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## **RESEARCH ARTICLE**

## **BIODEGRADATION OF POLY (ETHYLENE TEREPHTHALATE) BY BACILLUS SUBTILIS**

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ARTICLE INFO	ABSTRACT
<i>Article History:</i> Received 25 <sup>th</sup> , September 2015 Received in revised form 26 <sup>th</sup> , October 2015 Accepted 29 <sup>th</sup> , November 2015 Published online 30 <sup>th</sup> , December 2015	With the rapid development of poly (ethylene terephthalate) (PET) and its wide use in industry, there is a substantial fraction by volume added to the waste stream every year, which causes an environmental issue, since PET is highly resistant to atmospheric and biological agents. Currently, the handling methods of PET and other polymer wastes involve burying, burning, and recycling. But, as regards environmental protection, these methods have many shortcomings and cannot address poly (ethylene terephthalate) PET waste pollution from the source. Although the biodegradation ratio of PET was still weak, we demonstrated with SEM micrographs and IR analysis that Bacillus subtiliscould act on the PET.
Keywords:	
Biodegradation, PET, <i>Baccilus Subtulis</i> ,	

## **INTRODUCTION**

Polymer.

Polymeric materials have gained a wide influence due to their excellent mechanical and thermal properties and high stability. They are very unique in chemical composition, physical forms, mechanical properties and applications. Because of this structural versatility, polymeric materials are widely used in aerospace applications, aviation and space industries as paints, plastics, composites, rubbers, lubricants, etc adhesives. (Vassiliou et al., 2010). Environmental pollution by Polymeric materials, such as waste plastics, has been recognized as a large problem. In order to support continued sustainable development throughout the world, this problem must be addressed. In view of this, the biodegradation of plastics has been studied extensively for the past three decades. Some types of plastic have been shown to be biodegradable, and their degradation mechanisms have progressively become clearer (Masayuki, 2001). The biodegradation has the potential to avoid secondary pollution, while lowering the handling cost. work has chosen as polymer This poly(ethylene terephthalate). With the rapid development of poly(ethylene terephthalate) (PET) and its wide use in industry, there is a substantial fraction by volume added to the waste stream every vear, which causes an environmental issue, since PET is highly resistant to atmospheric and biological agents.

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Laboratory of Chemistry of Materials and Biotechnology natural products, University of Sciences Moulay Ismail, BP 11201, 50000Meknès, Morocco. Therefore, PET is a noxious material from a global environmental and ecological standpoint (Edge *et al.*, 1991; Allen *et al.*, 1994; Rudakova *et al.*, 1979).

### **MATERIALS AND METHODS**

**Polymer studied**: polyester is a polymer (a chain of repeating units) where the individual units are held together by ester linkages. The specific polyester used in this work is polyethylene tetrephtelete (Fig 1).



# Fig. 1. The structure of its monomer (n: is the number of repetitions)

**Organism**: *Bacillus subtilis* was isolated, identified and obtained from the Laboratory of Microbial Biotechnology of Faculty of Science and Technology, Fez, by its ability to degrade a many polymers.

**Degradation of Polyurethanes test, media and culture conditions**: Luria-Bertani (LB) medium was prepared by adding 0.5g yeast extract, 10.NaCl, and 1.0g tryptone to 100 ml dH2O. PET sample was also added to the medium.

The test of biodegradation was incubated 30 days at 37°C IR analysis of polyurethane degradation

Scanning electron microscopy: SEM was used to characterize surface modifications of PET samples. Images of experimental surfaces showed decomposition and penetration of bacterium on the PET samples.

Infra red: IR spectra of the PET samples were analyzed using an IR spectrometer VERTEX 70, after 30 days of incubation.

### **RESULTS AND DISCUSSION**

#### Surface morphology after biodegradation

The environmental scanning electron microscopy analysis before and after degradation of PET. The results of the effect of bacterial degradation on PET surface are presented in Fig 2. According to these findings, the surface of the sample control of PET is opaque and very smooth (Fig 2.A), compared to the samples the samples which have subjected to the treatment by bacterial solutions. Indeed, the samples of PET polymer obtained after the immersion in the bacterial medium of Bacillus subtilis showed the whitish cracks formation on its surface (Fig 2.B).



Fig. 2. (A) SEM images of PET before microbiological testing (B) SEM images of PET treated with Bacillus subtilis

From these results given by ESEM, it is found and noted that Bacillus subtilis strain has shown a significant interaction and effect on the PET surface producing low morphological heterogeneity and signs of erosion which show his ability to react and degrade the PET,.

These results are consistent with a study of biodegradation of PET fibers and which also showed low degradation of this polymer (Jianfei et al., 2004). Many researchers and institutes are engaged in the study of biodegradation of PET (Kohei, 2002; Huang et al., 1980; Yoon et al., 2001) but these studies are still in the primary stage due to the compact chemical structure of this polymer. PET spectrum is characterized by the presence of characteristic absorption bands of functions: (Fig 3A)



Fig. 3. (A): infrared spectrum of PET before test of biodegradation (B): infrared spectrum of PET after test of biodegradation

- Esters: 1050-1300 cm<sup>-1</sup> (deformation  $\delta_{C-O}$ ), 1720 to 1730 cm<sup>-1</sup> (elongation  $v_{C-O}$ ), 1240 cm<sup>-1</sup> (deformation  $\delta_{O-CO}$ );
- Aromatic at 1500-1600 cm<sup>-1</sup> (deformation  $\delta_{C=C}$ ) 3020 cm<sup>-1</sup>
- (elongation v  $_{\text{H-C}});$  lkyl at 1450 cm  $^{-1}$  (v\_{CH2} elongation) and 715-750 cm  $^{-1}$ (deformation  $\delta_{CH2}$ ).

The spectrum of the PET after biodegradation by Baccilus is characterized by the disappearance of absorption bands located at 2995, 2940, 2950 cm<sup>-1</sup> (elongation  $v_{(-CH-)}$ ), decrease in intensity of the band located at 1711 cm<sup>-1</sup> (ester carbonyl), the appearance of bands at 1339 and 1405 1449 cm<sup>-1</sup> (deformation  $\delta_{(-CH-)}$ , 1240, 1085 and 1014 cm<sup>-1</sup> (elongation v (-CO-)) and 870  $cm^{-1}$  (elongation v (-CC-) (Fig 3B). In this study the degradation of PET by Bacillus has been chemically demonstrated by infrared spectroscopy, which shows A progressive reduction in the relative intensity of the peak carbonyl was observed and was accompanied by more subtle changes at another wave number (Fig 3). In the previous studies on activity of Bacillus subtilis toward different polymers (Nakkabi et al., 2015). Such polyurethane were showed that Bacillus subtilis was able to degrade completely polyurethane.

The loss of this peak is consistent with hydrolysis of the ester bond in the urethane linkage (Nakkabi *et al.*, 2015). The growth of *Bacillus subtilis*, scattered on the surface of PET film, resulted in the emergence of a few small holes after one month (Fig 2A).

These results are the same given by other study who used the fungus *Penicillium funiculosum* (Bozena *et al.*, 2011) The current literature reports some fungi having the ability to degrade polyethylene tetrephtatate. This is the first study that demonstrates a capacity of the degradation of polyetheylen tetraphtalate by *bacillus subtillus*. Many researchers and institutes are engaged in the study of biodegradation of PET (Kohei, 2002; Huang *et al.*, 1980; Yoon *et al.*, 2001) but these studies are still in the primary stage due to the compact chemical structure of this polymer.

### Conclusion

Widespread studies on the biodegradation of plastics have been carried out in order to overcome the environmental problems associated with synthetic plastic waste. Biodegradation of plastics by microorganisms and enzymes seems to be the most effective process to fight against plastic waste.

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