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## BIODEGRADATION OF BP-3 USING GRAM-NEGATIVE BACTERIAL STRAINS

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### Introduction

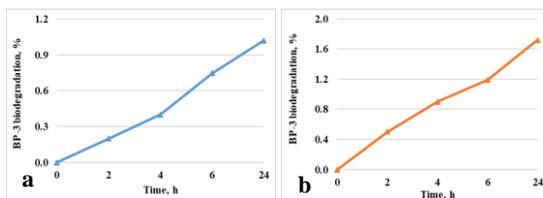
Organic UV filters are common compounds in the aquatic environment. These chemicals are used as active substances in chemical formulations of personal care products, in order to protect the skin, lips and hair against solar UV radiation. Organic UV filters easily reach the aquatic environment by incompletely removal in wastewater treatment plants and subsequent discharge of effluents into surface waters. These substances pose a threat to aquatic organisms because many of them exhibit hormonal activity. Benzophenone-3 (BP-3) is the most commonly used UV filter in cosmetic formulations worldwide. In surface waters in Romania, the concentration range detected for BP-3 ranged between 3-52 ng/L. Thus, degradation using bacterial strains can be a promising alternative to reduce the problems of environmental pollution with BP-3. Bacteria are a cost-effective alternative to catalytic processes. Finding suitable bacterial strains for BP-3 removal could improve the WWTP process by bioaugmentation. The aim of this study was the biodegradation of BP-3 in presence of two bacterial strains, namely *Salmonella typhimurium* and *Serratia rubidae*.

### Materials and methods

Initially, both gram-negative bacterial strains were seeded on solid nutrient medium (soybean agar tryptone) and incubated at 37°C for 24 h. After incubation, one colony of each bacterial strain was transferred to liquid broth - sodium lauryl sulphate specific culture medium. Incubation in liquid medium was performed for 24h in an incubator with stirring (130rpm) and heating at 37°C. The bacterial growth was determined at 600 nm using the UV-VIS spectrometer. Bacterial strains with 0.2 optical density (OD<sub>600nm</sub>) were incubated for 24 hours in the presence and absence of BP-3 at 10 mg/L. 1 mL aliquots were extracted from several incubation times such as 0h, 2h, 4h, 6h, 24h and stored in the freezer until LC-MS analysis.

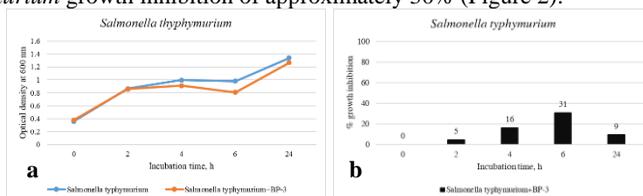
### Results and conclusions

Both experiments showed a similar behavior of BP-3 in the presence of *Salmonella typhimurium* and *Serratia rubidae* bacterial strains. BP-3 concentration decreased proportionally with increasing incubation time. However, compound biodegradation degree was not exceeded 2% after 24 hours from experiments initiation, in none of the cases (Figure 1).



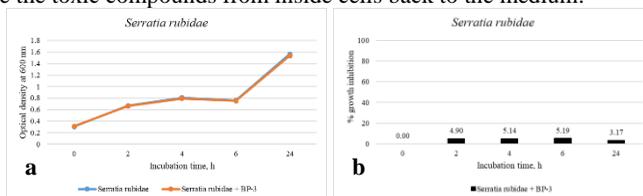
**Figure 1.** BP-3 evolution at different concentrations over time in the presence of *Salmonella typhimurium* (a) and *Serratia rubidae* (b)

Comparing the analytical and biological data, it was observed that after 6 hours incubation, the concentration of BP-3 decreased with the increase of *Salmonella typhimurium* growth inhibition of approximately 30% (Figure 2).



**Figure 2.** Optical density at 600 nm of *Salmonella typhimurium* in presence or absence of BP-3 (a) and growth inhibition (%) in presence of BP-3 (b)

Compared with *Salmonella typhimurium*, *Serratia rubidae* appeared to have a higher BP-3 biodegradation rate. The inhibition of bacterial growth in presence of BP-3 was lower than 10% (Figure 3b). The bacteria strain population had an increased growth rate slowly up to 6 hours, after which its growth in 24 hours was more accelerated. The increase in the bacterial population after 6 hours incubation time can be explained by one of the main defense bacterial mechanism based on efflux pumps which may eliminate the toxic compounds from inside cells back to the medium.



**Figure 3.** Optical density at 600 nm of *Serratia rubidae* in presence or absence of BP-3 (a) and growth inhibition (%) in presence of BP-3 (b)

In the context in which, so far, no studies have been reported on BP-3 biodegradation by reference bacterial strains, our results can be the basis for a large study involving other bacterial strains and higher incubation time.

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