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# Monitoring of biofilm growth on the microplastic surface

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## Abstract

In the aquatic environment, microplastics coexist with microorganisms that attach to the surface and form a biofilm - this process is often referred to as biofouling. Biofilm has a significant impact on the properties of microplastics, but its growth on the surface of microplastics is not well understood.

In this study, the formation of a biofilm on floating polyethylene microplastics was monitored under simulated laboratory conditions. Microplastics were exposed to natural stream water for 12 weeks, and the stream water was replaced every week to add new nutrients and microorganisms. Every two weeks, the microplastics were examined and the amount of biofilm developed, the content of extracellular polymeric substances (EPS), and the concentration of chlorophyll *a* were determined.

The growth rate of the biofilm corresponded to the common bacterial growth curve with three main phases: the lag phase (primo-colonization) in the first 14 days, the exponential growth phase from day 14 to day 42, and the stationary (maturation) phase from day 42 to the end of the experiment when the mass of the biofilm reached  $32.6 \pm 1.4$  % of the mass of the microplastic with biofilm. The EPS are an important component of any biofilm as they provide better adhesion of microorganisms and stability of the biofilm. During the incubation period, the EPS content was proportional to the amount of biofilm and had a constant specific concentration - at week 12, the concentration was  $0.47 \pm 0.08$  mg/g biofilm. The presence of chlorophyll *a* indicated the presence of photosynthetic organisms (e.g., green algae), and the concentration increased with time, reaching a maximum value of  $0.752 \pm 0.068$  mg/g biofilm. Our results shed light on the important interactions between microorganisms and microplastics and show the trend of various parameters related to the development of a biofilm on microplastics.

**Keywords:** aging, biofilm, characterization, microplastics

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