Impact of weathered and virgin polyethylene terephthalate nanoplastics on growth dynamics and the production of Extracellular Polymeric Substance (EPS) by microalgae

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In context of the ever-increasing plastic waste accumulation in the marine environment, it is important to understand the interaction between primary producers, i.e. microalgae, and the smallest plastic particles, i.e. nanoplastics (NP). A key unknown factor in this process is the role of extracellular polymeric substances (EPS). EPS production is a known algal stress response, and its adhesive properties may induce aggregation of algae, both with themselves and with other particles, which in turn may affect ecological and hydrodynamic processes. The ensuing effects can include the trophic transfer and vertical transport of nanoplastics. In this study, the impact of fragmented, polydisperse virgin polyethylene terephthalate (PET, $D_{average}$ = 1400 nm) and weathered polyethylene terephthalate (wPET , $D_{average}$ = 680 nm) on algae growth and the production of EPS was studied. We exposed the brackish marine microalgae *Rhodomonas salina* to a range of NP concentrations (10, 100 and 1000 and 10000 NPs ml^{-1}) for 12 days. A positive control with kaolin ($D_{average}$ = 2900 nm) a natural sediment particle, was included to distinguish particle effects from plastic effects. Baranyi parametric growth-models were fit to the data to analyze growth-dynamics. Exposure to all particles (plastics and kaolin) resulted in an initial increased growth rate, followed by significant decreases in algae population density. At low concentrations, the effect was independent of the particles' nature (natural or anthropogenic) or age (virgin or weathered). At the highest exposure concentration, the plastic particles caused significantly higher decreases in population density compared to kaolin, and the effects were amplified as the nanoplastics were weathered. The effects of weathered PET (10000 NPs ml^{-1}) on growth were accompanied by significant increases in cellular EPS production. This suggests that algae exhibit an increase in EPS-production as a stress response that is absent when exposed to natural particles or virgin nanoplastics. This raises questions about the toxicity mechanisms of NPs at concentrations of 10000 NPs ml^{-1} or higher, and hints towards the role of EPS production as a defence mechanism, which changes the energy budgets, with less energy allocated to growth. This study underscored the intricate interactions between particle types, age and concentrations, and their distinct impacts on algae density, growth inhibition and EPS production.

Keywords
Nanoplastics; Microalgae; Natural Particles; Weathering