Available online at www.scholarsresearchlibrary.com

Scholars Research Library

European Journal of Zoological Research, 2014, 3 (2):6-17
(http://scholarsresearchlibrary.com/archive.html)

ISSN: 2278–7356

Growth of *Actinomycetes* and *Pseudomonas sp.*, biofilms on abiotically pretreated polypropylene surface

Umamaheswari Sepperumal* and Murali Markandan

P.G and Research Department of Zoology, Periyar EVR College, Tiruchirapalli, Tamil Nadu, India

---

ABSTRACT

Doggedness of polypropylene flake in the environment is known to cause detrimental effects on the biota. The present study reports the role of Pseudomonas sp. and Actinomycetes sp., as possible candidates for biodegradation of polypropylene flake (straw). Pre-treatment of polypropylene flakes with UV, heat and HNO_3, resulted in cracking ,granules and roughness of polypropylene surface as evinced by SEM images. It was motivating to observe that UV, heat, and HNO_3 treated polypropylene flake on inoculation with Pseudomonas sp. and Actinomycetes sp., lead to crystal formation. Further, the polypropylene flake degradation was confirmed by FTIR studies, which indicated the formation of aromatic ether , hydroxyl groups and ester groups as evinced in UV ; heat ; HNO_3 ; UV, heat and HNO_3 treated polypropylene flakes.

Key words: Polypropylene, *Actinomycetes sp., Pseudomonas sp.*, FTIR, SEM.

INTRODUCTION

Polypropylene (PP) is an attractive thermoplastic with cheap production cost, low density, excellent mechanical properties and wide industrial and commodity applications such as textile fibres, hygiene (diapers, disposals), medical devices (operation gowns and covers and drug packaging), food packaging, absorbents, wipes, filters and polypropylene brings a lot of convenient to peoples life as a result of the diversity of its applications in a high volume of consumer products. Large amount of plastic waste is also generated which will be deposited in the environment after their use and disposal. In environment this dumped plastic waste is subjected to solar radiation, UV rays and heat, which affect their surface as well as to some extent their bulk properties. But this deterioration or degradation process is very slow [1]. On the other hand, bacteria, fungi and other microorganisms present in the environment also attack the polymer in directly to utilize it as a carbon source. The crucial step for initiation of this biodegradation is the attachment of the microorganism to the polymer surface, thus forming biofilms [2]. However, Polypropylene, since it is highly hydrophobic with high molecular weight, lacking active functional group with continuous chain of repetitive methylene units, it shows resistance to biodegradation [3]. Treatments including UV, thermal and chemical lead to the oxidation of the polymer surface aiding in the formation of carbonyl, carboxyl and ester functional groups [4, 5], which decreases the hydrophobicity of the surface [6]. This ultimately helps in the formation of microbial biofilm on its surface [7]. In this study, *Pseudomonas sp*. and *Actinomycetes sp*., capable of colonizing PP surfaces were evaluated in order to elucidate whether the films are easily colonised by autochthonous microorganisms. The present study aims to visualise electron microscopically the event of changes that has occurred during the PP biodegradation process.

MATERIALS AND METHODS

Isolation of polypropylene degrading microorganisms from polypropylene waste

The collected polypropylene waste collected from soil in different areas were scrapped several times with care to remove the soil and were cut into small pieces . Further, Polypropylene was washed with distilled water and inoculated into Nutrient Broth medium at room temperature for 24 hour. Identification of bacteria was performed

on the basis of microscopic examination and biochemical test according to Bergey's manual of determinative bacteriology [8].

Biotic and abiotic treatment of Polypropylene samples

Polypropylene (PP) straw was purchased from shop and was cut into small flakes about 0.5×0.5 mm size and were washed with distilled water. Further, they were treated with HNO_3 (boiled at 100 ^{0}C for 15 minutes), UV radiation (10 days in UV chamber) and heat treated (100 ^{0}C for 15 minutes) as shown in table-1. Treated PP samples were washed thoroughly with 70% ethanol and finally washed with distilled water thereafter kept in oven at 50 ^{0}C for one hour [9]. Further, they were inoculated into nutrient broth medium along with *Pseudomonas sp.* and *Actinomycetes sp* ., for a period of one month.

Table-1: **Treatment of PET flakes with UV, heat, HNO_3 and inoculation with *Pseudomonas sp.* and *Actinomycetes sp.*,**

Treatment factors	Control	Treated PET inoculated with *Pseudomonas species.*, and *Actinomycetes sp.*,
UV	-	+
HNO_3	-	+
Heat	-	+
Heat + HNO_3+UV	-	+

_ Not exposed to treatment factors
+ Exposed to treatment factors

FT-IR Spectrophotometer studies

Fourier transform infrared (FT-IR) measurements were carried out with a BIO-RAD spectrometer (model FTS 40A) in the range of 4000-400 cm^{-1}. The FT-IR spectra were recorded at a resolution of 2 cm^{-1} and an accumulation of 32 scans.

Scanning Electron Microscopy (SEM)

The scanning electron microscopic analysis of fractured surface of PP film was carried out using Scanning electron microscope (VEGA3 TESCAN). The surface of the treated PP samples were coated with conductive heavy metals such as gold/ palladium.

RESULTS

Analysis of surface degradation by SEM

The electron micrograph of the untreated polypropylene flakes exhibited smooth surface (Fig-1). There was no visible structural changes on the surface of the UV exposed polypropylene flake surface (Fig-2). Microscopic characterization revealed successful colonisation of *Pseudomonas sp.,* and *Actinomycetes sp.*, on the surface UV treated PP flakes inoculated with *Pseudomonas sp.,* and *Actinomycetes sp.,* (Fig-3). On other hand, morphological changes such as crack formation and granules were observed on treatment of polypropylene flakes with HNO_3 (Fig-4). Further, on inoculation with *Pseudomonas sp.,* and *Actinomycetes sp.,* rough surface along with *Actinomycetes sp.,* colonies were evident (Fig-5). Uneven and rough surface was observed in heat treated polypropylene flake surface (Fig-6). In addition to the *Actinomycetes sp.* colonies, a number of irregularities appeared on the surface of heat treated PP inoculated with *Pseudomonas sp.,* and *Actinomycetes sp.,* (Fig-7). Roughness of PP surface was observed in the SEM micrograph on exposure to heat, UV and HNO_3 (Fig-8). Furthermore, on inoculation with *Pseudomonas sp.,* and *Actinomycetes sp.,* granules and roughness of the PP surface was evident . In addition , growth of *Actinomycetes sp.,* on the PP surfaces was evident (Fig-9).Thus the abiotically treated PP on inoculation with microbes like *Pseudomonas sp.,* and *Actinomycetes sp.,* could cause structural modifications of the PP flake surface.

Fig 1

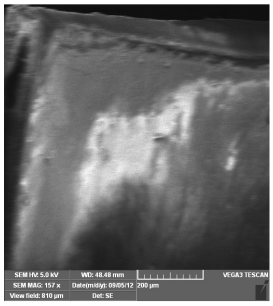


Fig 2

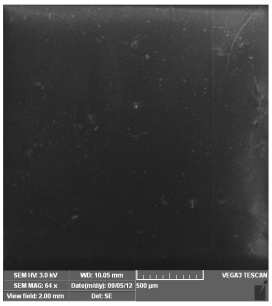


Fig 3

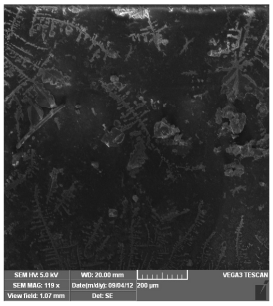


Fig 4

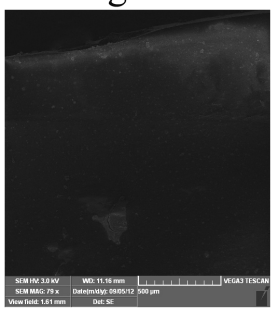


Fig 5

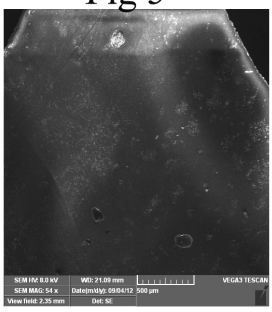


Fig 6

Fig 7

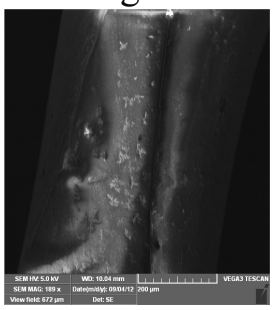


Fig 8

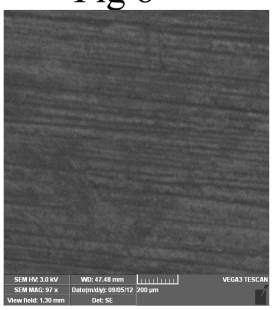


Fig 9

Fig -1 SEM imge of untreated Polypropylene flakes
Fig -2 SEM image of UV treated Polypropylene flakes
Fig -3 SEM image of UV treated Polypropylene flakes inoculated with *Pseudomonas sp*. and *Actinomycetes sp.,*
Fig-4 SEM image of HNO_3 treated Polypropylene flakes
Fig-5 SEM image of HNO_3 treated Polypropylene flakes inoculated with *Pseudomonas sp.* and *Actinomycetes sp.,*
Fig -6 SEM image of heat treated Polypropylene flakes
Fig-7 SEM image of heat treated Polypropylene flakes inoculated with *Pseudomonas sp.* and *Actinomycetes sp.,*
Fig-8 SEM image of UV, HNO_3 and heat treated polypropylene flakes
Fig-9 SEM image of UV, HNO_3 and heat treated polypropylene flakes inoculated with *Pseudomonas sp. and Actinomycetes sp.*,

Analysis of surface degradation by FT-IR microscopy

Fourier transform infrared spectrophotometer (FTIR) spectra of untreated and treated PP flakes were performed from 4000 cm^{-1} to 400 cm^{-1} wave numbers. The absorptions bands at 2875 cm^{-1} , 2723 cm^{-1}, 2582 cm^{-1}, 2369 cm-1,1446 cm^{-1} ,1380 cm^{-1} , 1304 cm^{-1} ,1167 cm^{-1},998 cm^{-1}973 cm^{-1},899 cm^{-1} , 875 cm^{-1},841cm^{-1},809 cm^{-1},and 713 cm^{-1} has been attributed to C-H bond stretching (methylene group) ,1256 cm^{-1} and 1103 cm^{-1} to C-O bond stretching (ether or alcohol formation). Absorption band at 1044 cm^{-1} correspond to O-C-O bond stretching (Fig-10). It is inferred from fig -11 that polypropylene flakes on exposure to UV radiation elicited appreance of new absorption peaks at 2926 cm^{-1} and 2354 cm^{-1} , which has been assigned to C-H bond stretching, 1685 cm^{-1} and 1524 cm^{-1} which has been attributed to C=O bond stretching (ester group formation), 1220 cm^{-1} has been assigned to O-C-O bond stretching and 772 cm^{-1} to C-H bond stretching when compared to untreated PP FTIR spectrum (Fig -11).On inoculation of UV treated PP flakes with *Pseudomonas sp.*, and *Actinomycetes sp* ., appearance of new absorption peaks at 2840 cm^{-1},2345 cm^{-1},1456 cm^{-1} ,1304 cm^{-1},940 cm^{-1} and 713 cm^{-1} which has been assigned to C-H bond stretching , 2256 cm^{-1} and 2173 cm^{-1} has been attributed to C=C bond stretching (alkynes). The absorption peaks at 1103 cm^{-1} has been attributed to C-O bond stretching when compared to the untreated PP. Furthermore , on inoculation of the UV treated PP with *Pseudomonas sp.*, and *Actinomycetes sp.*, certain remarkable chemical changes were evident (Fig-12). The absorption band observed at 3144 cm^{-1} ,2840 cm^{-1}, 2617 cm^{-1} ,1456 cm^{-1},1304 cm^{-1} and 940 cm^{-1}, reflect C-H bond stretching, 2582 cm^{-1} to O-H bond stretching and 2256 cm^{-1} and 2173 to C=C bond stretching .

The structural difference revealed by FTIR spectra of HNO_3 treated PP is presented in fig-13 . In comparison to the FTIR spectra of untreated PP flakes, appearance of new absorption peaks was evinced . Absorption intensity at 3404 cm^{-1} which has been attributed to O-H bond stretching and 2921 cm^{-1}, 2840 $cm^{-1}$and 2345 cm^{-1} to C-H bond stretching (CH_2 or CH_3), 1708 cm^{-1}, 1560 cm^{-1} to C=O bond stretching , 731 cm^{-1} to C-H bond stretching . The FTIR spectrum of HNO_3 treated PP inoculated with *Pseudomonas sp.*, and *Actinomycetes sp.*, displayed a number of new group formation at 2922 cm^{-1}, 2840 cm^{-1} and 2345 cm^{-1} , which has been attributed C-H bond stretching , 1708 cm^{-1} ,1560 cm^{-1} , and 1132 cm^{-1} to C=O bond stretching ,1459 cm^{-1} ,1377 cm^{-1},1279 cm^{-1} , 781 cm^{-1} , and 730 cm^{-1}. which has been attributed to C-H bond stretching (methylene)(fig-14) On further inoculation of HNO_3 treated PP flakes with *Pseudomonas sp.*, and *Actinomycetes sp.*, new peaks were evinced in the FTIR spectra, which is depicted in Fig-14 .These peaks include 1279cm^{-1} and 781cm^{-1} (C-H bond stretching) and 1132cm^{-1} (C=O stretching).

It can be seen from fig-15 that new absorption peaks appeared at 2926 cm^{-1} , 2369 cm^{-1}, 2345 cm^{-1}, 900 cm^{-1} , 773 cm^{-1} which correspond to C-H bond stretching, 1685 cm^{-1} and 1526 cm^{-1} to C=C and C=O bond stretching, respectively. FTIR spectrum of *Pseudomonas sp.*, and *Actinomycetes sp*., inoculated in heat treated PP exhibited absorption peaks at 3422 cm^{-1} , 3199 cm^{-1} and 2345 cm^{-1} , which has been assigned to O-H bond stretching , 2273 cm^{-1} to C-H bond stretching, 1654 cm^{-1} , and 1523 cm^{-1} to C=O bond stretching, 1256 cm^{-1}, 1219cm^{-1},1103 cm^{-1} 1044 cm^{-1} to C-O bond stretching, 940 cm^{-1} and 776 cm^{-1} to C-H bond stretching . New absorption bands were evinced between the FTIR spectra of heat treated PP and *Pseudomonas sp.*, and *Actinomycetes sp*., inoculated heat treated PP. Absorption peaks at 2724 cm^{-1} and 2583 reveal C-H and O-H bond stretching , respectively. The absorption peaks at 1256 cm^{-1}, 1219 cm^{-1} ,1103 cm^{-1} and 1044 cm^{-1} depict C-O band stretching (fig-16). Appearance of many new spectral absorption frequencies were visualised in the FTIR spectra of heat ,UV, and HNO_3 treated PP when compared to the control (Fig-17). Absorption peaks at 3771 cm^{-1} , 3691cm^{-1}, 3677cm^{-1} and 3651cm^{-1}has been attributed to O-H bond stretching (hydroxyl group), 2962 cm^{-1}, 2839 cm^{-1}, 2345 cm^{-1}, 1459 cm^{-1}, and 1377 $cm^{-1}$and 782 cm^{-1} and 732 cm^{-1}, to C-H bond stretching, 1693 cm^{-1}, and 1561 cm^{-1}, due to C=O bond stretching (ester group formation), 1282 cm^{-1} due to aromatic ether formation (C=O). In comparison to the control, FTIR spectrum of *Pseudomonas sp.*, and *Actinomycetes sp*., inoculated with heat, UV and HNO_3 treated PP exhibited absorption peaks at 3403cm^{-1} , due to O-H bond stretching (hydroxyl group), 2921 cm^{-1},1458 cm^{-1},1376 cm^{-1},1130 cm^{-1},972 cm^{-1},841 cm^{-1},781 cm^{-1}, and 728 cm^{-1} which has been assigned to C-H bond stretching, 1718 cm^{-1} due to ester group formation (C=O) ,1277 cm^{-1} which has been attributed to C-O bond stretching (ether group formation) (Fig 18) .On Further inoculation of the heat , UV and HNO_3 treated and UV treated with *Pseudomonas sp.*, and *Actinomycetes sp*., new peaks were evident at 3403 cm^{-1} (O-H bond stretching), 2921 cm^{-1} and 1130 cm^{-1} (C-H bond stretching), 1718 cm^{-1} and 1277 cm^{-1} (C-O bond stretching).

Fig -10 FTIR spectra of untreated Polypropylene flakes

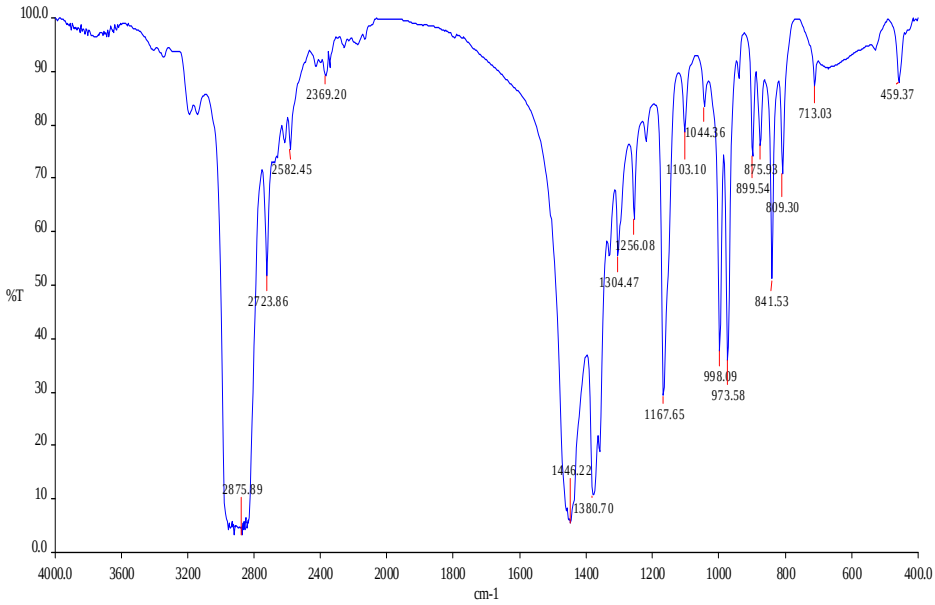


Fig -11 FTIR spectra of UV treated polypropylene flakes

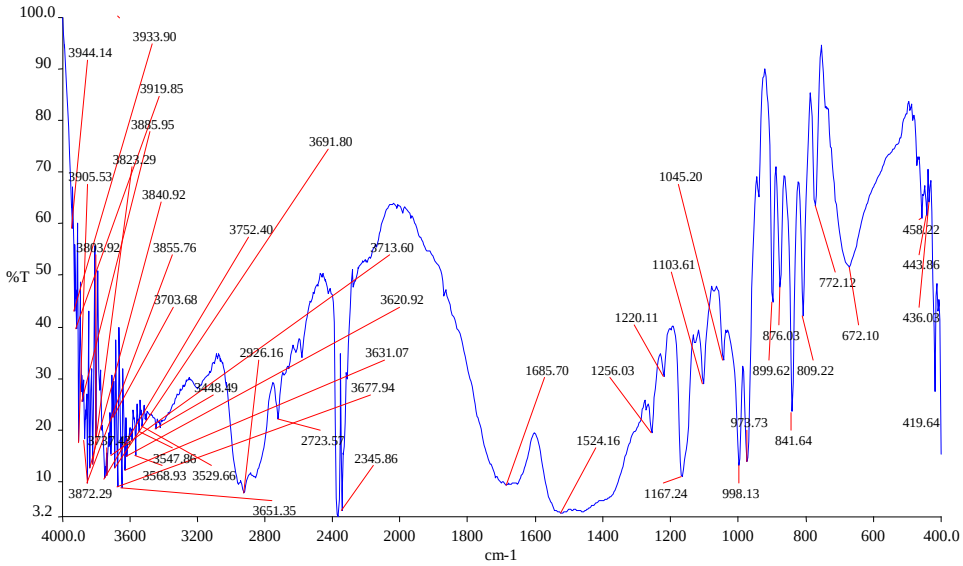


Fig 12: FTIR spectra of UV treated PP flakes inoculated with *Pseudomonas sp.*, and *Actinomycetes sp.*,

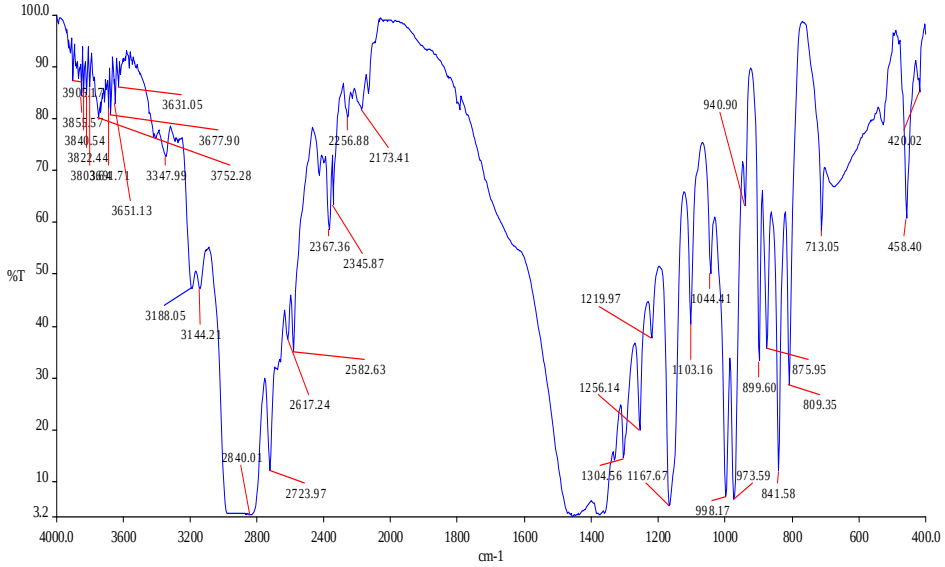


Fig -13: FTIR spectra of HNO_3 treated PP flakes

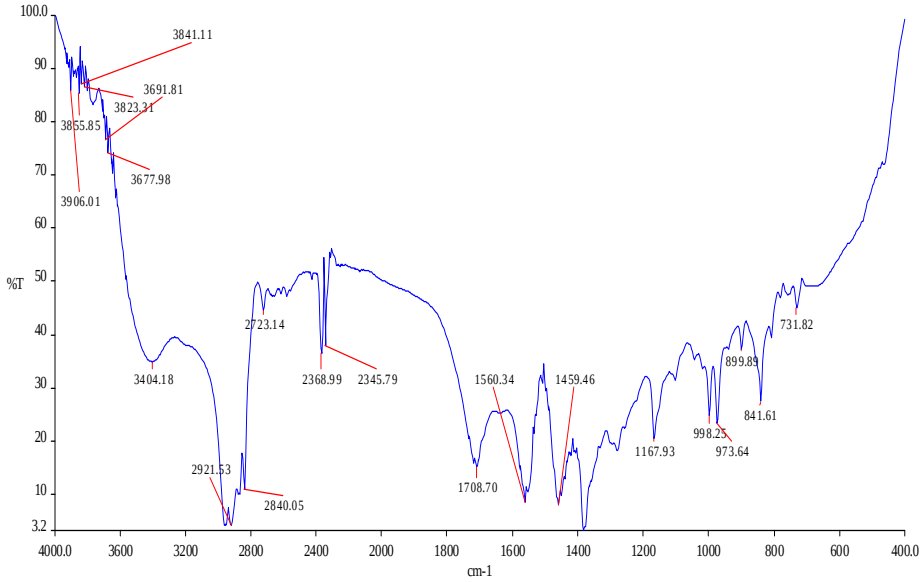


Fig -14: FTIR spectra of HNO_3 treated PP flakes inoculated with *Pseudomonas sp. and Actinomycetes sp.,*

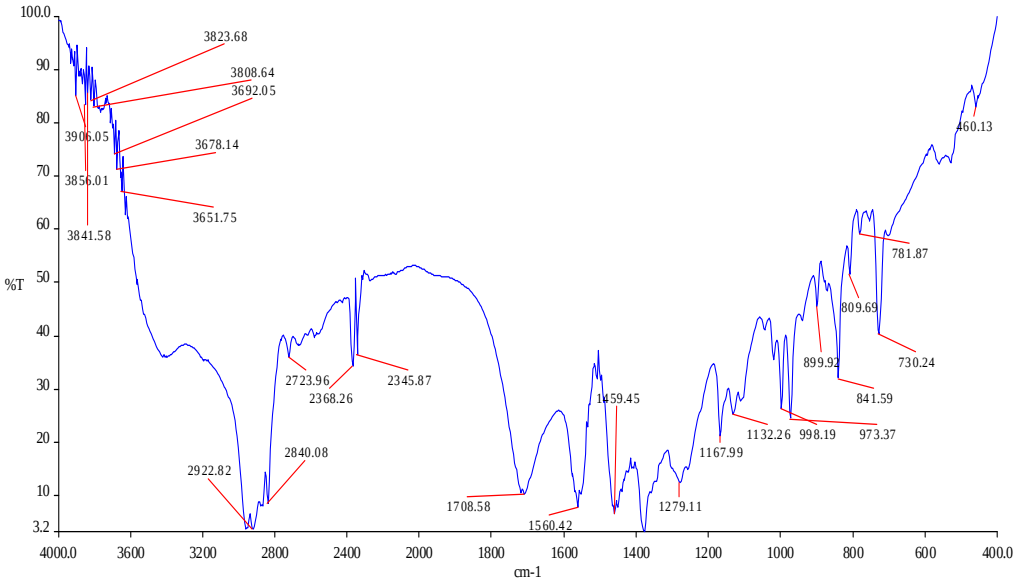


Fig -15: FTIR spectra of Heat treated PP flakes

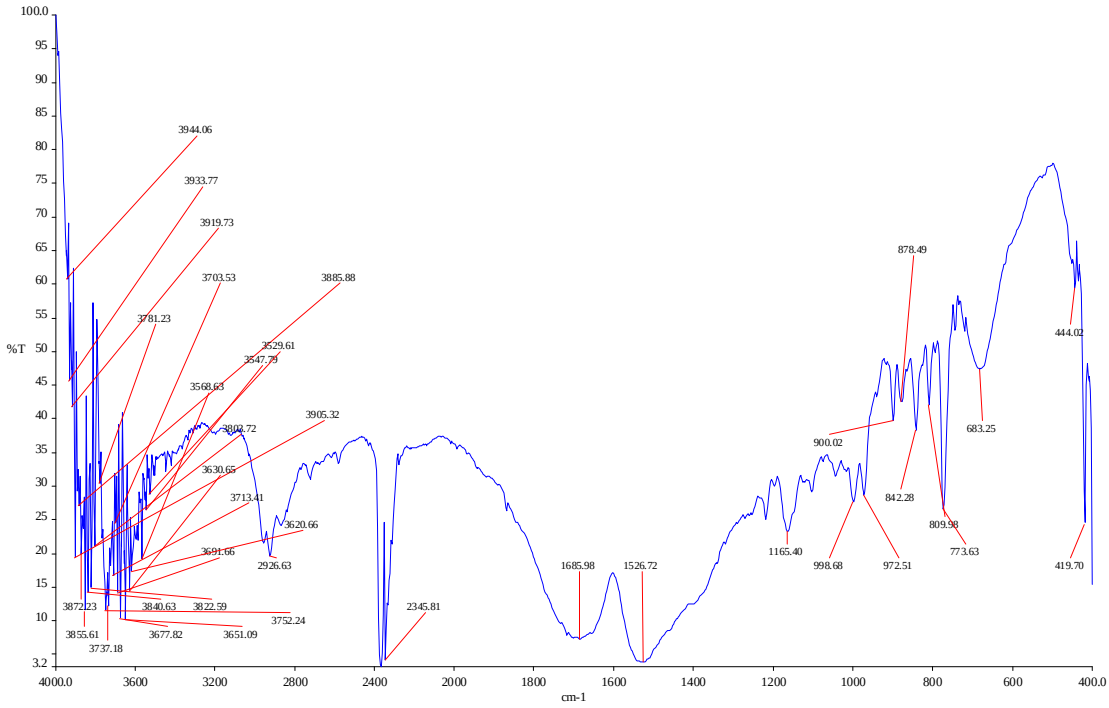


Fig -16: FTIR spectra of heat treated PP flakes inoculated with *Pseudomonas sp., and Actinomycetes sp.*

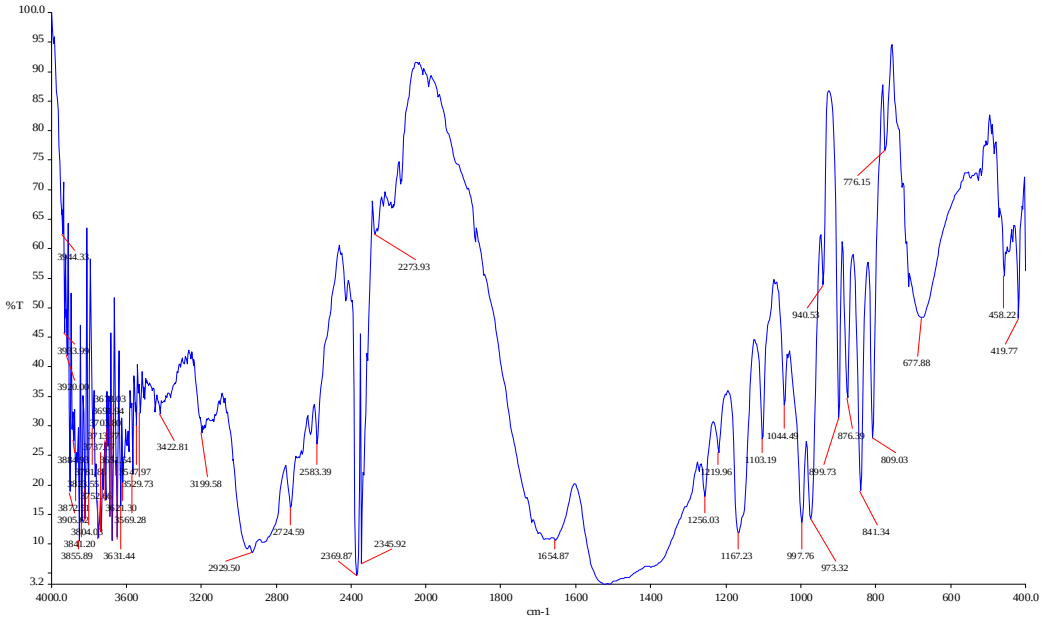


Fig -17: FTIR spectra of UV , HNO_3 and heat treated polypropylene flakes

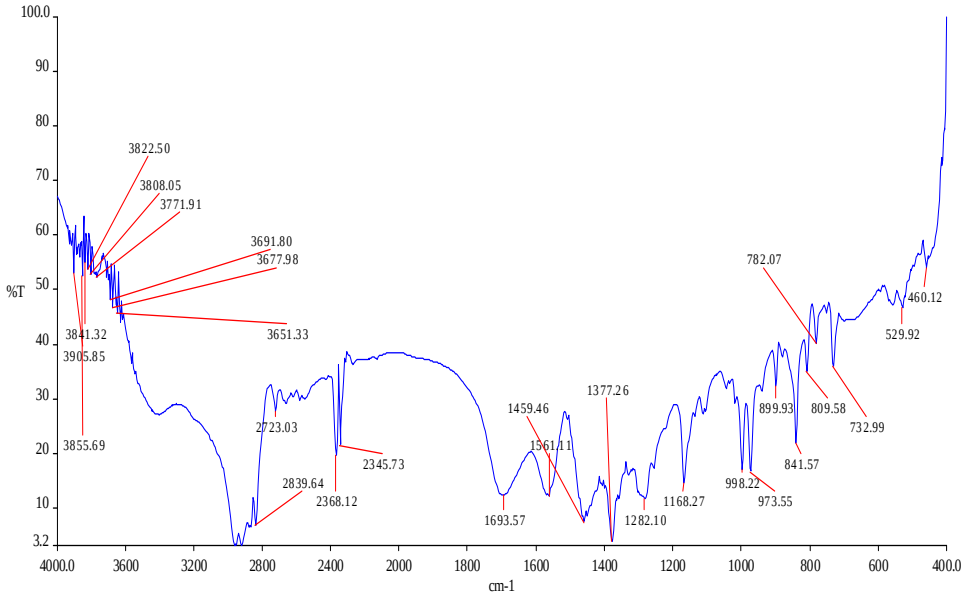
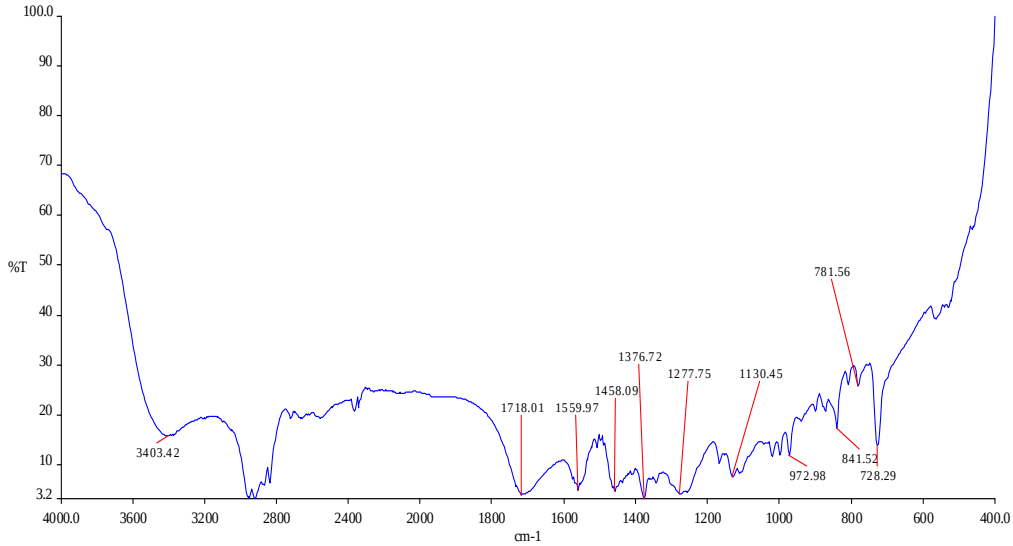


Fig 18 : FTIR spectra of UV , HNO_3 and heat treated PP flakes inoculated with *Pseudomonas sp. and Actinomycetes sp.,*



DISCUSSION

The non-degraded PP is smooth, without fracture and free from defects . The smooth surface of UV treated PP observed in this study is contradictory to the findings of Santhoshkumar et al., who have noticed development of some fracture and grooves on the PP surface due to UV exposure [10]. They have further observed that UV exposed PP showed rapid disintegration of the surface for 15 and 45 days. Similar to the present result, they have also observed typical patterns of microbial growth (*Aspergillus niger* and *Penicillium funculosum*) on the top surface of PP . In addition, they have noticed erosion of the surface and found that microorganism grows fissure resulting from fracture propagation, none grow within the fracture suggesting that the most of the low molecular weight nutrient migrate to the surface from the (oxidized) layer of the polymer. Further, they have observed surface agglomerations and have attributed it to biodegradation involving deterioration of molecular chains [10]. The presence of colonies of *Pseudomonas sp.* and *Actinomycetes sp.*, on the PP surface is well supported by Ambika Arkatkar et al ., who have observed that the attachment of *Pseudomonas azotoformans,* and *Bacillus subtilis* is considerably higher than the *Bacillus flexus and Pseudomonas stutzeri* and have ascribed it to the production of biosurfactant, making the PP films relatively more hydrophilic. The surface of UV treated PP is smooth without fracture and free from imperfection except the presence of *Pseudomonas sp.* and *Actinomycetes sp* ., colonies [11]. This observation is not in good accord with the findings of Santhoskumar and Palanivelu who have detected fractured surface of PP films after UV exposure in the SEM image [12]. As evinced in the SEM image of untreated PP, Favaro et al ., also observed flat and clean surface of untreated PP film under SEM .They also noticed increased surface roughness and defects in several regions of PP surface due to sequence of oxidative treatments . Furthermore, they observed white point on the $KMnO_4$ / HCl treated PP surface and have ascribed it to the residues produced in the oxidative process such as MnO_2[13].

Similar results were found by Grisa and Zeni in a study on PP degradation in the sanitary landfill , cell C_3 at 9 m depth. Surface colonization of *Pseudomonas sp.* and *Actinomycetes sp.*, on the surface of UV and HNO_3 treated PP could be a cause for the degradation for PP [14] . According to Arkatkar, the ability of a microorganism to utilize any substrate depends on its growth and its adherence to form a biofilms [11] . This observation also gain supports from the findings of Carina Longo. who have viewed through optical micrography on disposed PP the adherence of microorganisms visible as chromatic alteration and surface rupture of the polymeric material [15]. According to Fleming, the color spectrum observed in the cracks and in the colony of microorganisms in caused by excretion of microbial lipophylic pigments , bringing evidence of biodegradation process in the polymer [16].

The absorption peaks observed in the FTIR spectra of PP coincides with the observation of Santhoshkumar et al., who have reported absorption bands at 1167 cm^{-1} , 997 cm^{-1} and 973 cm^{-1}which correspond to C-H bond stretching. During pretreatment, polypropylene is also reported to undergo oxidation due to generation of radicals that further propagate to form peroxides. There are three carbon atoms in the PP chain where radicals can form namely, at primary (methyl group), secondary (in the chain, not attached to methyl group) and/or tertiary position (in the chain to which methyl group is attached) [10]. Ambika Arkatkar et al. and Gijsman and Hennekens , have proposed that there can be probably three paths that lead to the formation of peracid. The first path is the formation

of methyl radical due to the oxidation at the primary carbon. The second path is the formation of primary alkyl radical formed due to intermolecular hydrogen abstraction of tertiary alkoxy radical. The third path can be the formation of peracid from the oxidation of oxidized products[11, 17]. The peroxides formed initially decompose at a slower rate due to the restricted mobility of the polymer, but the primary oxidation products formed, lead to the formation of fast decomposing peroxides such as peracids [18, 17]. After the induction stage, decomposition of these peracids plays a major role in determining the rate of oxidation. These decomposition reactions produce ketones and esters. These oxidized products are analysed through FTIR analysis. The 1700 cm^{-1} -1800 cm^{-1} region in FTIR spectra indicates the presence of oxidized groups [11] .Peaks at 1708 cm^{-1} and 1178 cm^{-1} corresponding to the formation of keto carboxyl group were observed after pretreatment with (HNO_3 , HNO_3 + *Pseudomonas sp.* and *Actinimyctes sp*.) and (UV , Heat + HNO_3 + *Pseudomonas sp.* and *Actinimyctes sp*.) ,respectively indicating oxidation of the polymer. Similar peaks were also displayed (1715 cm^{-1} and 1748 cm^{-1}) in the FTIR spectra of pretreated PP at the end 12 month [11]. Our results gain further support from the reports of Ambika Arkatkar et al ., who have observed that *Pseudomonas azotoformans*, *Pseudomonas stutzeri* and *Bacillus subtilis* biotically degrade PP SUV (Short UV pretretment) at the end of the 12 month experiment . No such changes were observed in the positive control (without *Pseudomonas sp.* and *Actinimyctes sp* .,) indicating that changes observed in the carbonyl group after 12 month were entirely due to microorganisms [11].

Our observations are also in agreement with the findings of Albertsson et al. and Sudhakar et al. who reported that in the case of low and high density polyethylene the carbonyl and ester groups initially increased and then decreased during the course of the biotic process [19 ,20]. Several reports on polyethylene mention an increase in carbonyl group when exposed to abiotic environment and decrease in carbonyl group when exposed to biotic environment [21,24,25,4,2] .

During thermal decomposition of an aliphatic polyester , several byproducts are formed such as gaseous , liquids with different boiling points and heavy non-volatile products [26]. The characteristic peaks recorded at 1000 -1400 cm^{-1} and 2850-3050 cm^{-1} indicating formation of hydrocarbon in both the treated and untreated PP coincides with the observations of Chrissafis et al ., who have noticed these peaks in the volatile products released during thermal decomposition of poly (Propylene sebacate) (PPSeb) and poly(propylene azelate) PPAB [26].

The peaks in the range of 1650 -8500 cm^{-1} observed in the UV (1685 cm^{-1}), HNO_3(1708 cm^{-1}), HNO_3 + *Pseudomonas sp.* and *Actinomycetes sp*., (1708 cm^{-1}), Heat (1685 cm^{-1}), Heat + *Pseudomonas sp.* and *Actinomycetes sp* . (1654 cm^{-1}) , HNO_3+UV (1693cm^{-1}) and HNO_3+ Heat + UV+ *Pseudomonas sp.* and *Actinomycetes sp*. (1718 cm^{-1}) treated PP is in good accord with the observations of Chrissafis et al ., who have reported these peak (1685 -1800 cm^{-1}) in the FTIR spectra of volatile products obtained during thermal decomposition of PPAz and PPSeb and have attributed it to the formation of carbonyl compounds (acids, ketones, aldehydes , anhydrides, etc.). They have also observed absorption peaks at 2250 cm^{-1}-2400 cm^{-1} and have reasoned out that it is due to the formation of CO_2 and CO. These absorption peaks were evident in the present study except, in the PP exposed to HNO_3 +Heat +UV+ *Pseudomonas sp.* and *Actinomycetes sp*. Vrigin PP elicted peak at 2369 cm^{-1} , UV treated PP at 2369 cm^{-1} and 2345 cm^{-1},UV and *Pseudomonas sp.* and *Actinomycetes sp* ., at 2367 cm^{-1}, 2345 $cm^{-1}$and 2256 cm^{-1}, HNO_3 at 2368 cm^{-1} and 2345 cm^{-1}, HNO_3 + *Pseudomonas sp.* and *Actinomycetes sp* ., at 2369 cm^{-1} and 2345 cm^{-1}, Heat at 2369 cm^{-1} and 2345 cm^{-1}, Heat + *Pseudomonas sp.* and *Actinomycetes sp* ., at 2369 cm^{-1} , 2345 cm^{-1} and 2273 cm^{-1}, and HNO_3 +Heat + UV *at* 2368 cm^{-1} and 2345 cm^{-1} . They have further stated that the strong absorption band at 1711cm^{-1} is an indication that most of the produced byproducts of thermal decomposition of PPAz and PPSeb have carboxylic acids .This explanation could be extended to the present observation (HNO_3 treated PP: 1708 cm^{-1}; HNO_3 + *Pseudomonas sp.* and *Actinimyctes sp* .,treated PP 1708 cm^{-1} ; HNO_3 +UV+ Heat + *Pseudomonas sp.* and *Actinomycetes sp* .: 1718 cm^{-1}). Carbonyl end groups and vinyl groups are formed during decomposition of aliphatic polyesters via β –hydrogen bond scission , which is the main decomposition mechanism [26].

The present result also agrees with Adurafimihan Abiona and Gabrid Osinkolu who have reported that Gamma radiation induced remarkable changes in the physico- chemical properties of the material due to chain scission , oxygen effects and cross linking activities , which resulted in the production of water vapour, carbon monoxide from hydroxyl and carboxyl groups. Further, they noticed that the melting temperature and crystallinty of polypropylene reduce as the gamma radiation dose increases which they confirmed by Raman and UV visible spectroscopic analysis . In addition they have also observed reduction in elongation to fracture and reduction in tensile strength of the materials as dose increases [27]. Chase et al ., have reported that the volatile peroxides may be produced from the products of the photolysis of hydro peroxides formed in the polypropylene during oxidation [28]. Appearance of new absorption peaks at 1560 cm^{-1} , 1708 cm^{-1}, 3404 cm^{-1}in the HNO_3 treated PP lies in paralled to the findings of Favaro et al ., who have also observed development of distinct absorption bands at 1550 $cm^{-1}$and 1730 cm^{-1} and have reasoned out that it could be due to COO [29] and C=O [30] stretching

vibration in the FTIR –ATR spectra of PP film treated with $KMnO_4$ / Hcl 0.05/0.1 $molL^{-1}$ for 8h at 45 ^0C. while at $KMnO_4$ / Hcl 0.25/ 0.5 $molL^{-1}$ intensification of the band at 1730 cm^{-1} with the appearance of a broad band around 3200 cm^{-1} and have attributed it to O-H stretching vibrations and a large shoulder at 1080 cm^{-1} generated by C-O stretching vibration and have suggested that the oxidation of these films increased highly with increase in the concentration of the $KMNO_4$ solution. The FTIR spectra of the present study displayed absorption peak at 1524 cm^{-1} (UV treated PP), 1526 cm^{-1} (Heat treated PP),3199 cm^{-1} and 1523 cm^{-1} (Heat + *Pseudomonas sp.* and *Actinomycetes sp* ., treated PP) ,1561 cm^{-1} (HNO_3 + Heat +UV treated PP) and 1718 cm^{-1} (HNO_3 + Heat +UV treated *Pseudomonas sp.* and *Actinomycetes sp* ., PP) which coincides with the observation of Favaro et al [13].

The presence of carbonyl group in the FTIR spectra of UV treated PP, (1685 cm^{-1}) HNO_3 treated PP (1708 cm^{-1}), HNO_3 treated PP+ *Pseudomonas sp.* and *Actinomycetes sp* ., (1703 cm^{-1}), HNO_3 + Heat + UV treated (1693 cm^{-1}) and HNO_3 + Heat + UV *Pseudomonas sp.* and *Actinomycetes sp* ., treated PP (1718 cm^{-1}) corroborates with the findings of Carina Longo et al ., who have also detected the carbonyl group at 1722 cm^{-1} in the FTIR spectra of buried PP and have associated it to the degrading mechanism. Carina Longo et al., have stated that the incidence of absorbance measured in the regions 872 cm^{-1}, 741 cm^{-1},695 cm^{-1} and 614 cm^{-1} may possibly signal changes in crystallinity of buried PP [16] .

The most important degradative mechanism associated with majority of synthetic macromolecules, used as plastics and synthetic fibers are associated with the absorption of UV light . Polymer like polyethylene , polypropylene and polystyrene tend to embrittle under the action of UV light which could be caused by one or a combination of three effects : (1) scission of the main chain (2) Photo induced crystallization , (3) cross linking [31].The present observation correlates with that of Cacciari et al ., who have found that aerobic and anaerobic species of microbes with different catabolic capabilities can act in close cooperation to degrade polypropylene films [32]. We have also observed that *Pseudomonas sp., Penicillium sp, Aspergillus sp* could play a vital role in the acceleration of degradation process of PET (33,34,35)

CONCLUSION

The huge amount of synthetic polymers like polyethylene and polypropylene waste released into the environment is of global concern. Hence methods to convert these polymers into lower weight intermediates have to be devised. The present study focuses on biotic (*Pseudomonas sp.* and *Actinomycetes sp* .,) treatments of abiotically (UV, Heat ,HNO_3) treated PP flakes resulting in several molecular changes which is gauged through SEM analysis and FTIR studies SEM images reveal micromorphological changes likes formation of cracks and granules , roughness , irregularities and unevenness of the PP surface on exposure to HNO_3, Heat, *Pseudomonas sp.* and *Actinomycetes sp* ., . The surface colonisation the inoculated *Pseudomonas sp.* and *Actinomycetes sp* ., was evident in the SEM images . The formation of methylene group, ester group, alkynes carbonyl group were evident the FTIR spectra of abiotically and biotically treated PP .

Acknowledgement
The authors register their thanks to the University Grants Commission, New Delhi, India for providing financial assistance for conducting this research work.

REFERENCES

[1] LD Suits , YG Hsuan , *Geotextiles and Geomembranes*, **2003**,21, 111-122.
[2] D Hadad , S Geresh , A Sivan , *Journal of Applied Microbiology*,**2005**, 98,1093- 1100.
[3] A Arkatkar , J Arutchelvi , M Sudhakar , S Bhaduri , PV Uppara , M Doble ,*The Open Environmental Engineering Journal* , **2009**, 2, 68-80.
[4] M Iring , F Tudos , *Progress in Polymer Science*,**1990** ,15, 217-262.
[5] AG Graeme ,C Mathew , Homogeneous and heterogeneous oxidation of polypropylene. In: Hamid, S.H. (Ed.), Handbook of polymer degradation. Marcel Dekker, New York, **2000**, 277-313.
[6] M Sudhakar , M Doble , M P Sriyutha , R Venkatesan, *International Biodeterioration and Biodegradation*, **2008,** 61,203-213.
[7] I Gilan, Y Hadar , A Sivan, *Applied Microbiology and Biotechnology* **2004**, 65, 97-104.
[8] PHA Sneath ,SN Mair ,M Elisabeth Sharp , GJ Holt . Bergeys Manual of systematic Bacteriology, Williams ailkines, Baltimore, USA.**1994.**
[9] Y Magdy ,R Abdelaal Tariq ,S Sobahi Mohamed ,I Makki , *Int J Polymer Material*, **2008**, 57,73-80.
[10] AU Santhoskumar , K Palanivelu , SK Sharma , SK Nayak, *J Bioremed Biodegrad*, **2010,** 1,108,2155-6199.

[11] A Arkatkar, J Arutchelvi,S Bhaduri ,PV Uppara , M Doble. *International Biodeterioration & Biodegradation*, **2010**,64, 530-536.
[12] AU Santhoskumar, Palanivelu, *Research journal of Pharmaceuticla and Biological and Chemical science*, **2011**, 2(3), 299.
[13] SL Favaro , AF Rubira , EC Muniz , E Radovanovic . *Polymer Degradation and Stability*, **2009,** 92, 1219-1226.
[14] AMC Grisa ,C Longo ,M Zeni , Caracterizaco de filmes de poli(cloreto de vinila) biodegradadosem aterro sanitaria II . In : Anais do simposio Latino Americano , IX Congresso Iberoamericano de Polimeros; Lima, per.Lima: pontificia Universided Catolica del Per ; **2008**.
[15] L Carina , S Michele , Z Mara ,NB Rosmary , Ana Maria Coulon Grisa, .*Mat. Res*, **2011**.
[16] HC Fleming .*Polymer Degradation and Stability* , **1998,** 59, 309-15.
[17] P Gijsman , J Hennekens , *Polymer Degradation and Stability*, **1993**, 42, 95-105.
[18] JL Bolland ,G Gee, *Transactions of the Faraday Society* ,**2010**, 42, 236-252.
[19] AC Albertsson, SO Andersson , S Karlsson , *Polymer Degradation and Stability*, **1987**,18, 73-87.
[20] M Sudhakar , M Doble , MP Sriyutha , R Venkatesan , *International Biodeterioration and Biodegradation*, **2008,** 61, 203-213.
[21] B Dolezel, *British Journal of Plastic Surgery*, **1967**,49, 105-113.
[22] AC Albertsson ,S Karlsson , *Progress in Polymer Science*, **1990**,15, 177-192.
[23] AC Albertson ,C Barensted, S Karlsson ,T Lindberg . *Polymer*, **1995**,36, 3075-3083.
[24] M Weiland , A Daro ,C David .*Polymer Degradation and Stability* ,**1995**,48, 275-289.
[25] E Chiellini , A Corti ,G Swift , *Polymer Degradation and Stability*,**2003**, 81,341-351.
[26] K Chrissafis, KM Paraskevopoulos , GZ Papageorgiou , D N Bikiaris, *Journal of Analytical and Applied Pyrolysis* , **2011**,92, 123-130 .
[27] A Adurafimihan Abiona , A Gabriel Osinkolu , *International Journal of Physical Sciences*, **2010**, 5(7) , 960-967.
[28] H Chase, Butler, M Paul. *Polymer Degradation and Stability*, **2013**, 98, 471-473.
[29] M Muller, T Rieser, K Lunkwitz , S Berwald , J Meier –Haack , D Jehnichen,. *Macromol Rapid Commun*, **1998**, 19(7), 333-6.
[30] NV Bhat , DJ Upadhyay , *J Appl Polym Sci* , **2002** , 86(4),925-36.
[31] JE Guillet, *Pure and Applied chem*, **1972**, 30, 1(2), 135-144.
[32] I Cacciari ,P Quatrini ,G Zirletta,E Mincione , V Vinciguerra ,P Lupattelli ,G Giovannozzi Sermanni. *Appli and Envi Microbiology* , **1993**,59,(11),3695-3700.
[33] S Umamaheswari , M Murali, A Anbusaravanan. *J. Microbiol. Biotech. Res.*, **2013**, 3 (1)104-110
[34] S Umamaheswari , M Murali, P Ilayaraja. *J. Microbiol. Biotech. Res*, **2013**, 3 (4):47-53
[35] S Umamaheswari , M Murali, *Elixir Chem. Engg*.**2013,** 64,19159-19164.