## Potential risks of PET micro- and nanoplastics to the human gastrointestinal system

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## Abstract

Polyethylene terephthalate (PET) is one of the most widely used plastic polymer, ranking as the second most popular plastic in the food packaging industry, covering almost 16% of the total plastic consumption in Europe. In the last years, several studies have reported the presence of PET micro-and nanoplastics (MNPs) in a large variety of food products and beverages raising concerns about the health effects of PET MNPs after ingestion. In the present study, we evaluated the potential risks of PET MNPs to the human digestive system, using in vitro approaches based on gastrointestinal cell lines. For it, Caco-2 and Hep-G2 cells were exposed to 0.001 to 100mg/mL of PET MNPs for up to 24h, and effects on the gastrointestinal barrier integrity, cell viability and inflammatory responses were evaluated. PET MNPs were obtained following a top-down process that first cryomilled PET pellets to microplastic size  $(200-300\mu m)$  and second a high energy mechanically milling in water. PET MNPs were characterised by Fourier-transform infrared spectroscopy (FTIR), Raman spectroscopy, scanning electron microscopy (SEM), and dynamic light scattering (DLS). According to the characterisation results, PET MNPs showed primary size range between 50nm to 2mm, irregular shape and enhanced electrical charge of  $9.31\pm0.29$  nC (net charge) and  $17.34\pm2.15$  nC (tribocharge). Results of the *in vitro* experiments indicated that exposure for up to 4h to PET MNPs does not affect the gastrointestinal barrier integrity. Exposure for up to 24h to PET MNPs does not affect Caco-2 or Hep-G2 cells viability, however, induce inflammatory responses (increase of IL-6 and IL-8) in the two cell models. Despite not inducing cell death, PET MNPs induced important cellular responses that can trigger a cascade of events that can culminate in severe cell and tissue damage. Thus, the exposure to PET MNPs represent a potential risk to the human gastrointestinal system.

**Keywords:** Polyethylene terephthalate (PET), microplastics, nanoplastics, toxicity, human gastrointestinal cells

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