

BIODEGRADABLE SILK

YC WORLD COSMETICS

DONGGUAN YACAI COSMETICS CO.,LTD.

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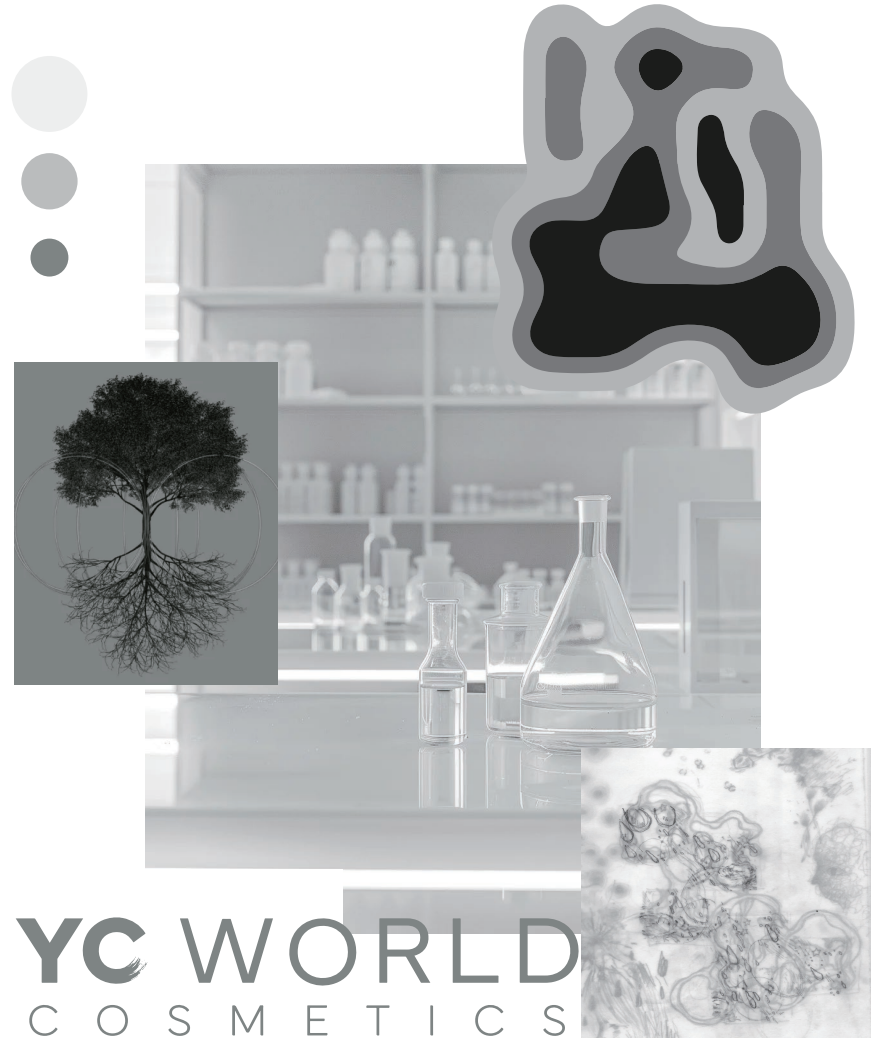
Biodegradable



Environmentally Friendly

PROJECT DESCRIPTION

Biodegradable PBT fibers have been submitted for the ASTM D5511 test. This method measures the degree and rate of anaerobic biodegradation of plastics in high - solid anaerobic conditions. The test material is exposed to methanogenic inoculum from an anaerobic digester for household waste pretreatment. Anaerobic decomposition occurs under high - solid (total solid content > 30%) and static, non - mixed conditions. The method aims to determine the percentage of sample carbon converted to gaseous carbon during municipal solid waste treatment in a high - solid - content anaerobic digester.



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1. INOCULUM COLLECTION AND CONDITIONING

The anaerobically digested sewage sludge (see figure) is mixed with domestic waste. To acclimatize and stabilize it during the short 53 °C post-fermentation, the sludge is pre-cultured at 53 °C for a week. The concentrated inoculum isn't fed; instead, pre - added organic residues degrade during post - fermentation, lowering its biogas background level.

2. Characteristics of the inoculum

The samples of anaerobically digested sewage sludge were analyzed for pH, percentage of dry solids, and volatile solids, as well as the amounts of CO₂ and CH₄ produced during the testing process. Table 1 lists the results of this preliminary test.



Standard Test Method for Determining the Anaerobic Biodegradability of Plastic Materials under Anaerobic Digestion Conditions with High Solid Content (ASTM D5511)

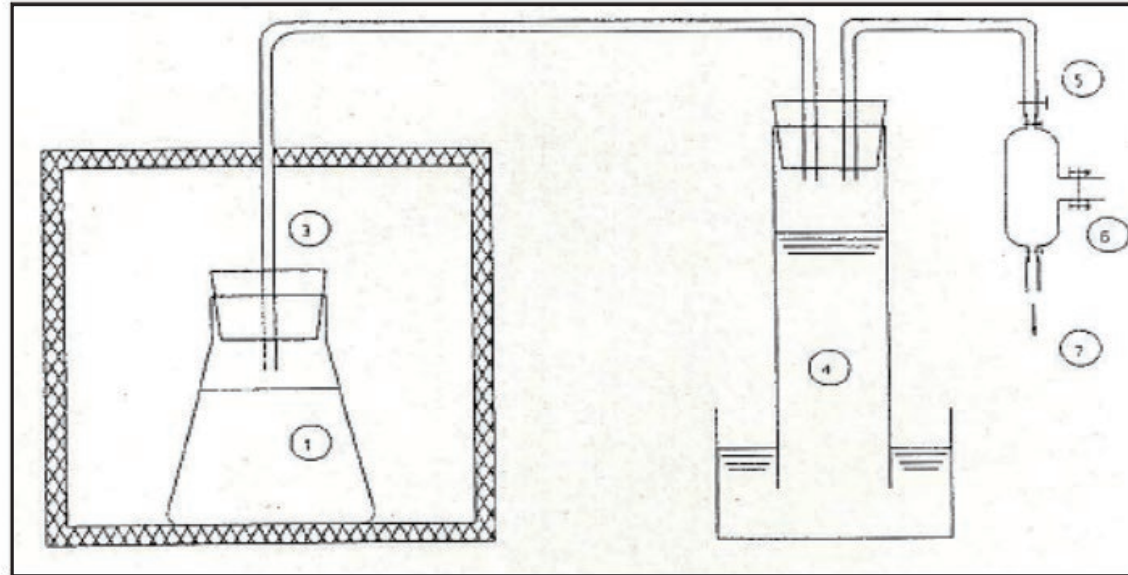
<div>1</div> <div>PREPARATION OF THE INOCULUM MEDIUM</div>	<p>Source: Take out approximately 15 kilograms of inoculum from the container after fermentation.</p> <p>Treatment: Stir it carefully and evenly by hand to ensure the uniformity of the culture medium.</p>
<div>2</div> <div>TEST SETUP</div>	<p>Blank sample: Contains only the inoculum, repeated three times.</p> <p>Positive control: Thin-layer chromatography cellulose, repeated three times.</p> <p>Negative control (optional): Set up as required.</p> <p>Test substance: The sample to be evaluated, repeated three times.</p>
<div>3</div> <div>SAMPLE MIXING</div>	<p>Mixing ratio: 1000 grams of inoculum with wet weight (containing at least 20% dry solids) is mixed with the volatile solids of 15 to 100 grams of the test substance or the control substance.</p> <p>Mixing time: Manually mix for 2 to 3 minutes.</p> <p>Container: Use a 2-liter wide-mouth conical flask. Ensure that the materials are evenly spread out and compacted to make the density inside the flask consistent.</p>
<div>4</div> <div>CULTIVATION CONDITIONS</div>	<p>Temperature: 52°C (± 2°C), under thermophilic conditions.</p> <p>Lighting: In a dark or diffused light environment.</p> <p>Cultivation time: Continue until there is no net gas production for at least five consecutive days in the reactors of both the positive control and the test substance.</p>
<div>5</div> <div>GAS COLLECTION AND MEASUREMENT</div>	<p>Gas collection device: Connect the conical flask to the gas collection device.</p> <p>pH control: Control the pH value of the water for measuring the biogas production to below 2 by adding hydrochloric acid.</p>
<div>6</div> <div>JUDGMENT OF THE RESULTS</div>	<p>Termination condition: There is no net gas production for five consecutive days in both the reactors of the positive control and the test substance.</p> <p>Data processing: Record the biogas production and analyze the biodegradability.</p> <p>Remarks: Ensure that all operation steps are consistent to guarantee the accuracy and reproducibility of the experimental results.</p>

ASTM D5511 - Standard Test Method for Determining the Anaerobic Biodegradability of Plastic Materials under Anaerobic Digestion Conditions with High Solid Content

1	Overview of the Test Method	Standard: ASTM D5511 Purpose: To determine the biodegradability of plastic materials under anaerobic digestion conditions with high solid content. Application: To evaluate the degradation performance of plastics in an anaerobic environment.
2	Preparation of the inoculum	Source: Take out approximately 15 kilograms of inoculum from the container after fermentation. Treatment: Stir it evenly by hand to ensure the uniformity of the culture medium.
3	Test Setup	Blank sample: Contains only the inoculum, repeated 3 times. Positive control: Thin-layer chromatography cellulose, repeated 3 times. Negative control: Optional. Test substance: The sample to be evaluated, repeated 3 times.
4	Sample Mixing and Loading	Mixing ratio: 1000 grams of inoculum with wet weight + 15-100 grams of the test substance or the control substance Mixing time: 2-3 minutes Container: A 2-liter wide-mouth conical flask. Spread the mixture evenly and compact it.
5	Cultivation Conditions	Temperature: 52°C (± 2°C), under thermophilic conditions Lighting: In the dark or under diffused light Cultivation time: Continue until there is no net gas production in both the positive control and the test substance for 5 consecutive days.
6	Gas Collection and Measurement	Gas collection device: Connect the conical flask to the gas collection equipment. pH control: Add hydrochloric acid to make the pH value of the measuring water lower than 2.

Remarks: Ensure that the operations are consistent to guarantee the accuracy and reproducibility of the experimental results.

The biodegradation testing of sample was performed in the digester as shown in the (Figure-3).



- 1.Digester
- 2.Incubator
- 3.Gas outlet
- 4.Gas collector
- 5.Valve
- 6.Gas Sampling
- 7.Gas Discharge

Figure-3: Digester setup

The most important biochemical characteristics of the inoculum such as pH, Volatile Fatty Acids, NH₄⁺-N— and dry solids was studied.

PARAMETERS	REQUIREMENT	ACTUAL RESULTS
PH	7.5-8.5	7.58
KJELDAHL NITROGEN	0.5 to 2 g/kg wet weight	1.40
DRY SOLIDS AT 105°C	>20%	44.00
VOLATILE SOLIDS AT 550°C	Below 1 g/kg wet weight	0.72

Table 1: Results of Initial testing of the anaerobic digested sewage sludge

The volume of biogas in the sampling bag was measured (Table 2). Gas in the positive control's (reference material) collector showed the inoculum was viable, and gas displacement occurred in both the positive control and test sample. ASTM D5511 requires the positive control to reach 70% within 30 days with replicate mean deviation < 20% for a valid test. The positive control reached 71.47% on day 27 with < 20% mean difference between replicates. Gas displacement after 45 days is presented in the table below.

BIODEGRADATION TEST	TOTAL VOLUME 45 DAYS (ML)
INOCULUM	2530
POSITIVE CONTROL (REFERENCE MATERIAL)	9650
BIODEGRADABLE PBT FIBER	4500

Table-2: Biogas volume of the evolved gas during the biodegradation process at 45 days



(A)



(B)



(C)



(D)

Colonization of bacteria at some places was observed under the microscope (Fig-4). This shows the process of biodegradation has begun.

Figure 4: Microscopic images of the test sample before and after 45 days of incubation.

A & B: Unexposed biodegradable PBT fiber to anaerobic biodegradation.

C & D: Exposed biodegradable PBT fiber to anaerobic biodegradation.

The percent biodegradation of the positive control (reference material) and test sample was calculated from the measured cumulative carbon dioxide and methane production in each flask. This was done by subtracting the carbon dioxide and methane evolution from the blank sample at the end of 45 days of testing. Calculations were based on the total organic carbon of both the positive control and test sample.

Table-3: Percentage biodegradability of Test sample with respect to Positive control (Reference material) Cellulose.

GROUP	Inoculum control	Positive control (Reference material)	BIODEGRADABLE PBT FIBER Sample
WEIGHT	1000ml	10.2300g	15.0493g
TOTAL VOLUME (ML)	2580.00	9650.00	4500.00
% CH ₄	12.70	43.20	19.20
VOLUME OF CH ₄ (ML)	327.66	4168.80	864.00
WEIGHT OF CH ₄ (G)	0.2149	2.7347	0.5668
% CO ₂	14.90	44.70	21.10
VOLUME OF CO ₂ (ML)	384.42	4313.55	949.50
WEIGHT OF CO ₂ (G)	0.7612	8.5408	1.8800
TOTAL WEIGHT OF CARBON IN GRAMS	0.3667	4.3571	0.9327
THEORETICAL WEIGHT OF CARBON IN GRAMS (CI)	-	4.3017	8.0318
BIODEGRADATION	-	0.9276	0.0705
% BIODEGRADATION	-	92.76	7.05

Table-3

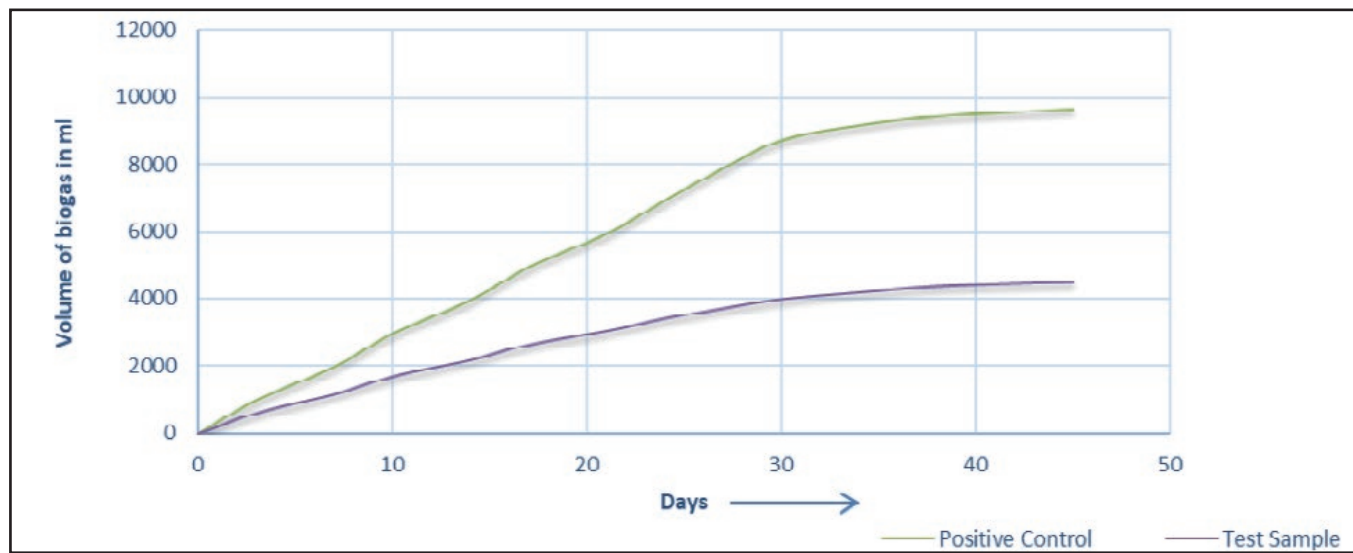
Table 4: Percent weight loss of BIODEGRADABLE PBT FIBER sample.

Average Initial Weight (grams)	15.0493
Average Final Weight (grams)	14.8933
Percent Weight Loss (%)	1.04

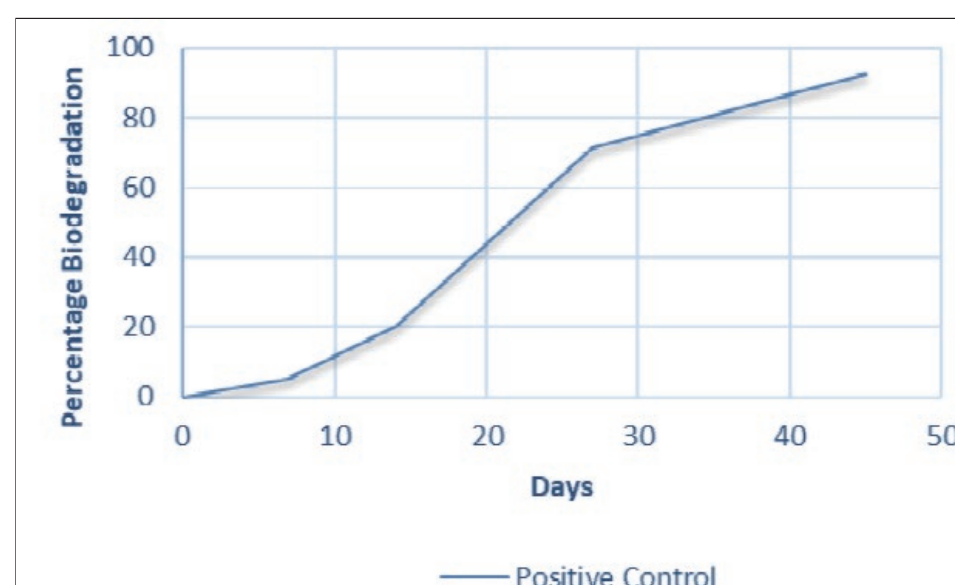
Table-4

The Percent weight loss was calculated based on the initial weight and final weight of the test sample after the 45 days study

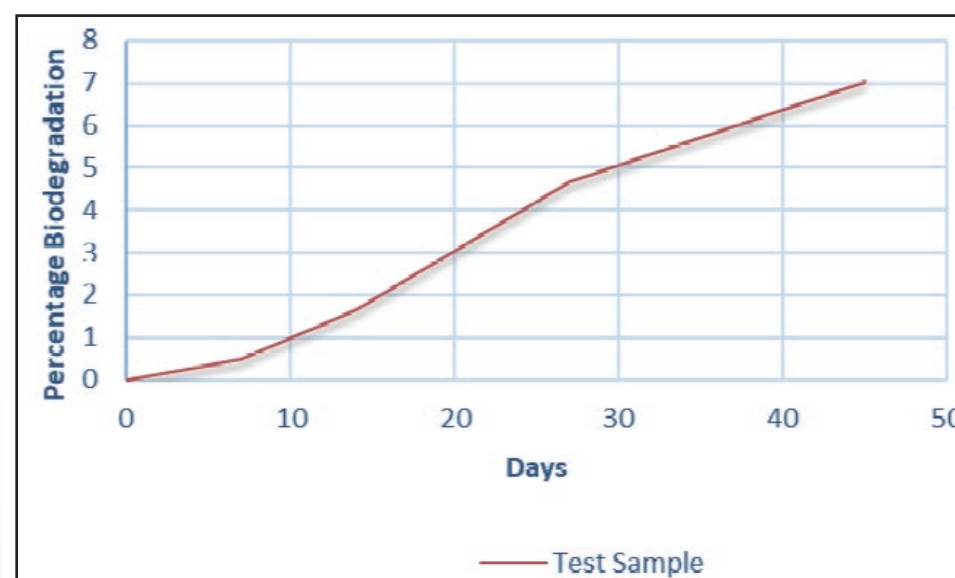
Biodegradation of the sample determined based on conversion of carbon from the test material to carbon in the gaseous phase (CH_4 and CO_2) can be observed in graph 1 and graph 2a & 2b.



Graph-1: Plot showing Net Biogas Production from Test sample (BIODEGRADABLE PBT FIBER) and Positive control (Reference material- Cellulose)



Graph-2a: The percent biodegradation of the Positive control (Reference material- Cellulose) determined based on conversion of carbon from cellulose to carbon in the gaseous phase (CH_4 and CO_2)



Graph-2b: The percent biodegradation of the Test sample (BIODEGRADABLE PBT FIBER Sample) determined based on conversion of carbon from the Test material to carbon in the gaseous phase (CH_4 and CO_2)

CONCLUSION:

Considering the cumulative gas production as observed in Table 2 & 3 and its analysis indicates that the process of biodegradation has occurred in BIODEGRADABLE PBT FIBER Sample. After 45 days of incubation, the level of biodegradation for the Positive control (Reference material) was 92.76 % while the BIODEGRADABLE PBT FIBER Sample submitted showed 7.05 %.

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