Determination of optimal conditions for the biodegradation of microplastics using bacteria

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UVOD

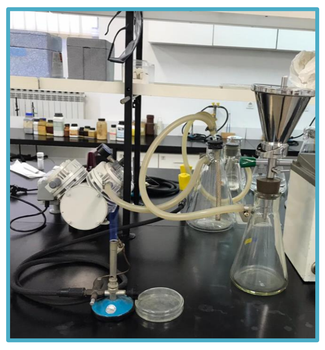
Since 1970, the attention of scientists has been focused on environmental pollution with microplastics. Microplastics are defined as particles smaller than 5 mm in size. They are found in all components of the environment, such as oceans, seas, rivers, lakes, sediment and soil, but also in food. By remaining in the environment and bioaccumulating microplastic particles in aquatic organisms, it represents an ecological risk. In view of the above, it is necessary to investigate procedures for removing microplastics from the environment. A promising procedure is the bioremediation of the polluted environment using bacteria.

MATERIALS AND METHODS

Grinding of plastic

Cultivation of *Bacillus cereus*

Sieving of microplastics



Sterilization of microplastics

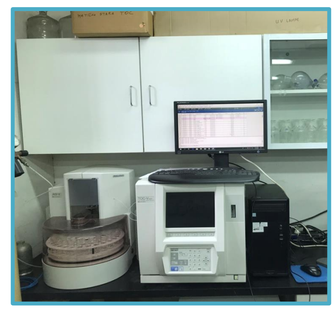
The biodegradation experiment

Parameters and their values that were investigated for PS biodegradation by *Bacillus cereus*.

Parameters	Level 1	Level 2	Level 3
γ_{MP} / mg/L	50	500	1000
size of MP / µm	100-300	300-500	500-700
agitation speed / rpm	100	150	200

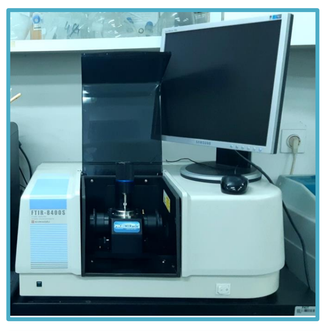
Initial values for PS biodegradation by *Bacillus cereus*.

Parameters	*Bacillus cereus*
$OD_{\lambda = 600\ nm}$	0.30
CFU / st/mL	$9.3 \cdot 10^6$
log CFU / -	6.97
pH-value / -	7.33
$\gamma(O_2)$ / mg/L	8.34
T / °C	24.60



TOC and IC analysis

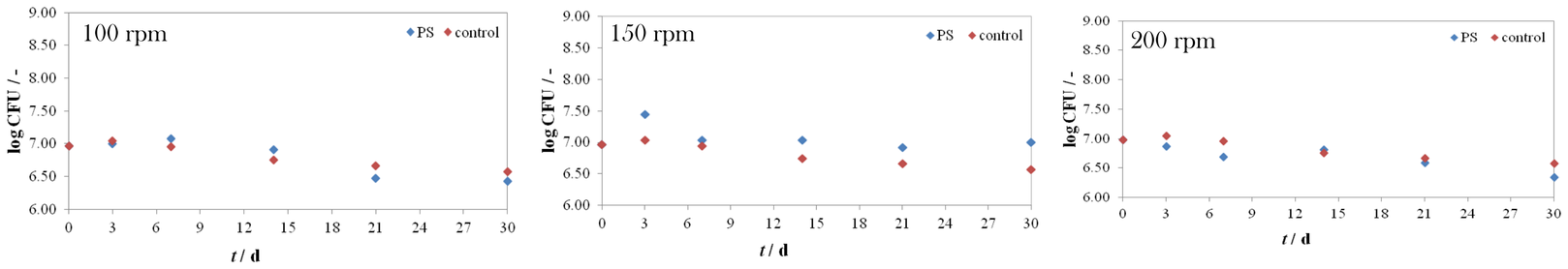
Ionic chromatography



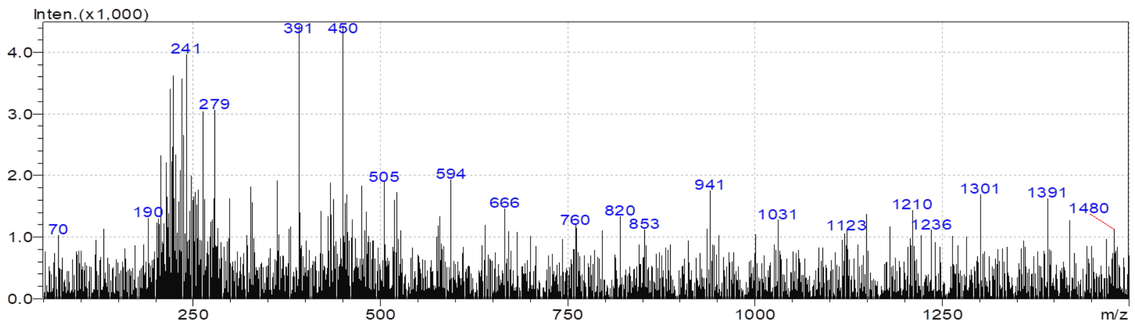
FTIR-ATR spectroscopy

The experiment for biodegradation of PS by *Bacillus cereus* was conducted for 30 days at room temperature. *OD* was constant and it was 0.3.

RESULTS

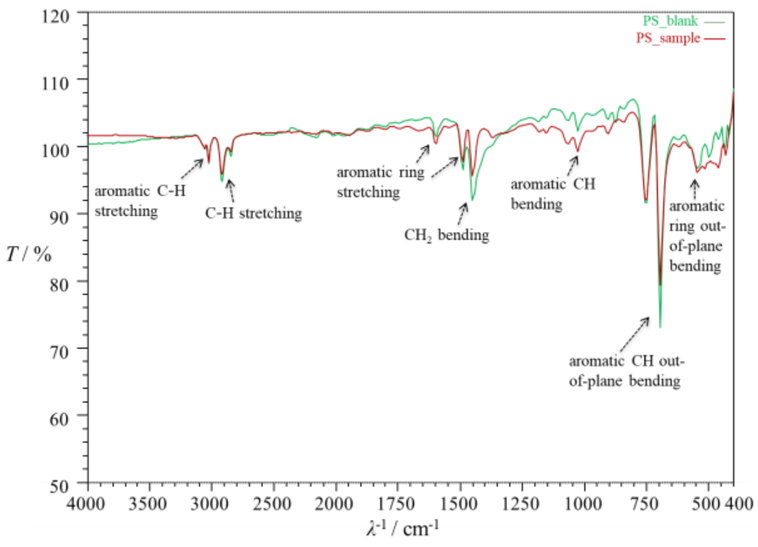


Changes of logarithmic number of living cells (log CFU) of *Bacillus cereus* during 30 days of PS biodegradation by *Bacillus cereus*.



m/z ratio	species
279	plasticiser , phalate ester
391	diisooctyl phthalate, plasticiser

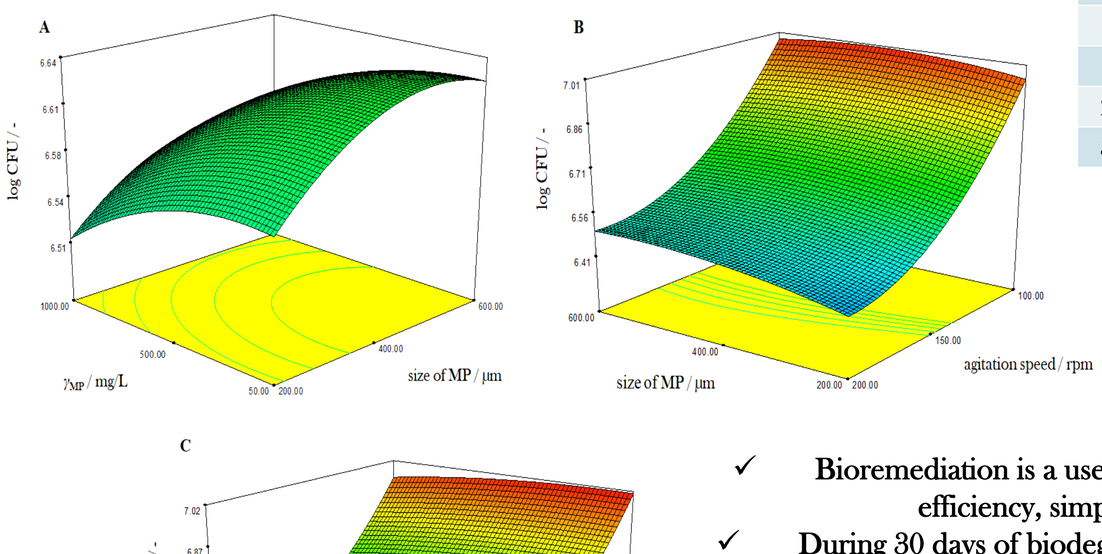
LC/MS analysis of aqueous phase during biodegradation process.



FTIR -ATR spectroscopy of PS particles before and after 30 days of PS biodegradation by *Bacillus cereus*.

Obtained significant factors according to experimental design by Design Expert.

Parameters	Level
γ_{MP} / mg/L	150.65
size of MP / µm	449.87
agitation speed / rpm	100



Response surfaces obtained for the experiment of PS biodegradation by *Bacillus cereus*.

Changes of total carbon (TC), total organic carbon (TOC) and inorganic carbon (TIC) during 30 days of PS biodegradation by *Bacillus cereus*.

t / d	Control			PS		
	TC / mg/L	TOC / mg/L	TIC / mg/L	TC / mg/L	TOC / mg/L	TIC / mg/L
0	3.28	2.51	0.76	3.28	2.51	0.76
3	2.20	2.10	0.10	5.60	3.37	2.23
7	2.29	2.12	0.17	5.02	3.29	1.73
14	3.22	3.13	0.09	4.29	3.42	0.87
21	3.17	3.07	0.10	4.09	3.30	0.79
30	3.01	2.92	0.09	4.89	4.14	0.75

Changes of ions concentrations during 30 days of PS biodegradation by *Bacillus cereus*.

t / d	Control		PS	
	PO_4^{3-} / mg/L	K^+ / mg/L	PO_4^{3-} / mg/L	K^+/ mg/L
0	5.17	3.40	5.17	3.40
3	5.54	3.62	1.47	1.88
7	7.04	3.44	5.51	3.76
14	7.69	3.81	5.68	3.94
21	6.41	3.38	4.67	3.62
30	10.81	4.80	5.05	4.23

CONCLUSIONS

- ✓ Bioremediation is a useful method for water purification because of its efficiency, simple performance and cost-effectiveness.
- ✓ During 30 days of biodegradation experiments, the number of living cells of studied Gram positive bacteria were raising.
- ✓ TOC and IC values had the same trend as a log CFU and indicate on the leaching of additives from microplastics surface and release of CO_2 and biodegradation of PS by bacterium, respectively.
- ✓ By ionic chromatography, phosphate and potassium were the most frequently analyzed as anions and cations, respectively. These ions make the composition of bacterial cells and are important for bacterial homeostasis.
- ✓ By FTIR-ATR spectroscopy, the biodegradation of PS was confirmed.
- ✓ Factors that affect PS biodegradation were concentration of MP, size of MP particles, and agitation speed of rotary shaker.

The obtained results show that tested bacterial culture has the ability to degrade PS particles.

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