragile and water-sensitive, additional natural biopolymers are incorporated to enhance their qualities (Noor Fatimah Kader Sultan & Wan Lutfi Wan Johari 2017). In addition to starch, plasticizers such formamide, glycerol, sorbitol, xylitol, and urea have been utilized extensively to create bioplastic films (Usha & Meera 2023).

Banana peels, which include cellulose, starch, and other biopolymers, are a viable source of bioplastics (Siluvai Kirubagari Aneeshia *et al*. 2022). There is approximately 220 tons of residue (mainly lignocellulosic material) produced per hectare of banana plantations. The peel, which makes up around 35–50% of a banana's mass, is usually thrown away untreated. Banana peels are generated in over 36 million tons annually, the majority of which are solid waste that produces unpleasant aromas as a result of anaerobic digestion. In the process of making goods like chips, grain flour, beverage, jelly,

and baby food, the food manufacturing sector also throws away a lot of banana peels (Mai Al-Dairi *et al*. 2023).

The problem of waste plastic can be solved sustainably by employing banana peels to generate bioplastics, which also makes use of a resource that would otherwise be discarded. In an effort to lessen the negative impacts of plastic waste and promote the development of renewable resources, this study investigates potential uses of bioplastics formed from banana peels. Plant is grown by using biodegradable pot and commercial plastic cover. Water holding capacity is more for biodegradable pot when compared to plastic cover it can hold water up to 1 day so water is need to be poured only one time a day no need to pour two times a day (Arjun *et al*.2023).

2. Materials and Methods:

2.1. Extraction of starch from *Musa acuminata*

Fresh *Musa acuminata* (banana) were gathered from farmers, the peels were removed and sliced into little pieces to prevent browning. They were then immersed for forty-five minutes in a 0.5% Na_2SO_4 solution. After that, the peels were cooked for 30 minutes in ethanol (1:2 w/v) to soften and extract the starch particles. The peels were boiled, then decanted and let to dry for half an hour to get rid of extra moisture. The dry peels were pureed into a homogeneous paste using a mortar and pestle (Pratiksha Kadam & Saswati Datta 2020)

2.2. Production of Bioplastics

A 500 ml beaker was filled with 25 ml of measured banana peel starch. A glass stirring stick was then used to mix the mixture after 3 milliliters of acetic acid had been added. The beaker was then filled with 2 milliliters of propan-1, 2, 3-triol (were tested at different glycerol content levels 2ml, 4ml, 6ml, and 8ml). Once more, the mixture was mixed. After being transferred to a petridish, the mixture was baked at 60°C. It was cooked for thirty minutes (Pratiksha Kadam & Saswati Datta 2020).

2.3. Biodegradation test for bioplastics

The 2.5 cm × 2.5 cm biodegradable film was cut. The film was then buried 5 cm below the surface. Water will be sprinkled on a regular basis. The soil samples were removed after a period of roughly two days and cleaned with distilled water. Following that, the specimens were dried and their weight was determined (Deeneshwaran *et al*. 2016). A statistical t-test is used to determine whether there is a statistically significant difference between the means of two groups (comparison of biodegradable bioplastics- Noorjahan *et al.* 2022).

2.4. Elongation test for bioplastics

The biodegradable plastic was sliced. The biodegradable plastic's original length was measured and noted. The biodegradable plastic was stretched beyond its original length. Another measurement of the plastic's length was made and noted. The formula for calculating elongation % is:

$$E(\%) = \left(\frac{L_f - L_i}{L_i}\right) \times 100 \qquad \text{.....(1)}$$

E(%) = Elongation percentage

L_f = Final weight of the bioplastic

L_i = Initial weight of the bioplastic

2.5. Water solubility test for bioplastics

The samples were weighed after being divided into 2.0 cm^2 squares. The samples were agitated for six hours at 25ºC at 30 rpm in 100 ml of distilled water. After six hours, the remaining samples were filtered. They were then dried at 110^0C in a hot air oven until a constant weight was achieved (Patkar *et al*. 2020). The following formula was used to get the solubility percentage:

$$S(\%) = \left(\frac{W_i - Wf}{Wf}\right) \times 100 \quad \text{.....(2)}$$

S(%) = Solubility percentage

W_i = Initial weight of the bioplastic

W_f = Final weight of the bioplastic

2.6. Bioplastics' morphology and chemical interactions

Fourier Transform Infrared Spectroscopy (Thermo Fisher Scientific, Nicolet iS5) was used to assess the intermolecular bonding of bioplastics in a 4000-400 cm^{-1} wave range. Purified samples were mixed with 10 mg of potassium bromide (KBr) powder. An average of 16 scans were taken in order to acquire IR spectra and integrate the spectrum. A scanning electron microscope (Carel Zeiss EVO 18) was used to examine the cross-sectional shape of the produced bioplastics. The samples were evaluated at an accelerating voltage of 20 Kv after being coated with copper grid and carbon wand prior to SEM examination (Usha & Meera 2023).

2.7. Antimicrobial activity of Bioplastics:

Bacillus subtilis, S*taphylococcus aureus, Pseudomonas aeruginosa* and *Klebsiella pneumoniae* were isolated from sewage samples respectively, identified and cultivated on nutrient agar medium. Antimicrobial activity test was performed based on Kirby -Bauer well diffusion method using Muller Hinton agar to determine the susceptibility or resistance of bacterial cultures to antimicrobial compounds/extract by measuring zone of inhibition.

A loop full of bacterial culture was inoculated in the nutrient broth to create the liquid culture, which was then incubated at 37 °C for additional steps. The colonies were transferred to the Muller Hinton Agar using a loop. After that, the sterile cotton swab was dipped into the inoculum and used to swab the agar plate's surface. With cork borer a 5 mm diameter was used to create wells on each plate, and the ethanolic banana extract was then loaded to each well. The plates were then placed in an incubator set at 37°C for 18 to 24 hours.Wells were created, one for the sample (an ethanolic extract of banana peel using a sterile micropipette at varied extract quantities 20, 40, 60, 80, and 100µl), one for the positive control (an antibiotic-Streptomycin), and one for the negative control (DMSO). Following

the addition of the ethanolic extract to the wells, the plates were incubated at 37°C. After the incubation time, zones of inhibition will be measured around the wells, showing antimicrobial efficacy.

2.9.Statistical analysis:

The triplicate data obtained from biodegradability test were statistically analysed and expressed in terms of Mean, Standard Deviation and 't' Test (Noorjahan *et al.* 2022). ANOVA (Analysis of Variance) is used to compare the means of multiple groups to determine if there is a statistically significant difference between them for antimicrobial activity test.

3. Result & Discussion:

The bioplastic film that were fabricated shown in Fig. 1. We found that bioplastic samples made from banana peel starch that had higher glycerol concentrations (8 ml) broke down much more quickly in 10 days graphically represented in Fig. 2, Table. 1 explains biodegradability test of synthesized bioplastics for a period of 15 days in the soil burial shows drastic change in weight (g) and In a biodegradability test, a t-test assess bioplastics degrades significantly differently under different concentrations.

. Glycerol serves as a plasticizer, enhancing the flexibility and permeability of the bioplastic matrix, which facilitates microbial colonization and enzymatic degradation (Pratiksha Kadam & Saswati Datta 2020). Glycerol has a hygroscopic nature. Its molecular structure has three hydroxyl (OH) groups, which is the cause of this. Hydrogen bonds can be formed between glycerol and water molecules. Consequently, the glycerol content was adjusted to be 2 ml, 4 ml, 6 ml, and 8 ml in order to improve the bioplastic's solubility, moisture content, and water absorption (Deepali Elizabeth *et al*. 2024).

All the soil buried bioplastic samples were taken from the soil at various time intervals. The rate of biodegradation was observed and compared with the synthetic polypropylene. Significant degradation was observed on day 10 in all the bioplastic films. Degradation at 30 % was observed in B1, 28 % in B2, 26 % in film B3, 27 % in commercial bioplastic (CB). Degradation of all the bioplastic films in soil was almost 80% in 30 days which shows a remarkable degradation period compared to the reports of Carlos *et al*. 2016.

Fig.1: Extraction of starch and production of thin, flexible bioplastic film

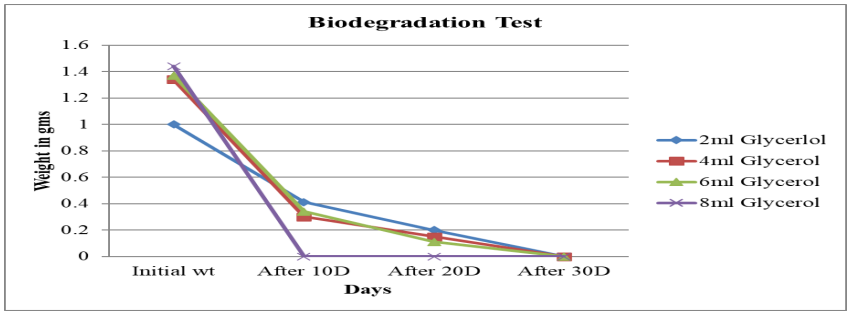

Fig.2: Graphical comparison of biodegraded bioplastic weight loss over time

Table. 1: Biodegradability test of synthesized bioplastics for a period of days in the soil.

S.No	Days	Appearance	Weight (g) of the sample				Mean ± S.D.	't' test
1	5th day	Brown	1.00	1.34	1.37	1.44	1.280 ±0.1699	5%
2	10th day	Brownish black	0.81	0.82	0.93	0.94	0.875 ±0.0602	
3	15th day	Black	0.731	0.735	0.734	0.734	0.734 ±0.0015	

The elongation or stretchability test for banana peel bioplastic starch-based using glycerol plasticizer indicates as the highest elongation percentage, 36.66%, was achieved with 8ml of glycerol, suggesting an optimal concentration for maximum stretchability as depicted in Fig. 3.

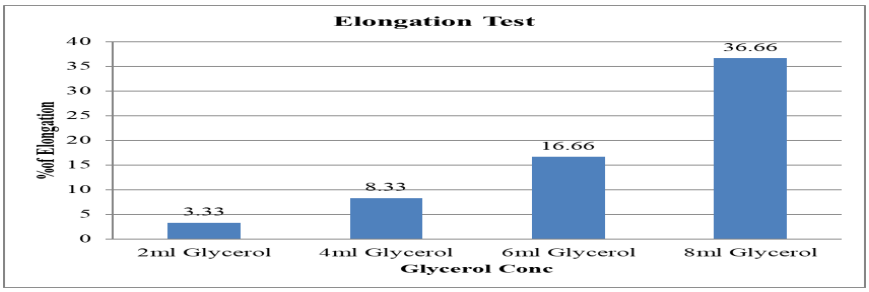

Fig:3. Graphical representation of elongation test

The presence of hydrophilic chemicals as the main element was shown by the bioplastic films' solubility. Due to their ability to absorb water, bioplastic sheets exhibited a 70% greater solubility. Since crosslinking and high levels of intermolecular interaction within the matrix result in a reduced ability to interact with water, synthetic polypropylene plastic was only 2% soluble (Usha & Meera 2023). Thus, bioplastic sheets were significantly soluble in water, whereas polypropylene plastic was the least soluble. Using cassava flour and different glycerol proportions ranging from 1 to 3%, (Wahyuningtiyas & Suryanto 2017) created bioplastic and assessed its biodegradation.

The produced bioplastic films were analyzed using FT-IR analysis to determine the functional groups, spectra ranging from 500 to 4000 cm^{-1} were ascribed to the hydrogen bonds produced by the starch and plasticizer's O-H group interactions. The absorption band at 3289.96 cm^{-1} confirms the presence of hydrogen bonding, including hydrate (H_2O) and hydroxyl (-OH) groups. Sharp peaks at 2936.09 cm^{-1} indicate stretching of CH_2 groups in starch structure. The peak at 1636.3 cm^{-1} indicates

unsaturation bonds, specifically C=C double bonds. The 1415.49 cm^{-1} peak corresponds to O-H bending, showing a hydrogen link between starch and bioplastic as shown in Fig. 4. The 1032.69 cm^{-1} band represents C-O bending grew as glycerol was added, and the enhancement of the -OH stretching vibration at about 3269 cm-1 indicated the occurrence of association (Jiang *et al*. 2020).

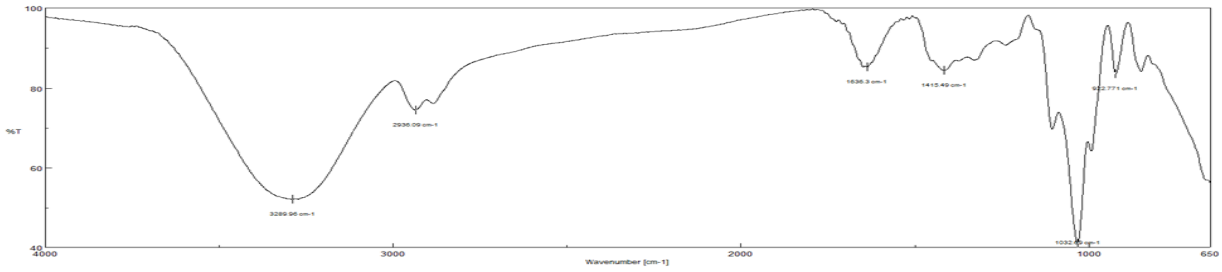

Fig. 4: FTIR analysis of Bioplasic films

SEM was used to examine the surface morphology of bioplastic sheets. Due to their brittle nature, the produced bioplastic sheets showed few surface fissures as observed in Fig. 5. Although there are less gaps and microcrystals in the SEM image, the plasticizer was well integrated. When hydrogen bonds in lengthy chains of starch break down at gelatinization temperatures, water molecules infiltrate the hydroxyl groups of the starch molecules, resulting in voids and microvoids (Anitha *et al.* 2024).

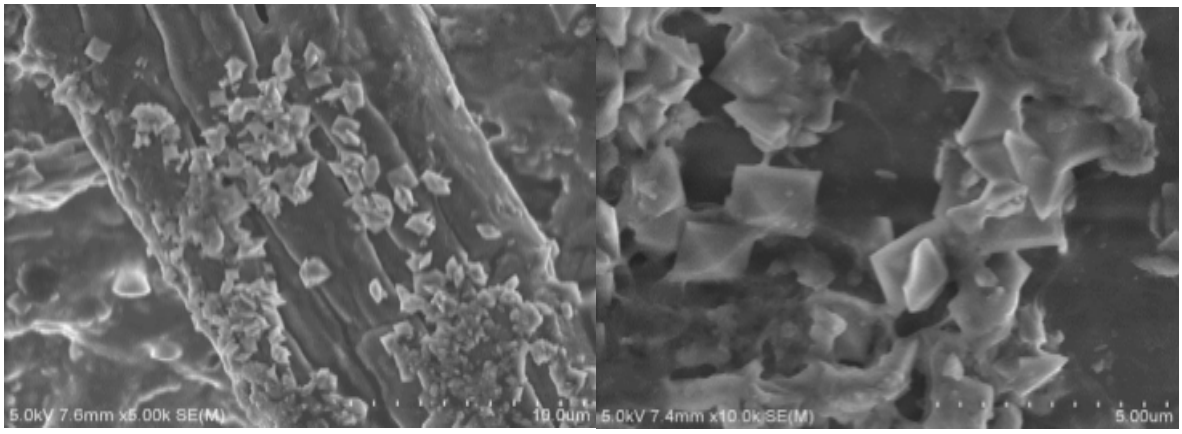

Fig. 5: The SEM micrograph of banana peels bioplastics

The antimicrobial activity of banana peel bioplastic extract against *Klebsiella pneumoniae, Pseudomonas aeruginosa, Staphylococcus aureus*, and *Bacillus subtilis*, using varying extract volumes (20µl, 40µl, 60µl, 80µl, 100µl), shows consistent trends: increasing extract volume correlates with larger zones of inhibition for all bacterial strains. *Staphylococcus aureus* is most sensitive, with the largest inhibition zone (up to 23mm), while *Klebsiella pneumoniae* is less sensitive as shown in Fig. 6. Differential sensitivity among bacterial strains underscores the need to understand interactions between bioplastic extract compounds and specific bacteria.These findings illustrated the advantages of glycerol as plasticizer (Deepali *et al.* 2024). Table. 2 indicates ANOVA test (e.g., in Excel using Data Analysis Toolpak) for antimicrobial activity, this test compared means of multiple group and determined P-value ≤ 0.05 supports rejecting the null hypothesis (Significant difference exists) and higher F-value means greater variation between groups compared to within groups. Different treatments have significantly different effects on bacterial inhibition zones.Using banana peels, biodegradable plastic was created. The weight decreased from 72.05 g to 44.70 g in the first 6 days of the biodegradation test, and in the next 60 days, 100% decomposition is anticipated (Arjun et al., 2023).It was discovered that as starch and glycerol concentrations rose, antibacterial activity fell. Zinc oxide added to bioplastic showed a

stronger antibacterial activity. 5 ml of glycerol with 5% zinc oxide were found to have a high zone of clearance (Jerlin Vinoth *et al*. 2021).

Banana peel bioplastics interact with microbial colonies through surface adhesion, enzymatic biodegradation, and antimicrobial effects. Optimizing their composition and processing can enhance either their biodegradability or antimicrobial properties for specific applications. Banana peels contain phenolic compounds, flavonoids, and tannins, which exhibit antimicrobial properties against bacteria like *Staphylococcus aureus*, and *Pseudomonas aeruginosa*. This antimicrobial effect can inhibit biofilm formation or reduce microbial colonization. The extent of antimicrobial activity depends on factors like plasticization, extraction method, and aging of the bioplastic.

To protect the food from spoilage due to microorganisms, antimicrobial packaging is one of the most promising active packaging systems. Environmental concerns associated with plastic waste emphasized the development of packaging film from natural polymers such as starch.Bioplastics has been named as the eyes of biomaterial because it is highly applicable in skins replacements for burnings and wounds, scaffolds for tissue engineering, bone reconstruction, nerves and gum reconstruction, drugs releasing system, blood vessel growth and stent covering. Besides, in dental industry bioplastics based nanocellulose has been used in dental tissue regeneration in humans which is produced from microbial cellulose by the Glucanacetobacter xylinus strain. Bioplastics have been the great of interesting exploration such as in construction and building industry. On the other hand, bioplastics also is applied in membrane for reinforcement for high quality electronic paper (e-paper), combustible cells (hydrogen) and as an ultra-filtration membrane for water treatment (Veena and Rani 2022).

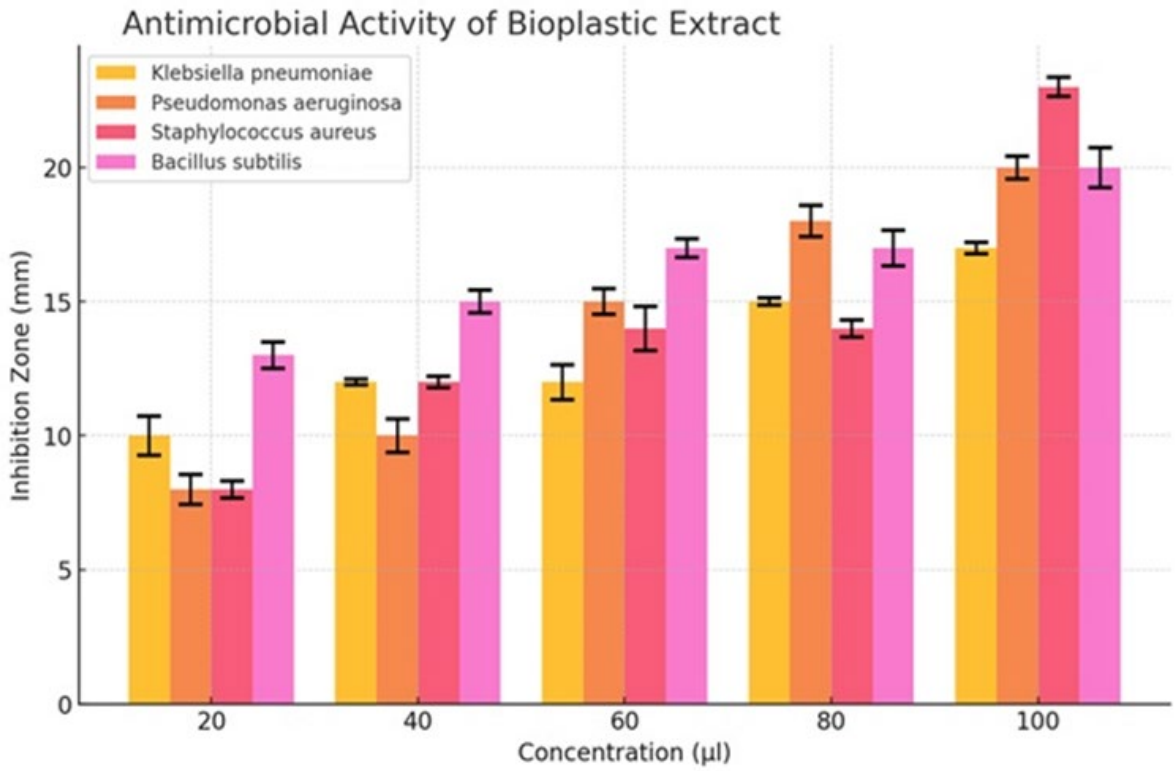

Fig. 6: Graphical representation of antibacterial activity of bioplastics.

Table. 2: ANOVA (Single Factor) analysis for antibacterial activity of bioplastics.

S.No.	Microorganisms used	F value	P value	F Crit
1	*Klebsiella pneumoniae*	10.86752009	0.010914174	5.317655072
2	*Pseudomonas aeruginosa*	10.22042487	0.012670490	5.317655072
3	*Staphylococcus aureus*	10.1807416	0.012789393	5.317655072
4	*Bacillus subtilis*	9.440603894	0.015288506	5.317655072

4. Conclusions

We conclude that plastic pollution can negatively impact lands, waterways, and oceans after taking into account all of the aforementioned points. Plastic pollution also affects humans, for example, by interfering with hormone levels or the thyroid hormone axis. As a result, biodegradable plastic emerges as a viable remedy for all of these issues. The biodegradable plastic made from banana peels is intended to serve as a replacement for traditional plastic and demonstrate that the starch found in banana peels can be utilized to make biodegradable plastic. This indicates that bioplastics made from banana peels may be a practical substitute for artificial antimicrobials in the treatment of some bacterial illnesses. In summary, the films made from banana peels have the potential to be used as food packaging since they can both improve food quality and safeguard the environment.

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