

***In vitro* bioassays as a tool to evaluate risk assessment of micro and nanoplastics**

M. Pérez*, C.Furió, A. Fernandez, & J. F. Ferrer

AIMPLAS, Valencia, Spain

* marinperez@aimplas.es

The concern regarding micro and nanoplastics (MNPs) has significantly increased as they can be present in food, water and environment. Once the particles enter the food chain, they can cross the biological barriers, as well as cell membranes, leading to different molecular effects.

To conduct a robust risk assessment of MNPs, two key challenges must be addressed: the **availability of well-characterized MNPs standards**, and the **establishment of a standardized battery of bioassays** to assess their effects across different cellular levels. Non-animal approaches, such as *in vitro* bioassays based on cell cultures, are especially relevant for evaluating complex mixtures of low-level contaminants. For particulate substances, it is essential to consider properties such as surface chemistry, impurities, dissolution rate, and stability under biological conditions.

The aim of the study is to develop standards of micro and nanoplastics to be used, first of all, in an *in vitro* gastrointestinal digestion to evaluate their stability and dissolution under biological conditions and, finally, to assess cytotoxicity (Alamar Blue), oxidative stress and genotoxicity (Micronucleus and Comet assay) in different cell lines.

The results showed no cytotoxic effects or induction of reactive oxygen species. However, certain plastic particles induced genotoxic responses in specific conditions, highlighting the need for further investigation. These findings underscore the importance of using well-characterized reference materials and standardized *in vitro* methods for hazard assessment.

In conclusion, this study reinforces the necessity of developing harmonized protocols for the preparation and testing of MNPs to generate reproducible data and ensure consumer safety.

Funding: This work was funded by IVACE (Institut Valencià de Competitivitat Empresarial).