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RESEARCH ARTICLE

Evaluation of Biodegradability Characteristics of Cellulose-based Film as per IS/ISO 14855-1

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ABSTRACT

Biodegradable polymers (especially those derived from plant sources) begin their lifecycle as renewable resources, usually in the form of starch or cellulose. In this paper, the evaluation of biodegradability of cellulose-based polymer film under controlled composting conditions as per the guidelines of IS/ISO 14855 (Part-1) standard has been described. Microcrystalline cellulose (MCC) powder was taken as positive-control polymer. The apparatus used to analyse the degree and rate of biodegradation was developed indigenously. The validation of the biodegradability testing apparatus was also performed as per the prescribed test method given in IS/ISO 14855-1.

KEY WORDS

Biodegradability, Cellulose, Composting, IS/ISO 14855-1

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INTRODUCTION

The development of innovative biodegradable polymers (or biopolymers) is in progress for a number of years, and continues to be an area of interest for many scientists. In addition, eco-concern played a key role in encouraging the development of biomaterials based on agricultural resources as alternative feed-stocks [1]. Cellulose and starch, proteins and peptides, DNA and RNA are all examples of biopolymers, in which the monomeric units are sugars, amino acids, and nucleotides respectively [2]. Cellulose is both the most common biopolymer and the most common organic compound on Earth, which amounts to about 33% of all plant matter is cellulose [3, 4]. The use of biodegradable polymers and their final method of biodegradation are dependent on the composition and processing method employed [5]. The biodegradable plastics must also be microbially and environmentally degraded upon disposal, without any adverse environmental impact [6].

Cellophane is a thin, flexible and transparent organic polymeric material made from cellulose which is a biodegradable polysaccharide. It is fabricated by dissolving cellulose in a mixture of sodium hydroxide and carbon disulphide to obtain cellulose xanthate which is then dipped into an acid solution (sulphuric acid) to yield cellophane film [7]. It is widely used as wrappers and in packaging food and merchandize since it is impermeable to gases, grease, or bacteria. It is a transparent, strong, flexible sheet, and is produced to many different specifications [8]. Cellophane can be left uncoated; however, it is often coated to improve the water vapour barrier and to make it heat-sealable. The coating may be a cellulose nitrate lacquer, such as pyroxylin, or more recently polyvinylidene chloride (PVDC), polyurethane/ poly(methylacrylate) interpenetrating polymer networks (IPN), etc. are the common waterproof coatings. Coated cellophane has low permeability to gasses, good resistance to oils, greases, and water, which is suitable for food packaging [9]. It also offers a moderate moisture barrier and is printable with conventional and offset printing methods. Cellophane has an interesting and unique optical property known as birefringence, i.e., when placed between two sheets of polarizing material, cellophane molecules act as tiny prisms, emitting colored light when white light passes through them. This characteristic, more common in crystalline structures like quartz, where changing the angle and orientation of the polarizing material and the cellophane changes the appearance of the resulting light has led some artists to use it in interactive artwork [10]. Cellophane is fully recyclable in composting environments, and will typically break down in just a few weeks. Due to its biodegradability, cellophane is gaining in popularity, especially in food production industry, where cellophane is used to wrap food [11].

There are several different methods used to determine the degree and rate at which materials biodegrade. Samples can be exposed to degrading enzymes or microbes in laboratory culture, or biodegraded by being submerged in soil, activated sludge or compost. Numerous metrics can be obtained from such experiments to quantify and assess the rate and extent of biodegradation [12, 13]. The most predominant form of aerobic biodegradation is through composting. Composting is considered to be nature's way of recycling, because compost is a hummus-like substance comprised of decomposed organic material, such as animal waste, vegetable waste, grass clippings, leaves, etc. that decomposes material into biomass to complete the lifecycle [14]. The measurement of carbon dioxide evolution under aerobic conditions, however, provides a quantitative measure of degradation. The use of such respirometric data can allow the calculation of the degree and rate of biodegradation during aerobic composting process itself, which is very convenient [15, 16]. The complete set of requirements for determining ultimate aerobic biodegradability of plastic materials under controlled compositing conditions test is mandated under the IS/ISO 14855-1 (2005) standard [17].

The present research paper discusses about the measurement of biodegradability of a commercially available cellophane film by using the precise method of evolution of carbon dioxide as per the guidelines of IS/ISO 14855-1 standard through indigenously developed biodegradability measuring apparatus and its validation.

MATERIALS AND METHODS

Materials

Cellophane film

Cellophane film having trade name Kesophane® having thickness 24-25 µm and substance 40 g/m^2 was supplied by M/s. Kesoram Rayon, Kolkata, India. The tensile strength, elongation at break and bursting strength of the film, as provided by the supplier, are 4 kg/15 mm, 40% and 2.2 kg/cm^2 respectively.

Compost medium

For biodegradability testing, the compost medium was provided by the Solid Waste Management unit, K/East ward, Brihanmumbai Mahanagarpalika Corporation (BMC), Mumbai, India. It was made mainly of household waste and yard trimming including grasses and leaves. It was screened through a mesh sieve of 10 mm size to separate outsized inert substances (stones, glass pieces, metallic parts, etc.). The measurements were conducted in triplicate and the moisture content result obtained was 52.47(±1.5) %. First, compost sample of 30 g was dried in an oven at 105 °C overnight to obtain the total solids (TS) content. The dried sample was then heated in a furnace at 550 °C for 1 hour to obtain the volatile solids (VS) content. The measurements yielded 51.72(±1.5) % TS and 17.04(±1.5) % VS. The microbial count of the compost was verified in our microbiology laboratory which came out to be 6.5×10^7 CFU per g of the compost. The pH of the compost was determined with a pH Meter (DBK Instruments, Mumbai, India) and it came out to be 7.93(±0.4). The carbon content of the compost was determined using elemental analysis method and nitrogen content by Kjeldahl method. The values of carbon and nitrogen content were 34.08% and 2.20%, respectively. These results reveal that the compost medium has a C/N ratio of 15.49 and falls within the required C/N range between 10 and 40.

Microcrystalline cellulose (MCC)

Microcrystalline cellulose powder of thin-layer chromatography grade with a particle size of less than 20 µm was procured from Sigma-Aldrich, India and used as the positive-control reference degradable polymer.

Reagents

Sodium hydroxide (NaOH) pellets having molecular weight of 40 and hydrochloric acid (HCl), having 35% concentration, were purchased from Merck Specialities Pvt. Ltd., Mumbai, India.

Apparatus

The apparatus for measurement of biodegradability was developed indigenously as per the guidelines of IS/ISO 14855-1 standard. The apparatus is basically an assembly of four parts – (a) Supply arrangement of CO_2-free air; (b) Composting vessels (or bioreactors); (c) Composting chamber with temperature control mechanism containing composting vessels; and (d) CO_2 absorbing vessels to quantify the amount of CO_2 release. The various parts of the apparatus were considered separately and the accuracy, compactness, reliability, and of course, the economy etc. were the main factors which were taken care of while designing. The apparatus consisted of nine composting vessels placed in three level partitions (3 vessels on each level) of the temperature controlled chamber. On each level, the first vessel was taken as blank (containing compost only); the second one was taken as positive control (containing microcrystalline cellulose); and the last one was taken as the test (containing the test sample). The CO_2 gas evolved from each composting vessel was absorbed in three vessels (in series) containing sodium hydroxide solution.

Validation of the Process

All the composting vessels were filled with 250 g of the compost. In six of the nine composting vessels, 2 × 2 cm strips of the cellophane film were placed in the middle of the compost medium to ensure maximum surface contact between the compost and the sample. After weighing, all the composting vessels were incubated under optimum air flow rate, temperature and moisture for a period of 15 days. CO_2-free air (also known as Zero Air) was supplied to the composting vessels with a flow rate of 60 mL per minute throughout the experiment to ensure enough oxygen for the biodegradation process. The temperature and moisture content were kept at 58±2 °C and 55-60%, respectively. The water content in the composting vessels was maintained every 4 days to adjust the moisture level to 55-60 %. Once in a week, the experiment was stopped for a while, the composting vessels were emptied, the test samples were taken out, the compost was re-mixed homogenously using a spatula, distilled water was added (if required) to maintain the humidity and the contents were added back to their respective composting vessels.

Sample Testing

After validating the process of the system for 15 days, the degree and rate of the biodegradation was analysed for next 180 days (6 months) under the same conditions (58±2°C temperature and 55-60 % relative humidity), which is representative of full-scale composting, as mentioned in IS/ISO 14855-1 standard. The incubation period was ended till a constant plateau phase was reached. Aerobic conditions were maintained by providing continuous supply of sufficient airflow to the composting vessels and to compensate for the water loss, the contents were hydrated and mixed well once a week. Biodegradation behaviour of microcrystalline cellulose powder was also measured to verify its suitability as positive reference material.

Calculations

During aerobic biodegradation, the carbon present in polymer molecules is converted into gaseous carbon dioxide, water, biomass (or humus) and carbon residues through the respiration process by microorganisms as shown in Eq. 1 [18].

$$C_{polymer} + O_2 \rightarrow CO_2 + H_2O + C_{biomass} + C_{residue} \qquad (1)$$

The theoretical amount of CO_2 produced by the total oxidation of the test sample was calculated by the using Eq. 2.

$$CO_2(Th) = W_{sample} \times C_{sample} \times \frac{44}{12} \qquad (2)$$

where W_{sample} is the mass of total dry solids in the test sample initially added to the bioreactors (g); C_{sample} is proportion of total organic carbon in the sample (g/g); 44 and 12 are the molecular weights of carbon dioxide and carbon, respectively (g/mol).

To estimate the actual amount of CO_2, the evolved gas from each bioreactor was absorbed in three flasks in series, each containing 1500 mL sodium hydroxide (NaOH) solution. When CO_2 enters the absorbing vessels, it reacs in the following manner:

$$2NaOH + CO_2 \rightarrow Na_2CO_3 + H_2O \qquad (3)$$

The Na_2CO_3 formed is insoluble and precipitates. The retrieved solution was determined by end-point titration with 0.05 N HCl solution using phenolphthalein as indicator solution. The mean value (from the three replicates) of the net CO_2 produced by controlled composting of the test substances was determined by subtracting the mean CO_2 production from the blank bioreactor, i.e., the compost only. The % biodegradation was calculated by using Eq. 4.

$$\%\,biodegradation = \frac{CO_2(test) - CO_2(blank)}{CO_2(Th)} \times 100 \qquad (4)$$

where CO_2 *(test)* is the cumulative amount of carbon dioxide evolved from each composting reactor containing a test sample (g) and CO_2 *(blank)* is the cumulative amount of carbon dioxide evolved from the blank reactor (g). Since the cumulative CO_2 depends largely on pH value of the compost, it was monitored continuously (by Hanna make Soil pH meter; Model: Groline HI981030) and maintained within the prescribed values.

Statistical Analysis

Results are reported as mean ± standard deviation (n = 3). Pearson correlation coefficient between various parameters was estimated by using Origin Software. The differences in the values of the observed parameters were considered statistically significant when $p < 0.05$.

RESULTS AND DISCUSSION

The investigations were carried out in controlled compost based on IS/ISO 14885-1 standard and the calculations/results of alternative day are shown in Table 1.

The graph obtained by putting the values obtained for percent biodegradation of cellulose (positive-control) and the test sample is shown in Figure 1.

The results obtained, as shown in Table 1 and Figure 1 above suggests that the degradation of cellulose was approx. 86% after 45 days and 97% after 180 days, which proves that the cellulose powders are effective as a reference material for IS/ISO 14855-1. No induction period of biodegradation curves is observed for the positive-control material, i.e., MCC, which may be due to the fact that cellulose can be directly degraded by enzymes such as lipase and cellulase. The biodegradation test was terminated when the biodegradation curve show plateau phase. After 45 days of incubation period, the test sample biodegraded to approx. 73% and the ultimate biodegradation of the test sample, after 180 days of the composting period was obtained as 96% approx. Since the carbon atoms from samples left in a compost cannot be accurately evaluated, therefore the final degree of biodegradation of cellulose are not equal to 100%. The results suggest that the cellophane film is conforming to the criteria of the standard for biodegradability.

The photographs of the cellulosic films, taken during initial period, after three months and after six months of biodegradation in compost were also taken and shown in Figure 2. As the time progressed, the film samples were rapidly biodegraded by compost microorganisms and by end of six months, the film fragments could no longer be distinguished from soil. These results are supported by several other research studies that have reported biodegradation of biodegradable polymer film by soil microorganisms [19, 20].

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Time (Days)	CO_2 Produced (g)		Biodegradation (%)	
	MCC (Positive Control)	Kesophane® (Test Sample)	MCC (Positive Control)	Kesophane® (Test Sample)
0	0.000	0.000	0.000	0.000
1	1.572	0.163	9.646	0.965
2	2.721	0.330	16.696	1.953
4	3.954	0.480	24.261	2.839
6	4.786	0.669	29.370	3.962
8	5.492	0.789	33.701	4.671
10	5.933	0.867	36.406	5.132
12	6.346	1.146	38.943	6.787
14	6.817	1.510	41.832	8.941
16	7.082	2.317	43.459	13.719
18	7.591	2.806	46.582	16.614
20	8.749	3.682	53.688	21.801
22	9.193	4.382	56.413	25.946
24	9.572	5.728	58.738	33.916
26	10.248	6.783	62.887	40.162
28	11.052	7.273	67.820	43.064
30	11.428	7.937	70.128	46.995
32	12.062	9.392	74.018	55.610
34	12.272	10.782	75.307	63.840
36	12.739	11.048	78.173	65.415
38	13.015	11.382	79.866	67.393
40	13.537	11.720	83.069	69.394
42	13.974	11.973	85.751	70.892
44	14.035	12.293	86.125	72.787
46	14.215	12.480	87.230	73.894
48	14.358	12.553	88.108	74.326
50	14.527	12.619	89.145	74.716
52	14.539	12.784	89.218	75.693
54	14.726	12.839	90.366	76.018

Table 1: Results obtained for CO_2 Produced and Percent Biodegradation

Time (Days)	CO_2 Produced (g)		Biodegradation (%)	
	MCC (Positive Control)	Kesophane® (Test Sample)	MCC (Positive Control)	Kesophane® (Test Sample)
56	14.821	12.860	90.949	76.145
58	14.829	12.959	90.998	76.731
60	14.830	13.036	91.004	77.186
62	14.902	13.091	91.446	77.511
64	14.918	13.091	91.544	77.510
66	14.921	13.135	91.562	77.770
68	14.925	13.178	91.587	78.026
70	14.930	13.266	91.618	78.546
72	14.951	13.309	91.746	78.804
74	14.983	13.364	91.943	79.128
76	15.000	13.419	92.047	79.451
78	15.008	13.462	92.096	79.710
80	15.030	13.484	92.231	79.840
82	15.071	13.655	92.483	80.852
84	15.075	13.691	92.507	81.065
86	15.103	13.705	92.679	81.147
88	15.219	13.775	93.391	81.561
90	15.328	13.845	94.060	81.975
92	15.372	13.915	94.330	82.389
94	15.440	13.985	94.747	82.803
96	15.471	14.055	94.937	83.217
98	15.498	14.124	95.103	83.631
100	15.507	14.194	95.158	84.045
102	15.516	14.264	95.214	84.459
104	15.528	14.334	95.287	84.872
106	15.604	14.404	95.754	85.286
108	15.661	14.474	96.103	85.700
110	15.682	14.873	96.232	88.063
112	15.750	15.108	96.649	89.455
114	15.800	15.361	96.956	90.953
116	15.809	15.582	97.012	92.261
118	15.818	15.609	97.067	92.421
120	15.824	15.629	97.104	92.540
122	15.827	15.631	97.122	92.551

Time (Days)	CO_2 Produced (g)		Biodegradation (%)	
	MCC (Positive Control)	Kesophane® (Test Sample)	MCC (Positive Control)	Kesophane® (Test Sample)
124	15.829	15.633	97.134	92.564
126	15.830	15.690	97.140	92.901
128	15.830	15.695	97.140	92.930
130	15.833	15.713	97.159	93.037
132	15.838	15.772	97.189	93.386
134	15.839	15.781	97.196	93.440
136	15.840	15.796	97.202	93.528
138	15.841	15.798	97.208	93.540
140	15.846	15.799	97.239	93.546
142	15.849	15.800	97.257	93.552
144	15.850	15.801	97.263	93.558
146	15.850	15.802	97.263	93.564
148	15.851	15.802	97.269	93.563
150	15.853	15.812	97.282	93.621
152	15.855	15.812	97.294	93.621
154	15.855	15.816	97.294	93.647
156	15.855	15.852	97.294	93.857
158	15.860	15.851	97.324	93.856
160	15.860	15.891	97.324	94.093
162	15.860	15.861	97.324	93.914
164	15.860	15.900	97.324	94.144
166	15.860	15.901	97.324	94.150
168	15.860	15.941	97.324	94.386
170	15.860	15.941	97.324	94.386
172	15.860	15.941	97.324	94.386
174	15.860	16.006	97.324	94.772
176	15.860	16.051	97.324	95.035
178	15.860	16.170	97.324	95.743
180	15.860	16.170	97.324	95.743

Time (Days)	CO_2 Produced (g)		Biodegradation (%)	
	MCC (Positive Control)	Kesophane® (Test Sample)	MCC (Positive Control)	Kesophane® (Test Sample)
124	15.829	15.633	97.134	92.564
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128	15.830	15.695	97.140	92.930
130	15.833	15.713	97.159	93.037
132	15.838	15.772	97.189	93.386
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136	15.840	15.796	97.202	93.528
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164	15.860	15.900	97.324	94.144
166	15.860	15.901	97.324	94.150
168	15.860	15.941	97.324	94.386
170	15.860	15.941	97.324	94.386
172	15.860	15.941	97.324	94.386
174	15.860	16.006	97.324	94.772
176	15.860	16.051	97.324	95.035
178	15.860	16.170	97.324	95.743
180	15.860	16.170	97.324	95.743

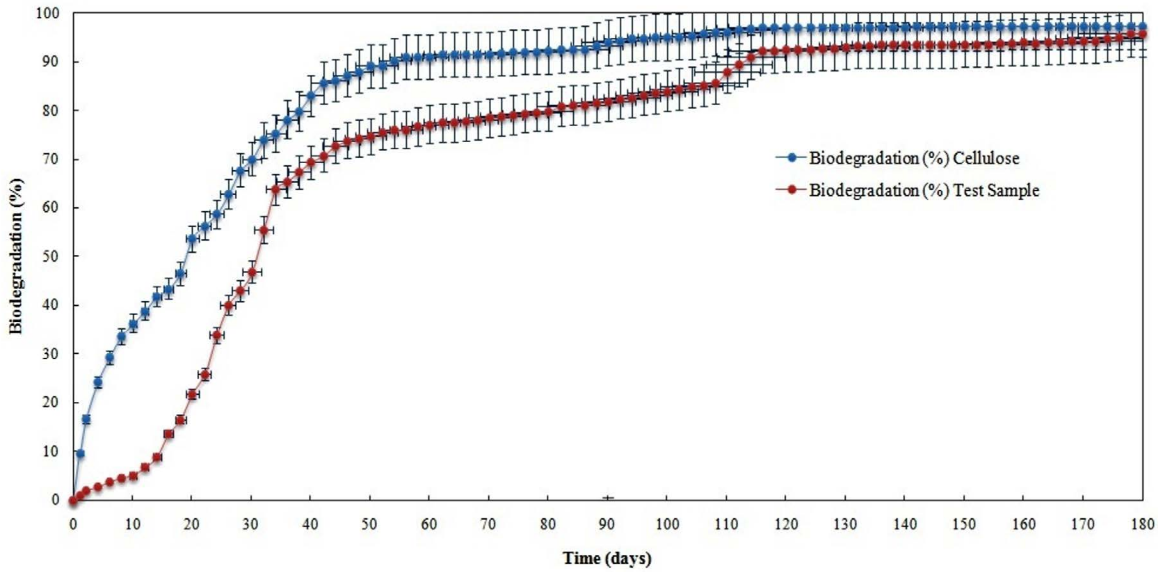

Figure 1: Biodegradation Graph for positive-control (cellulose) and the test sample

(a) **(b)** **(c)**

Figure 2: Photographs of cellophane film – (a) Before incubation; (b) After 3 months of incubation in compost; and (c) After 6 months of incubation

ultimate biodegradation of the test sample, after 180 days of the composting period was obtained as 96% approx. Since the carbon atoms from samples left in a compost cannot be accurately evaluated, therefore the final degree of biodegradation of cellulose are not equal to 100%. The results suggest that the cellophane film is conforming to the criteria of the standard for biodegradability.

The photographs of the cellulosic films, taken during initial period, after three months and after six months of biodegradation in compost were also taken and shown in Figure 2. As the time progressed, the film samples were rapidly biodegraded by compost microorganisms and by end of six months, the film fragments could no longer be distinguished from soil. These results are supported by several other research studies that have reported biodegradation of biodegradable polymer film by soil microorganisms [19, 20].

CONCLUSIONS

The biodegradability of cellulose-based plastic film sample (Kesophane®) under controlled composting conditions (at 58 °C and 50-60 % RH) were investigated according to IS/ISO 14855-1 in an indigenously developed biodegradability testing apparatus. The validation of the biodegradation tests using the apparatus was performed with cellulose powder as the reference material. It was found that the film samples fulfils the criteria of biodegradability under controlled composting conditions as per IS/ISO 14855-1 using the biodegradability testing apparatus.

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