STUDIES ON DEGRADATION OF SYNTHETIC POLYMER NYLON 6 BY LIGNOLYTIC FUNGUS *Phanerochaete chrysosporium* NCIM 1073

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ABSTRACT

A study of nylon 6 polymer degradation by lignolytic fungus *Phanerochaete crysosporium* has been carried out under submerged conditions. The analysis was carried out using weight and thickness measurements, and I. R. spectroscopy. Sheets of nylon 6 were inserted in fermentation broth which was incubated on a rotary shaker at 30^{0}C and 90 rpm. Nylon 6 was the sole source of nitrogen in the medium. A colony of fungus was developed that resulted in substantial degradation via crack within 75 days. The groove that weakening and breaking of polyamide bond have been confirmed by weakening in I R band and molar mass reductions have been observed in morphological studies.

Key Words : Nylon 6, Degradation, *Phanerochaete chrysosporium*, Plastic waste management, Fungus

INTRODUCTION

Biodegradation of polymers is seen as one of the solution for current plastic waste management problems. By definition biodegradable polymers are those that are degraded into carbon dioxide, water and biomass as a result of the action of living organism or enzymes. The rate of biodegradation may vary substantially and depends on the molecular structure, morphology, surface area etc[1]. Among polymer category polyamide types of polymer have been considered as a long standing existence. Therefore find, diversifies applications by all commodities. Polymers bearing recurring amide groups in their backbone are defined as polyamide. Nylon 6, one of the polyamide polymers finds its application in the bristles, ropes, fabrics, fishing nets, automobile parts etc.The very bad degradability of nylon is assumed to be due to its strong intermolecular cohesive force caused by hydrogen bonds between molecular chains[2]. Furthermore, composting has been attracting a great interest as a method of waste management for plastic material[3,4]. F Aliphatic polyamides, such as nylon 66 and nylon 6 have been produced industrially since the late 1930s and continue to be important materials[5,6]. Nylon 6 is a polymer obtained by ring - opening polymerization of ε caprolactam and has several commercial names including perlon, nylon and steelon[3]. **(Fig. 1)**

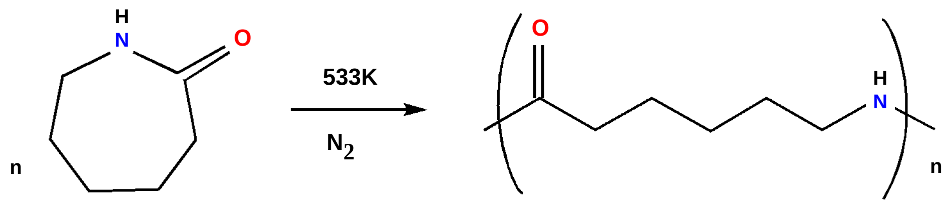

Fig.1:Chemical reaction showing preperation of Nylon 6

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Several attempts have been made to degrade these materials using microorganism. In an oligomer consisting of a small number of monomeric units of nylon - 6, biodegradation was observed only with *Pseudomonas and Flavobacterium*[7,8]. Nylon 6 monomer is metabolized by numerous microorganisms, including the bacterial genera *Pseudomonas, Achromobacter and Corynebacterium* and fungal genera *Bjerkundera adusta*[9]. In addition to the environmental point of view microbial degradation of synthetic compounds provides a good system for studying how microorganisms acquire the ability to degrade such compound[10,11].In this paper we report on nylon biodegradation using fungi *Phanerochaete chrysosporium* in a submerged cultivation using nitrogenous nutrient broth as stimulator. Degradation of polymer was evaluated by measurement of relative viscosity, mechanical methods and by using FTIR. So far, several investigations on nylon degradation using microorganisms have been reported. Kouke Temita in 2003 reported thermophilic bacterium degradation of some kinds of nylon.

MATERIAL AND METHODS

Materials

Commercial grade nylon 6 was provided by Sigma Aldrich. The material is in the form of pellet. All chemicals and solvents used in all experiment were AR grade. A thin sheet of nylon 6 was prepared from nylon 6 pellets by melting and pressing the pellets of nylon 6.

Selection of Fungi

The fungus selected in our study was a white rot fungus *Phanerochaete chrysosporium* that is well known for its lignolytic activity[10]. The degradation was confirmed by various observations. The tested fungal strain of the species *Phanerochaete chrysosporium* NCIM 1073 was obtained from the National Collection of Industrial Microorganisms (NCIM), NCL, Pune, Maharashtra, India.

Sterilization of the sample

The sample sheets were sterilized before they were inoculated into the test medium. The nylon sheets were dipped in 1 % hypochlorite for a few hours. Washed with distilled water thoroughly in order to remove all the hypochlorite and later dried. No physical or chemical changes were observed in the sample after hypochlorite treatment

Submerged Cultivation Procedure

A nylon sheet was exposed to submerged cultivation process Microbial degradation was performed in Erlenmeyer flasks in shaker condition. Each flask containing small sheet of nylon 6 and 100 ml of the liquid glucose - salt medium at a pH 6.25. The medium contained per liter of deionised water, 10 gm of glucose, 1 g KH_2PO_4, 0.5 gm of $MgSO_4$ $7H_2O$ and 0.5 gm of $(NH_4)_2$ SO_4 and 0.1 gm of $CaCl_2$. Aliquats (100 ml) of medium were poured into 500 ml Erlenmeyer flask and sterilized in an autoclave for 20 min at 121^{o}C and 1.2 atm, after cooling the medium was inoculated with 5 ml of the fungi spore suspension obtained by suspending spores from one agar slant tube in 20 ml sterile water. The fermentation broth was incubated on a rotary shaker at 30^{o}C and 90 rpm. Nylon 6 was the sole source of nitrogen in the medium. Degradation was left to proceed for a period up to 75 days.

RESULTS AND DISCUSSION

Degradation of nylon 6 sheets in submerged cultivation using *Phanerochaete chrysosporium* was carried out for 15, 30, 45, 60 and 75 days. Parallel to samples inoculated with the fungus

Table 1: Decrease in weight of Nylon 6 by treating with fungi *Phanerochaete chrysosporium* NCIM 1073

Time Duration	Control (A biotic) (Weight in gm)	Sample (Biotic) (Weight in gm)
0 Days	0.013	0.013
15 Days	0.013	0.011
30 Days	0.012	0.009
45 Days	0.010	0.008
60 Days	0.009	0.008
75 Days	0.009	0.006

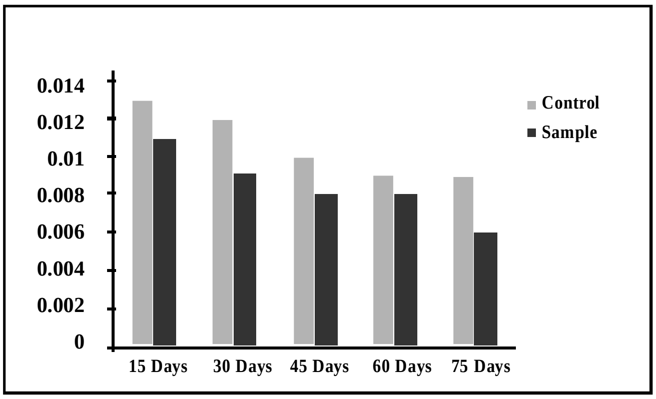

Fig. 2 : Decrease in weight of nylon 6 by treating with fungi *Phanerochaete chrysosporium* NCIM 1073

Table 2: Decrease in thickness of nylon 6 by treating with fungi *Phanerochaete chrysosporium* NCIM 1073

Time Duration	Control (A biotic) (Thickness in mm)	Sample (Biotic) (Thickness in mm)
0 Days	0.118	0.118
15 Days	0.118	0.101
30 Days	0.115	0.096
45 Days	0.114	0.087
60 Days	0.107	0.085
75 Days	0.105	0.035

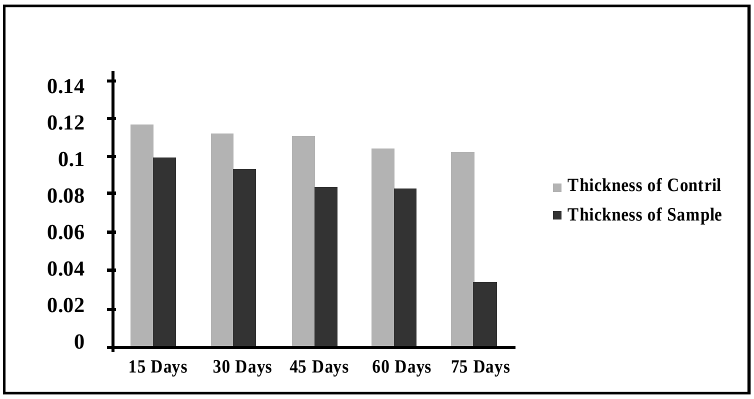

Fig. 3 : Decrease in weight of nylon 6 by treating with fungi *Phanerochaete chrysosporium* NCIM 1073

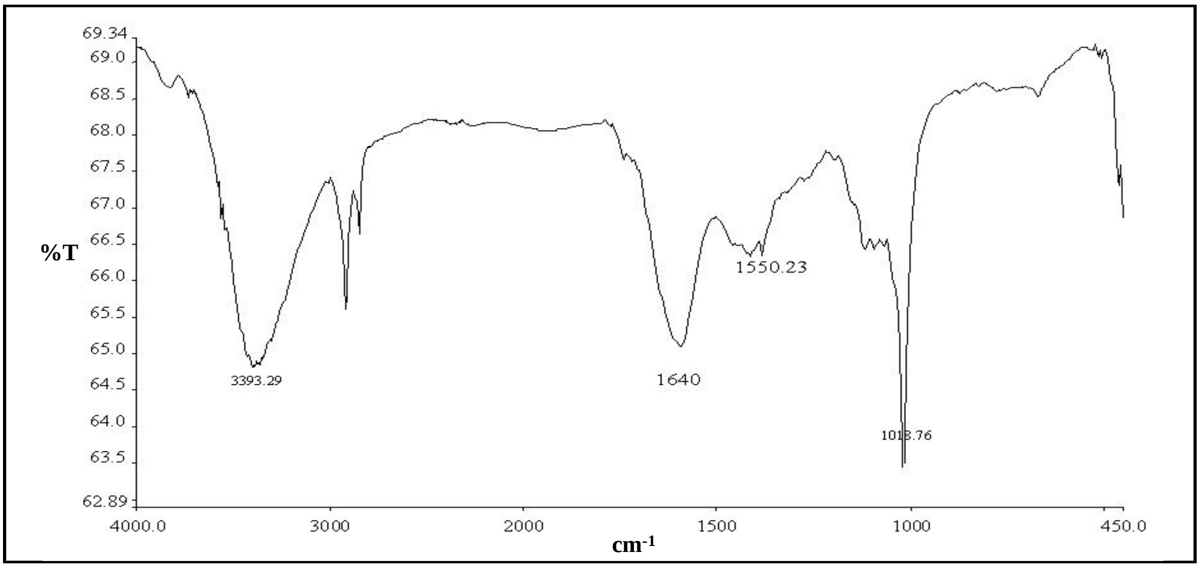

Fig. 4 : FTIR Spectra of Nylon 6 Untreated

(biotic sample) blank experiment were carried out with nylon 6 sheets placed equally composed media without the fungus (a biotic sample) results presented in the following sections were obtained from two independent sets of degradation experiments.

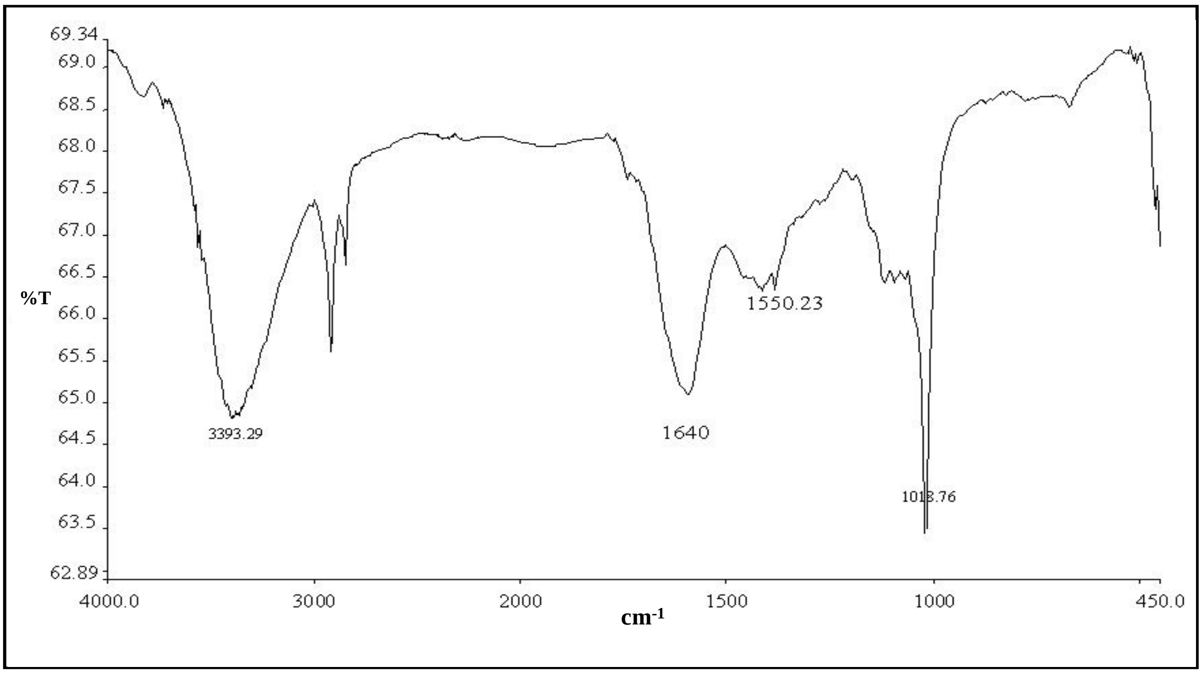

Fig. 5 : FTIR Spectra of Nylon 6 Treated.

Morphological Study

Degradation of sheets of nylon 6 was observed my measuring thickness of sheets and weight reduction of sheets as it is treated with selected fungi. The color of sheets becomes brownish and surface morphology become rough when sheets are exposed to fungi.

FTIR Spectra of Nylon 6 *Phanerochaete chrysosporium* NCIM 1073 treated 75 days samples (the strength of characteristic bands of C(O)NH occurring around 3300, 1640 and 1550 cm^{-1} decreased after 75 days)

Weight loss

The undegraded polymer sheets were removed from medium, washed with distilled water and dried. The amount of degradation was determined by studying weight loss of sheets. From **Table 1** which shows that weight of control sheets were reduced by 0.01 to 0.02 gm as 15 days time duration was passed. But in case of samples (biotic) sheets treated with fungi shows within 30 days and after this the weight reduces by 0.01mg of difference. **Fig. 2** shows weight loss of nylon 6 sheets samples exposed to fungi.

Decrease in thickness

Thickness of nylon 6 sheets were observed by micrometer. The initial thickness of nylon sheet sample was 0.118 mm. Sheets which were treating with fungi phanerochaete chrysosporium NCIM 1073 show maximum thickness decrease within 30 days while control sheets shows decrease in thickness by 0.1 mm after 30 days. After completion of time duration of 45 days the thickness was remain constant and finally the thickness was reduced from 0.118 mm to 0.035 mm in sample.

FTIR study of nylon 6

Infrared spectroscopy is one of the most often used spectroscopic tools for the study of polymers. The functional groups of nylon 6 mediated with fungus *Phanerochaete chrysosporium* NCIM 1073 at time intervals were determined using FTIR Spectroscopy (Fourier transformed Infrared Spectroscopy) at room temperature in the transmission mode.

The strength of characteristic bands of C (O) NH occurring around 3300, 1640, 1550 and 1018 cm^{-1} decreased after 75 days. Formation of new group like COOH and CHO may be form due to hydrolysis and oxidation.

CONCLUSION

It was found that in submerged cultures no polymer degradation. At different cultivation conditions used (surface vs. submerged) and after changing the medium, shows nylon degradation by ligninolytic white rot fungi, *P. chrysosporium*[12,13,14&15] . Fungus *Phanerochaete chrysosporium* NCIM 1073 mediated biodegradation of nylon 6 is reported in this paper. Degradation of polymer was confirmed by weight loss, decrease in thickness, decrease in intensity of functional groups like C (O) NH, C-O and formation of new functional groups like COOH and CHO etc. The color of sheets becomes brownish and surface morphology become rough when sheets are exposed to fungi which shows effect of enzymatic activity on nylon 6.

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