

Fluorescent labeling of micro/nanoplastics for biological applications with a focus on “true-to-life” tracking

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The increased environmental presence of micro/nanoplastics (MNPLs), and the potential health risks associated with their exposure, classifies them as environmental pollutants of special environmental/health concern. Consequently, there is an urgent need for studies on the potential risks of secondary MNPLs. In this context, the use of “true-to-life” MNPLs resulting from the laboratory degradation of plastic goods, looks like a sound approach. Nevertheless, these non-commercial secondary MNPLs need to be *labeled* to track their presence/journey inside cells or organisms by using methods such as flow cytometry and confocal microscopy. Since cells are commonly analyzed by fluorescence techniques, and the use of fluorescent dyes seems to be a simple way to stain MNPLs, five different compounds comprising two chemical dyes (Nile Red and Rhodamine-B), one optical brightener (Opticol), and two industrial dyes (Amarillo Luminoso and iDye PolyPink) have been tested to determine their potential for such use. Using commercial standards of polystyrene nanoplastics (PSNPLs) with an average size of 170 nm, different characteristics of the selected dyes such as the absence of cell viability, specificity for plastic staining, no leaching, and lack of interference with other fluorochromes were determined. From the overall data obtained data in wide battery of assays performed, iDye PolyPink was the dye showing more advantages, regarding the other compounds, to be chosen as an effective dye to label “true-to-life” MNPLs. These advantages were confirmed using titanium-doped PETNPLs (obtained from the degradation of milk PET plastic bottles), as an example of true-to-life secondary NPLs. The results confirmed the usefulness of iDye PolyPink to label MNPLs, permitting cell internalization detection.

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