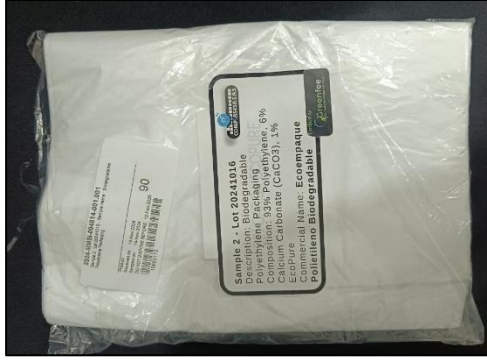


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Sample Registration Date	14/11/2024
Analysis Starting Date	14/11/2024
Analysis Completed on	22/01/2025
Sampling Done by	Sample drawn and supplied by customer
Deviation from the test methods:	None
Packaging Condition/sealed/unsealed	Sealed with box
Sample Image	 <p>Figure 1: Test Sample</p>

DETAILS PROVIDED BY THE CUSTOMER

Customer Name:	COMPLASESA SAS
Contact Person:	Mr. David Enriquez Ordonez
Address:	KM 25 Via Siberia Funza VD Cacique LA Playa BG 7
Sample submitted as	Sample 2: Lot 20241016
Description on label/Sample Bottle	Biodegradable Polyethylene Packaging
Sample type	93 % polyethylene, 6 % Calcium Carbonate (CaCO ₃), 1 % EcoPure
Quantity	1 Packet
Mode of Packaging	Courier Box
Sample condition	Sample was at ambient temperature in good condition
Test Method	ASTM D5511 Standard Test Method for Determining Anaerobic Biodegradation of Plastic Materials Under High-Solids Anaerobic-Digestion Conditions

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LABORATORY

Testing as presented in this report was conducted by Environmental division of Intertek India Private Limited. The testing facility is located at F wing, 2nd Floor, Chandivali Saki vihar Road, Andheri (East), Mumbai – 400 072, India.

SAMPLE RECEIPT

The Biodegradable Polyethylene Packaging samples were received on 14/11/2024 at the Intertek testing facility. The sample was sent through courier. The sample was at ambient temperature in good condition with no evidence of damage or contamination. No temperature preservation was required.

PROJECT DESCRIPTION

Biodegradable Polyethylene Packaging Sample was submitted by COMPLASESA SAS for testing under standard ASTM D5511. This test method covers the determination of the degree and rate of anaerobic biodegradation of plastic materials in high-solids anaerobic conditions. The test materials are exposed to a methanogenic inoculum derived from anaerobic digesters operating only on pretreated household waste. The anaerobic decomposition takes place under high-solids (more than 30 % total solids) and static non-mixed conditions. This test method is designed to yield a percentage of conversion of carbon in the sample to carbon in the gaseous form under conditions found in high-solids anaerobic digesters, treating municipal solid waste.

INOCULUM COLLECTION AND CONDITIONING

The anaerobic digested sewage sludge (Figure 2) mixed with household waste was obtained from the Chembur (Mumbai). To make the sludge adapted and stabilized during a short post-fermentation at 53°C, the sludge was pre-incubated (one week) at 53°C. This means that the concentrated inoculum was not fed but allowed to post ferment the remains of previously added organics allowing large easily biodegradable particles were degraded during this period and reduce the background level of biogas from the inoculums itself.

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Figure 2: Anaerobic microbial inoculum

INOCULUM PROPERTIES

A sample of the anaerobic digested sewage sludge was analyzed for pH, percent dry solids, and volatile solids, as well as, the amount of CO₂ and CH₄ evolution during the testing. Table 1 lists the results of this initial testing.

METHODOLOGY:

Inoculum Medium: Remove enough inoculum (approximately 15 kg) from the post-fermentation vessel and mix carefully and consistently by hand in order to obtain a homogeneous medium. Test three replicates each of a blank (inoculum only), Positive control (Reference material) (thin-layer chromatography cellulose), negative control (optional), and the test substance being evaluated.

Manually mix 1000 g wet weight (at least 20 % dry solids) of inoculum in a small container for a period of 2 to 3 min with 15 to 100 g of volatile solids of the test substance or the controls for each replicate. For the three blanks containing inoculum only, manually mix 1000 g of the same inoculum in a small container for a period of 2 to 3 min with the same intensity as was done for the other vessels containing test substance or controls. Determine the weight of the inoculum and test substance added to each individual Erlenmeyer flask accurately. Add the mixtures to a 2-L wide-mouth Erlenmeyer flask and gently spread and compact the material evenly in the flask to a uniform density.

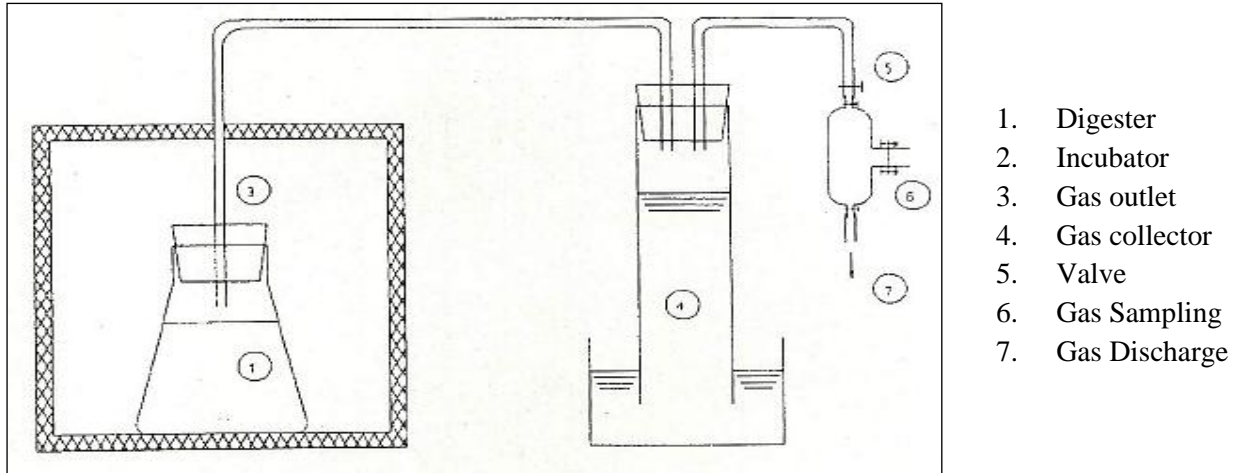
After placing the Erlenmeyer flask in incubator, connect it with the gas collection device. Incubate the Erlenmeyer flasks in the dark or in diffused light at 52°C (± 2°C) for thermophilic conditions, The incubation time shall be run until no net gas production is noted for at least five days from both the Positive control (Reference material) and test substance reactors. Control the pH of the water used to measure biogas production to less than two by adding HCl.

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ANAEROBIC DIGESTER SETUP FOR THE PLASTIC BIODEGRADATION

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



RESULT:

The most important biochemical characteristics of the inoculum such as pH, Volatile Fatty Acids, NH_4^{++}N — and dry solids were studied.

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

Parameters	Requirement	Actual results
pH	7.5 to 8.5	7.85
Kjeldahl nitrogen	0.5 to 2 g/kg wet weight	1.26
Dry Solids at 105 °C	>20%	45.99
Volatile Solids at 550 °C	Below 1 g/kg wet weight	0.71

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The biogas volume in the gas sampling bag was measured (Table- 2). Presence of gas in the gas collector of Positive control (Reference material) indicated that the inoculum was viable and gas displacement was observed both in Positive control (Reference material) and Test Sample.

ASTM D 5511 states that for the test to be considered valid, the Positive control (Reference material) must achieve 70 % within 30 days with deviation less than 20% of the mean between the replicates.

Positive control (Reference material) showed 70.90% on 27th day with less than 20% of the mean difference between the replicates.

The gas displacement observed after 45 days is as shown in the table below.

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

Biodegradation Test	Total Volume 45 days (mL)
Inoculum	2470
Positive control (Reference material)	9840
Biodegradable Polyethylene Packaging	4450

The percent biodegradation of Positive control (Reference material) and Test sample was calculated by the measured cumulative carbon dioxide and methane production from each flask after subtracting carbon dioxide evolution and methane evolution from the blank samples at the end of 45 days of testing. Calculations were based on Total Organic Carbon obtained of both Positive control (Reference material) and Test sample.

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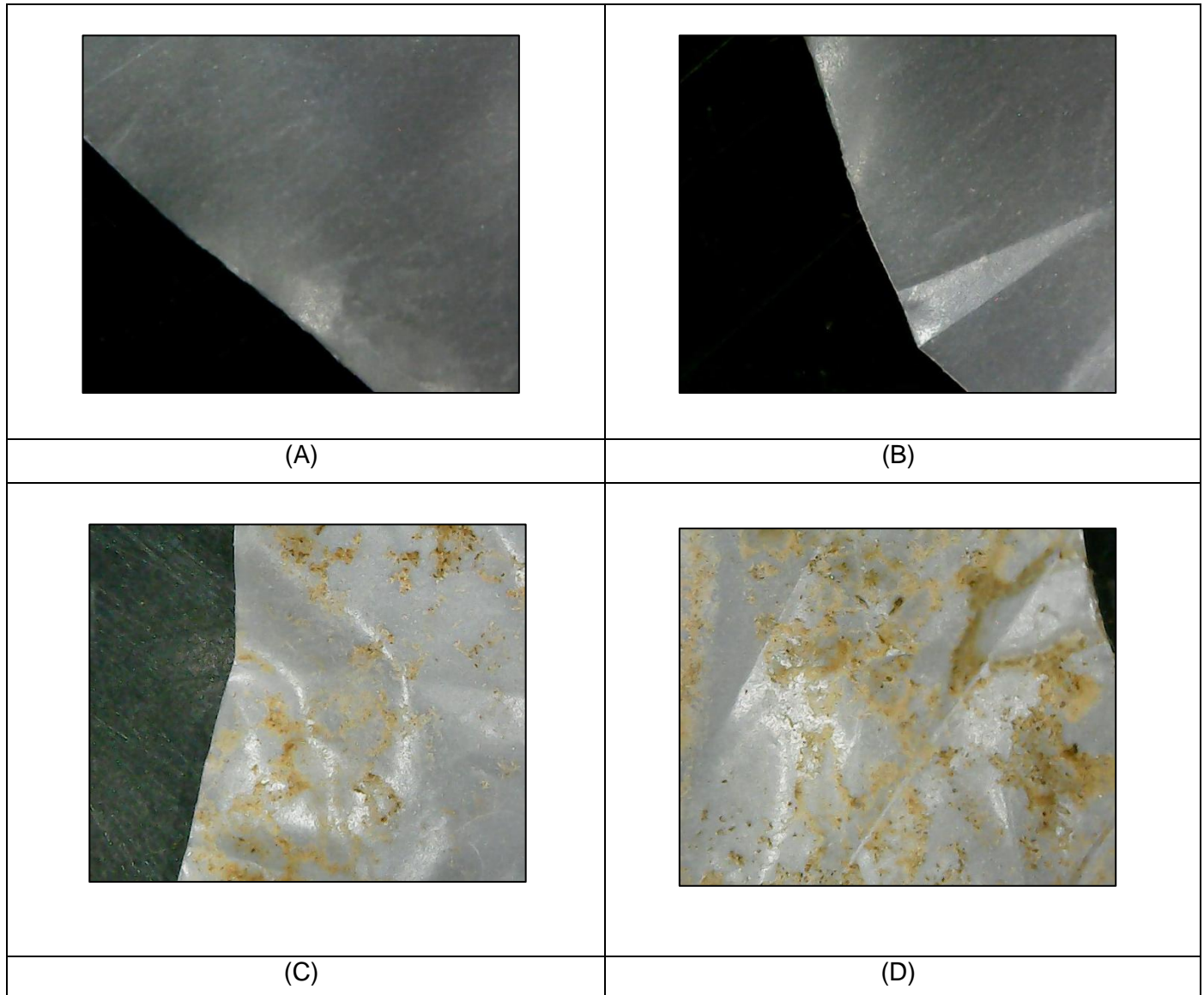


Figure 4: Microscopic image of Test samples Before and After 45 days Incubation Condition

A & B – Unexposed Test Sample Biodegradable Polyethylene Packaging to anaerobic biodegradation process
C & D – Exposed Test Sample Biodegradable Polyethylene Packaging to anaerobic biodegradation process

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The percent biodegradation of Positive control (Reference material) and Test sample was calculated by the measured cumulative carbon dioxide and methane production from each flask after subtracting carbon dioxide evolution and methane evolution from the blank samples at the end of 45 days of testing. Calculations were based on Total Organic Carbon obtained of both Positive control (Reference material) and Test sample.

Table-3a: Percentage biodegradability of Positive control (Reference material) Cellulose.

Group	Inoculum control	Positive control (Reference material)	Biodegradable Polyethylene Packaging Sample
Weight	1000 ml	10.4582 g	16.1992 g
Total volume (ml)	2470.00	9840.00	4450.00
% CH ₄	12.90	43.50	18.40
Volume of CH ₄ (ml)	318.63	4280.40	818.80
weight of CH ₄ (g)	0.2090	2.8079	0.5371
% CO ₂	14.10	43.80	19.50
Volume of CO ₂ (ml)	348.27	4309.92	867.75
Weight of CO ₂ (g)	0.6896	8.5336	1.7181
Total weight of carbon in grams	0.3430	4.4100	0.8667
Theoretical weight of carbon in grams (Ci)	-	4.3998	12.6295
Biodegradation	-	0.92439	0.04147
% Biodegradation	-	92.44	4.15

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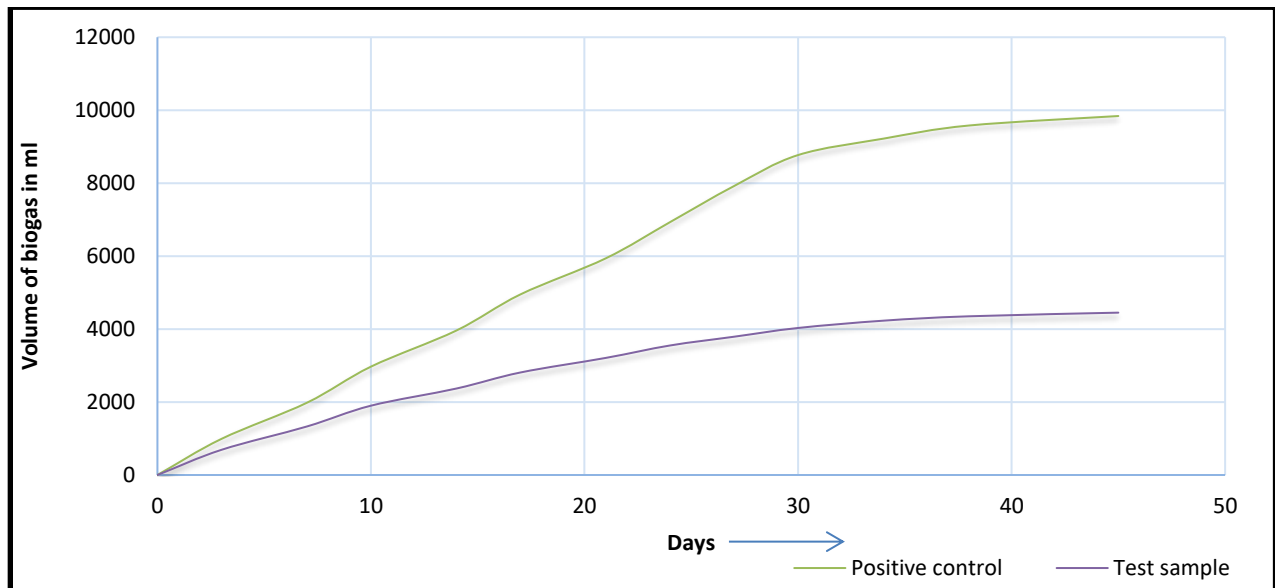
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Table 4: Percent weight loss of Biodegradable Polyethylene Packaging

Average Initial Weight (grams)	16.1992
Average Final Weight (grams)	16.1500
Percent Weight Loss (%)	0.30

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

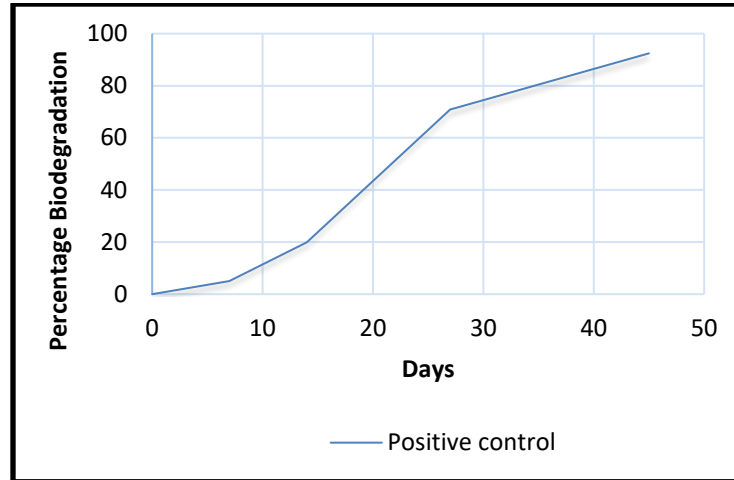
Biodegradation of the samples determined based on conversion of carbon from the test material to carbon in the gaseous phase (CH₄ and CO₂) can be observed in graph 1 and graph 2a & 2b.



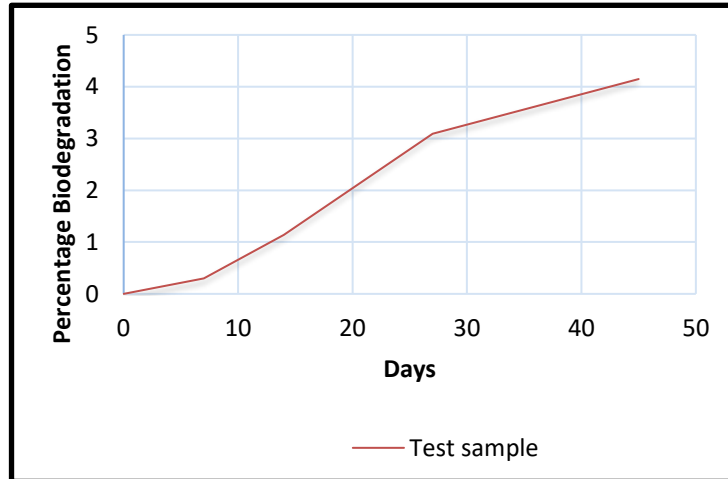
Graph-1: Plot showing Net Biogas Production from Test sample Biodegradable Polyethylene Packaging and Positive control (Reference material- Cellulose)

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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 (CH₄ and CO₂)



Graph-2b: The percent biodegradation of the Test sample Biodegradable Polyethylene Packaging determined based on conversion of carbon from the Test material to carbon in the gaseous phase (CH₄ and CO₂)

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INTERPRETATION:

Considering the cumulative gas production as observed in Table 2 & 3 and its analysis indicates that the process of biodegradation has occurred in **Biodegradable Polyethylene Packaging** Sample. After 45 days of incubation, the level of biodegradation for the Positive control (Reference material) was **92.44 %** while the **Biodegradable Polyethylene Packaging** sample submitted by Intertek Korea showed **4.15 %**.

Reviewed By

Authorized Signatory

Alok Pandey
Assistant Manager – Biodegradability Services

Ushadevi Yadav
Deputy Manager – Biodegradability Services

----- **End of Report** -----

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