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### Deciphering the effects induced by nanoplastics in peripheral blood immune cells

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Humans can internalize nanoplastics (NPLs) through inhalation and ingestion due to the small size, allowing for their absorption, reaching the bloodstream, permitting their systemic biodistribution. Indeed, NPLs have been detected in human whole blood samples representative of the general population. Although several studies in invertebrates have linked NPLs exposure to disruption of the immune system, and some have assessed the in vivo immunotoxicity of NPLs in small mammals, information on the effects of NPLs on the human immune system is still lacking. In the present study, we exposed ex vivo human peripheral blood from healthy donors to several representative NPLs, not only to the most used (polystyrene. PS) but also to polyethylene terephthalate (PET) and to polylactic acid (PLA). To explore the influence of the physicochemical characteristics of NPLs on the analyzed effects, different sizes were chosen: 50 nm PS with different surface characteristics (pristine PS, carboxyl (-COOH), and amino (-NH<sub>2</sub>)), 150 nm PET, and 250 nm PLA. After 24 h of blood exposure to NPLs, white blood cells (WBC) were isolated and processed for flow cytometry analysis and single-cell RNA sequencing (scRNA seq). Our results show that NPLs internalize in human WBC, suggesting that the internalization dynamics of the NPLs depend on the WBC sub-population, and that it is modulated according to the type and functionalization of the particle.

The analysis of the scRNA seq data has permitted us to identify differentially expressed genes between each WBC population for the different NPL exposures. Remarkably, relevant proinflammatory markers are enhanced by NPLs exposure, suggesting the influence of NPLs in the production of an inflammatory environment in the body, thus altering human health and conferring an immune system more prone to be altered. Overall, these findings show us for the first time the NPLs effects on the human immune blood cells and emphasize that may be fundamental to control the NPLs exposure.

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#### Keywords:

Nanoplastics; exposure; scRNA seq; immune system; inflammation.