Workshop for quantitative assessment of microplastics in bottled waters and identification of plastic materials

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What is plastic?

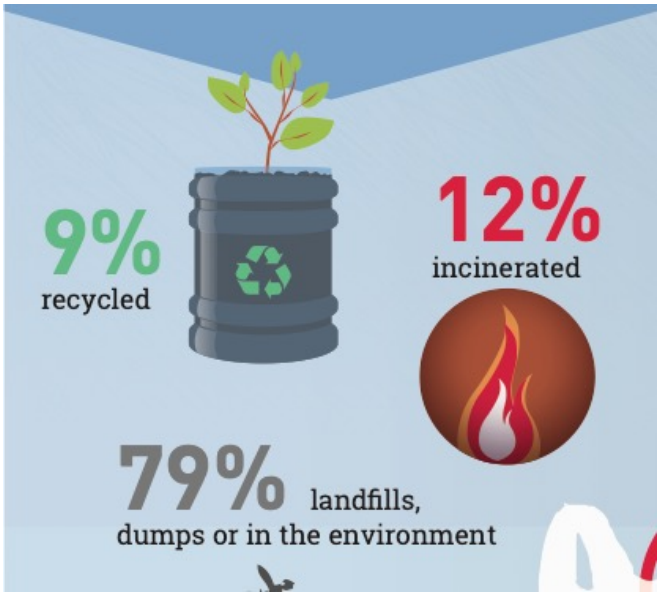
What are plastics?

- Most widely used non-metallic material today
- According to the ASTM Standard D883-20a, plastic is defined as a material that contains as an essential ingredient one or more organic polymeric substances of large molecular weight, is solid in its finished state, and, at some stage in its manufacture or processing into finished articles, can be shaped by flow
- Advantages such as light weight, good plasticity, good insulation and corrosion resistance
- Plastics are polymeric materials, which are not occurred naturally but are synthesized by small basic chemical molecules (i.e. monomers)

ASTM D883-20a, Standard Terminology Relating to Plastics, ASTM International, West Conshohocken, PA, 2020, www.astm.org

Problems from plastics

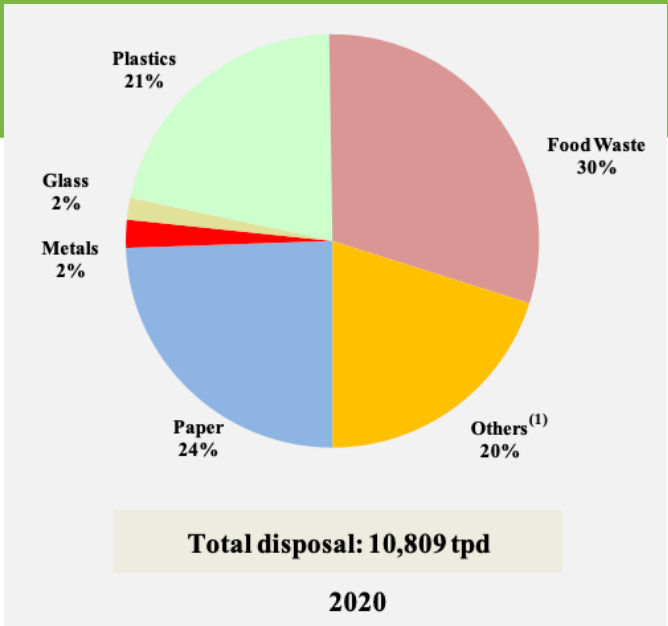
- Produced since 1950s
- 400 Million tonnes produced in 2015, 36 % are packaging materials (141 million tonnes)
- Impacts of plastic wastes:
  - Contaminate soil, water, and food chains
  - Affect wildlife through ingestions
  - Choke waterways and block sewage systems
  - Release toxic chemicals



https://www.unep.org/resources/report/single-use-plastics-roadmap-sustainability

Plastic wastes in Hong Kong (2020)

- 2,312 tonnes per day (21% of the municipal solid waste), ranked 3rd
- 11% of plastic wastes were recyclable
- With the Plastic Recycling Pilot Scheme, 94,700 tonnes of plastic recycled locally
- Apart from generated locally, plastics wastes can enter Hong Kong through importation or transboundary movement



Plastics						
- Plastic bags	**537**	(7.8%)	**267**	(6.7%)	**804**	(7.4%)
- Plastic bottles	**118**	(1.7%)	**61**	(1.5%)	**179**	(1.7%)
- Plastic / Polyfoam dining wares	**189**	(2.8%)	**77**	(1.9%)	**266**	(2.5%)
- Others(3)	**474**	(6.9%)	**588**	(14.8%)	**1,063**	(9.8%)
(Plastics) Sub-total	**1,318**	**(19.3%)**	**994**	**(25.1%)**	**2,312**	**(21.4%)**

https://www.wastereduction.gov.hk/sites/default/files/msw2020.pdf

Basel Convention and Wastes Disposal Ordinance

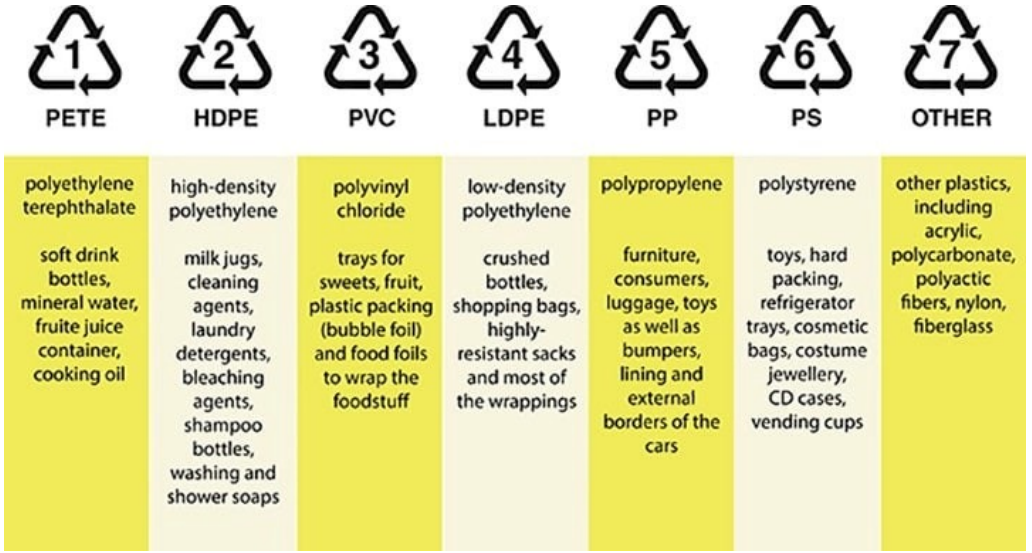
- In May 2019 the Basel Convention adopted the decision BC-14/12 to control transboundary movements of hazardous wastes
- Starting from January 1, 2021, except for unpolluted single material type plastic wastes listed in an environmentally sound manner, all other plastic wastes would be classified as "regulated plastic waste".
- Chapter 354 Waste Disposal Ordinance of the Hong Kong Legislation, unless waste is uncontaminated and is imported for the purpose of a reprocessing, recycling or recovery operation or the reuse of the waste, a permit issued by EPD is required

Photo 5

- http://www.basel.int/TheConvention/ConferenceoftheParties/ReportsandDecisions/tabid/3303/Default.aspx
- https://www.elegislation.gov.hk/hk/cap354

Recycling of plastics

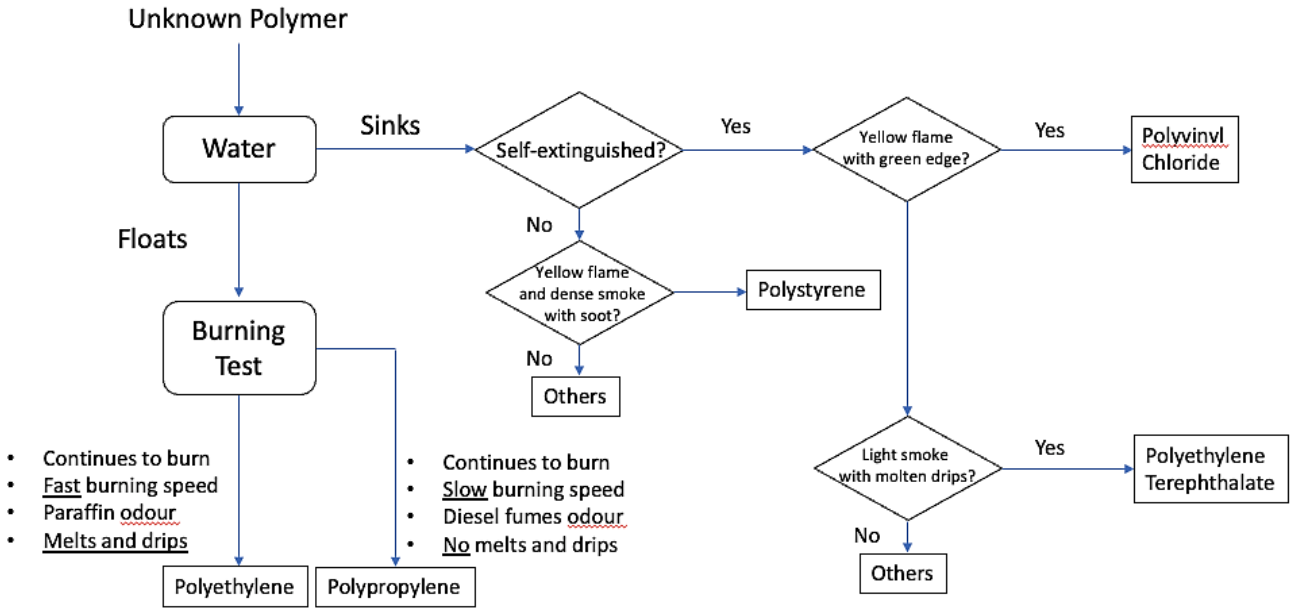
- Plastics are usually recycled mechanically: they are sorted, cleaned, shredded, melted and remoulded
- Different types of plastic recycled together would degrade the final plastic properties



1 PETE	2 HDPE	3 PVC	4 LDPE	5 PP	6 PS	7 OTHER
polyethylene terephthalate soft drink bottles, mineral water, fruite juice container, cooking oil	high-density polyethylene milk jugs, cleaning agents, laundry detergents, bleaching agents, shampoo bottles, washing and shower soaps	polyvinyl chloride trays for sweets, fruit, plastic packing (bubble foil) and food foils to wrap the foodstuff	low-density polyethylene crushed bottles, shopping bags, highly-resistant sacks and most of the wrappings	polypropylene furniture, consumers, luggage, toys as well as bumpers, lining and external borders of the cars	polystyrene toys, hard packing, refrigerator trays, cosmetic bags, costume jewellery, CD cases, vending cups	other plastics, including acrylic, polycarbonate, polyactic fibers, nylon, fiberglass

- https://learn.eartheasy.com/articles/plastics-by-the-numbers/

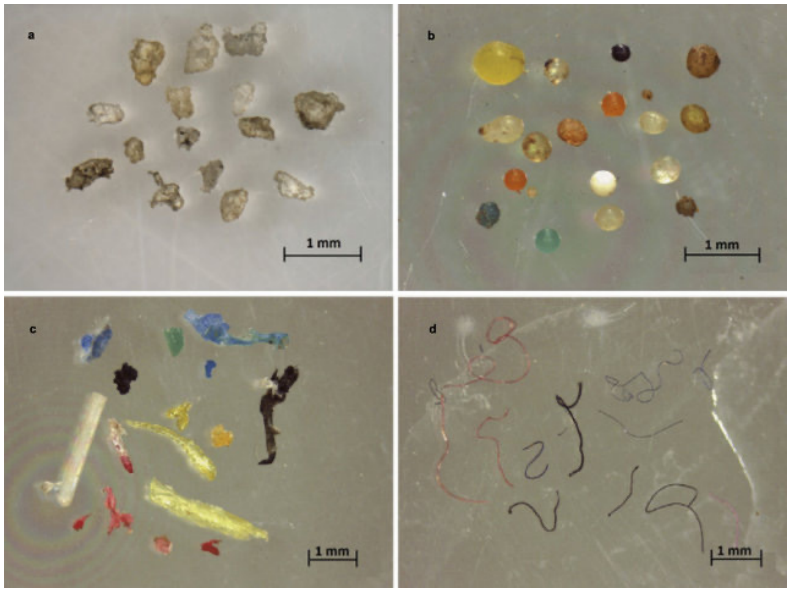
How to identify plastics preliminary?



What are microplastics?

What are microplastics?

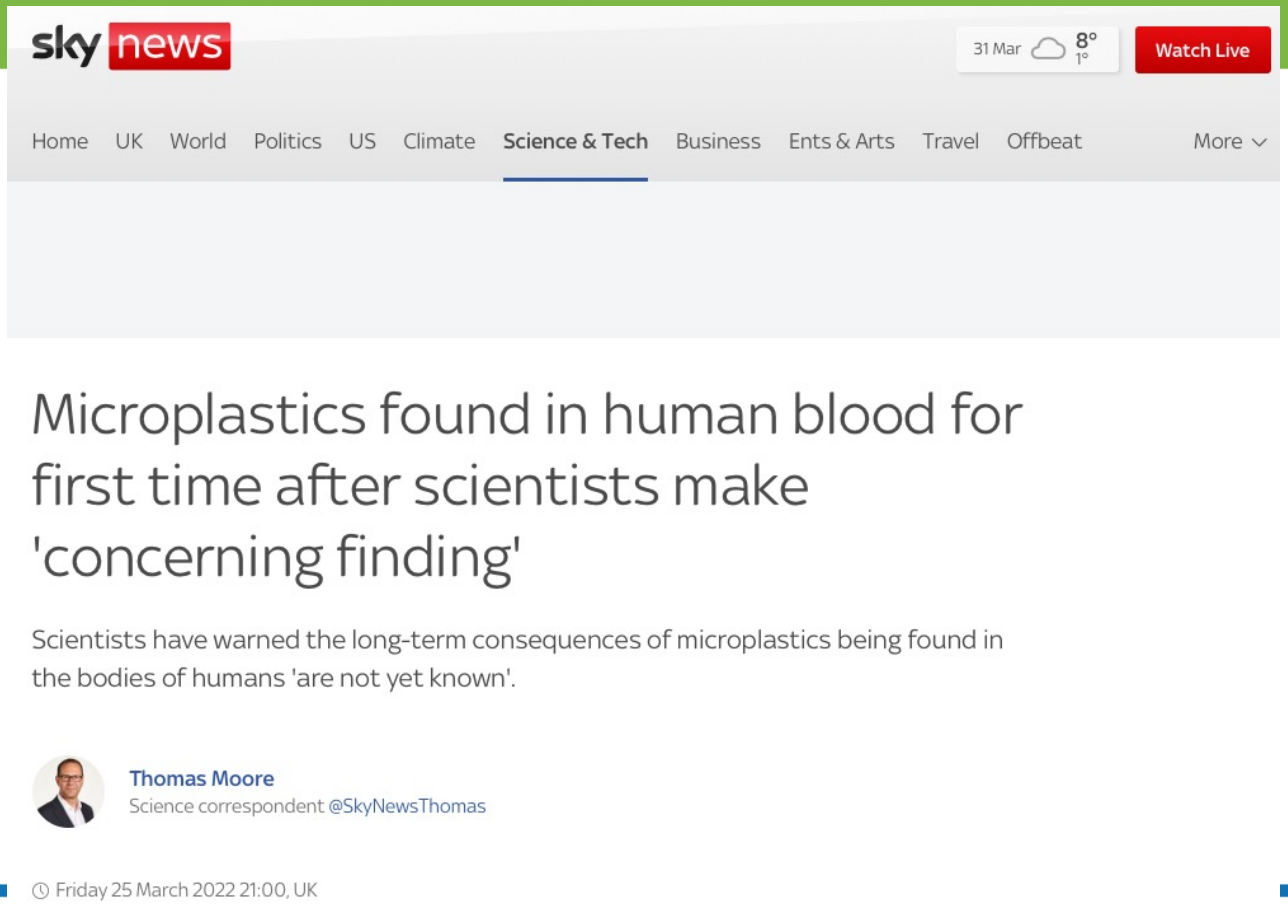
- Define plastic debris with a diameter less than 5 mm
- Primary microplastics: Directly released in the environment as small particles (~15 – 31%)
  - laundering of synthetic clothes
  - Abrasion of plastic articles
  - Cosmetics and personnel care products (e.g. face scrubs)
- Secondary microplastics: degradation of larger objects (~69 – 81%)



Koyuncuoğlu, Pelin & Erden, Gülbin. (2021). Sampling, pre-treatment, and identification methods of microplastics in sewage sludge and their effects in agricultural soils: a review. Environmental Monitoring and Assessment.

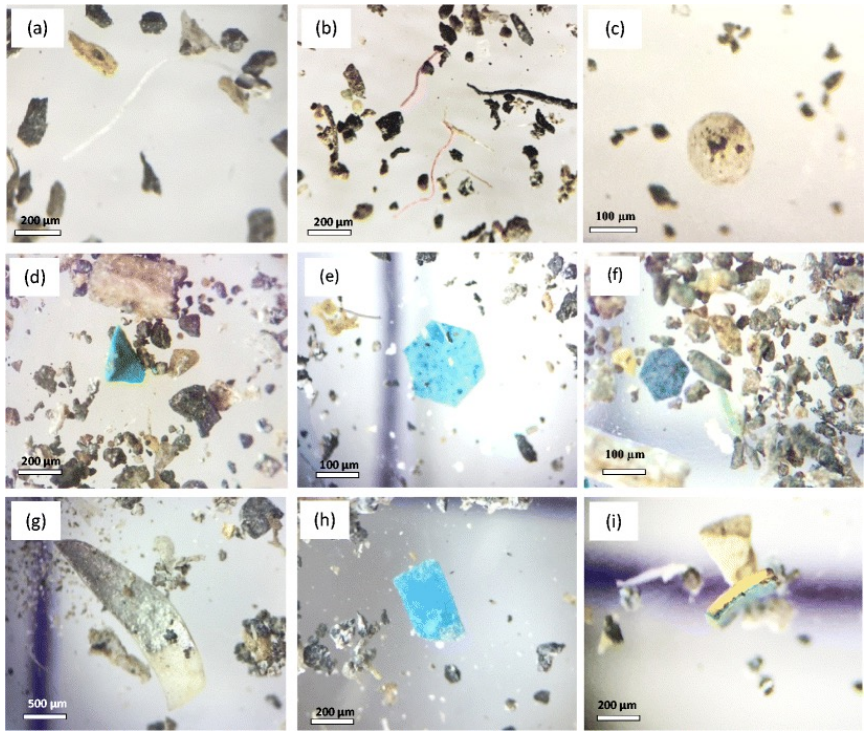
https://www.europarl.europa.eu/news/en/headlines/society/20181116STO19217/microplastics-sources-effects-and-solutions

Microplastics penetrate into blood stream

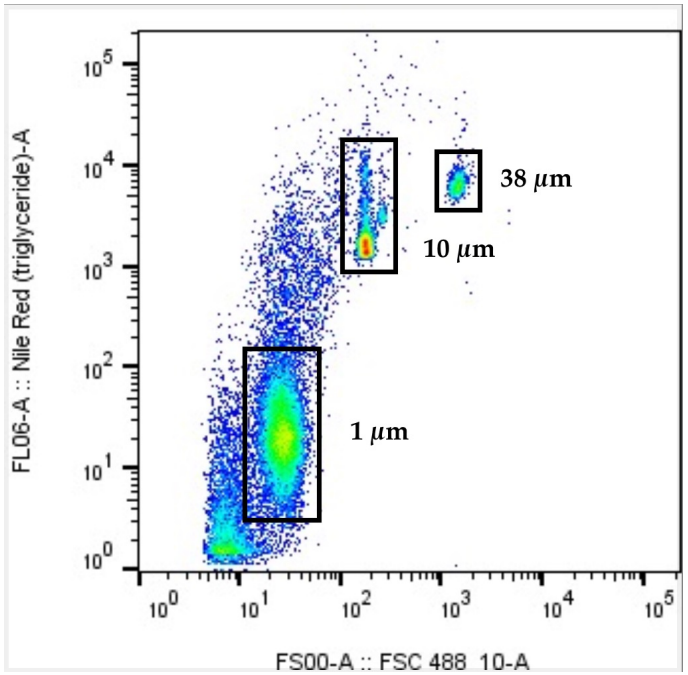


Risks and difficulties in microplastics study

- Size distribution of microplastics (MPs) is crucial
- Smaller sizes MPs may be able to penetrate through biological barriers
- Currently MPs below 50 µm quantified manually using microscope, which is time consuming and expertise required
- Microplastics lower than 50 µm is difficult to be identified under microscope



Methods to quantify small microplastics down to 1 µm



water

MDPI

Article

Flow Cytometry as a Rapid Alternative to Quantify Small Microplastics in Environmental Water Samples

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Abstract: The most frequently used method to quantify microplastics (MPs) visually by microscope is time consuming and labour intensive, where the method is also hindered by the size limitation at 10 µm or even higher. A method is proposed to perform pre-concentration of MPs by vacuum filtration, hydrogen peroxide wet digestion, fluorescent staining and flow cytometric determination to rapidly detect and quantify small MPs sized from 1–50 µm. The method performance was evaluated by the spiking of seven different types of polymer, including polystyrene (PS), low-density polyethylene (LDPE), polypropylene (PP), poly(methyl methacrylate) (PMMA), polyvinyl chloride (PVC), polylactic acid (PLA) and acrylonitrile butadiene styrene (ABS) at different levels (400, 4000, 40,000 particles mL^{-1}), with a satisfactory overall % recoveries (101 ± 19.4%) observed, where in general no significant difference between the two methods was observed. Furthermore, a pre-concentration process by vacuum filtration was introduced to reduce the matrix effect. After pre-concentration, satisfactory % recoveries and accuracy in MP counts resulted from both ultrapure water (94.33 ± 11.16%) and sea water (103.17 ± 9.50%) samples. The validated method using flow cytometry can be used to quantify MPs in environmental water samples that can reduce time and human resources.

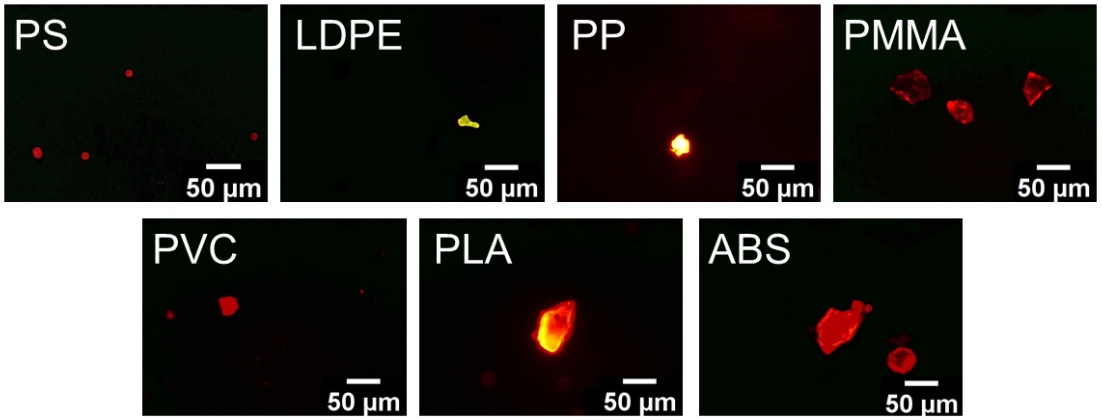
Keywords: microplastics; flow cytometry; Nile red fluorescence staining; visual identification

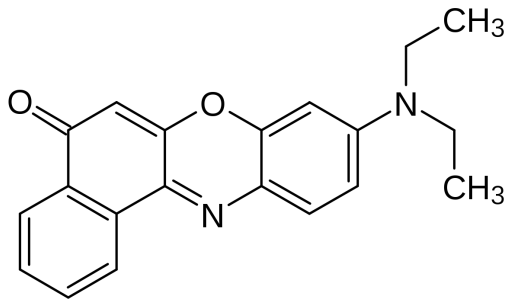
Citation: Tse, Y.-T.; Lo, H.-S.; Chan, S.M.-N.; Sze, E.T.-P. Flow Cytometry as a Rapid Alternative to Quantify Small Microplastics in Environmental Water Samples. *Water* **2022**, *14*, 1436.

https://www.mdpi.com/2073-4441/14/9/1436

Staining of microplastics by Nile red indicator

- Nile red (9-diethylamino-5H-benzo[a]phenoxazine-5-one) is the most commonly used lipophilic stain
- Poor solubility and fluorescence in water
- Colour emission varying from deep red to strong yellow gold
- Ideal to stain microplastics in bottled water





Microplastics by in bottled waters