Supplementary Information

Fractionating microplastics by density gradient centrifugation: A novel approach using LuerLock syringes in a low-cost density gradient maker.

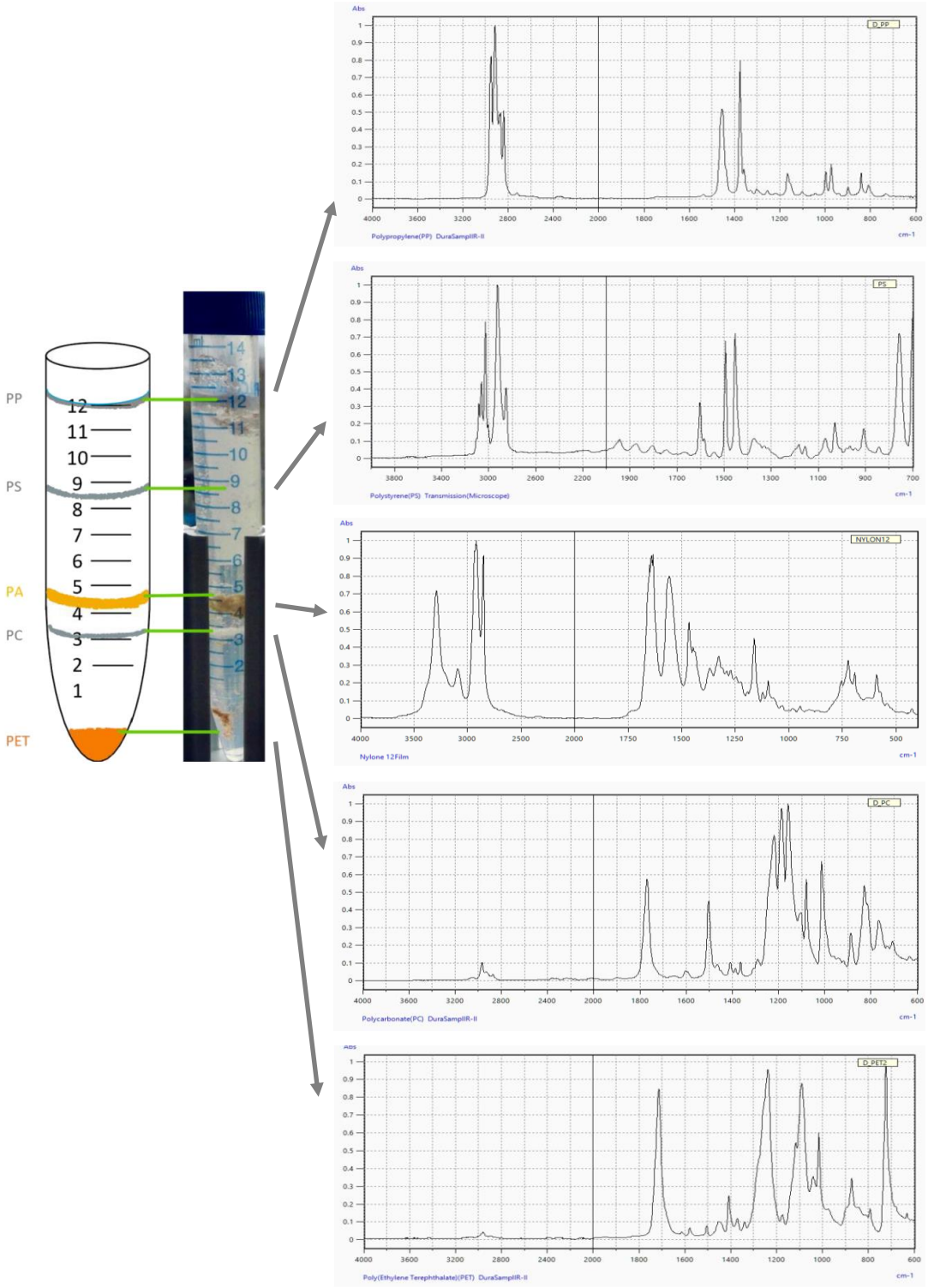


Figure A. *Tested microplastic particles of the types PP, PS, PA, PC and PET and their ATR Spektra.*

Hazards

Students should always use appropriate personal protective equipment. Piercing the needle through the stopper should only be done by a teacher. While using the gradient maker students should be aware about the needle. It is recommended to cover the needle when the maker is not in use. Potassium carbonate (K_2CO_3) reacts alkaline in water, so as precautions wearing safety glasses is obligatory. When working with potassium carbonate fully consider the safety data sheets for H- and P-statements.

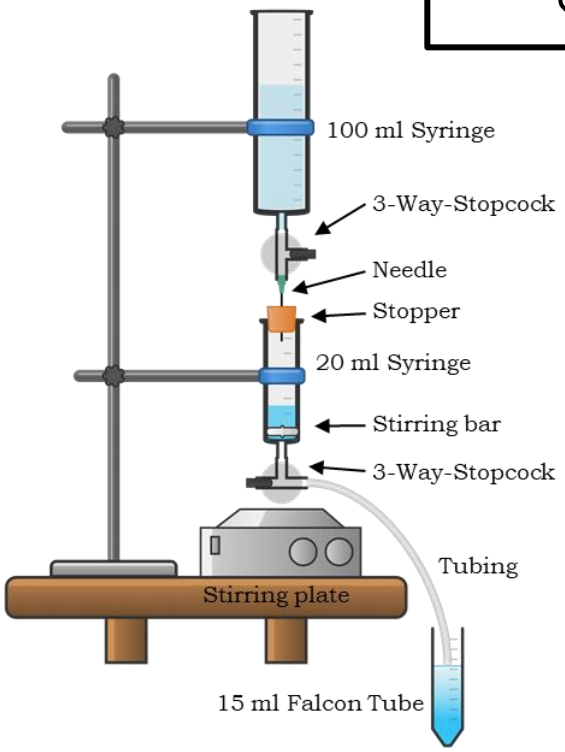
Density Centrifugation of Microplastik Particles

Instruction

Materials

- Retort Stand and Clamp
- Magnetic stirrer
- Centrifuge
- 100 mL syringe
- 20 mL syringe
- Rubber stopper with a pierced cannula
- 2 x Three-way stopcock
- Rubber tube
- Stirring bar
- Disposable pipette
- 15 mL Falcon tube
- Microplastic sample

- Distilled water
- Saturated potassium carbonate solution



Instructions

Set up the apparatus as shown in the figure above. Ensure that all three-way valves are closed at the start.

1) Add 5 mL of saturated potassium carbonate solution into the 20 ml syringe. Then add the stirring bar.
2) Fill the 100 mL syringe with approximately 70 mL of distilled water.
3) Assembly of the apparatus: Now place the 100 mL syringe with the rubber stopper and cannula onto the 20 mL syringe. Ensure that the rubber stopper seals tightly with the 20 mL syringe. Turn on the magnetic stirrer so that the stirring bar in the 20 mL syringe rotates evenly.
4) Now, first open the upper three-way valve and then the lower three-way valve.
5) Creation of the density gradient: Carefully catch the liquid with the help of the tubing in a Falcon tube. To do this, always carry the end of the tubing upwards, just above the surface of the liquid.
6) Now carefully add the microplastic sample into the density gradient. Seal the Falcon tube and place it in the centrifuge. Put identical prepared Falcons with the same weight in opposite to each other in the centrifuge to prevent damage to the rotor.Turn on the centrifuge for 30 minutes at 5000 rpm.

H A Z A R D S

	Potassium Carbonate	H315: Causes skin irritation. H319: Causes serious eye irritation. H335: May cause respiratory irritation.

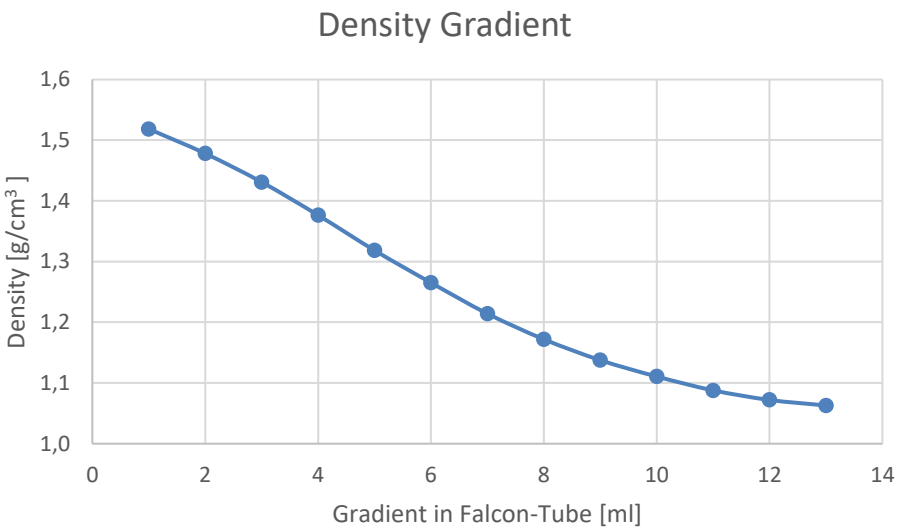
Density Centrifugation of Microplastic Particles

Protocol

Observation

Results

Type of Plastic	Density Range [g/cm³]
PA (Polyamide)	1,02-1,15
PUR (Polyurethane)	1,05-1,28
PET (Polyethylen terephthalate)	1,29-1,45
PVC (Polyvinylchloride)	1,10-1,58



Tasks

1) Make assumptions about your observations: how many microplastic particles of each plastic type have you got in your tube? Use the table and the graph to assist you. Then explain why you can only make assumptions about the plastic type using the table.

2) Use the internet for research on the applications of the four listed types of plastics.

Density Centrifugation of Microplastic Particles

Technical background

Microplastics have become widely dispersed in the environment and are turning into an ever-growing global issue. Microplastic particles were discovered in the oceans worldwide early on, and reports of their negative effects soon followed. Only in recent years has microplastic been increasingly detected on land, where its impact on the environment, organisms, and human health constitutes a large part of current research. Thus, detecting microplastics in the environment is becoming an increasingly important research field within environmental analytics, requiring various methods for detection and analysis. However, all methods share a common foundation: (Micro)plastics are separated and identified based on one of their specific chemical and/or physical properties.

A common characteristic relied upon in microplastic analysis is the physical size "density". By definition, density is a material constant and is the quotient of mass to the volume of a material. It indicates the ratio of mass to volume of a substance. But what lies behind this abstract description? Imagine you are holding an equal volume of two different materials in each of your hands; for example, cotton in one hand and stones in the other. It will be clear that the hand full of stones will be heavier than the hand with the same amount of cotton. Therefore, the stones have more weight for the same volume - they are denser.

The different types of plastics also have various but very specific densities. We will utilize this fact in this experiment to examine microplastic particles more closely. Since it would be very laborious to cut the particles all the same size and weigh them, the density is determined differently. With the help of two liquids, one with low density (water) and one with high density (potassium carbonate solution), any solution with a density between these two material-specific densities can be created in principle. Instead of creating many individual solutions, the two liquids are filled into a tube while mixing, creating a smooth transition from a solution with high density to a solution with progressively lower density. When the microplastic particles are added, they will settle exactly at the spot in the solution that has the exact same density as the microplastic particle itself.

Literature:

- Okunola A, A., Kehinde I, O., Oluwaseun, A., & Olufiropo E, A. (2019). *Public and Environmental Health Effects of Plastic Wastes Disposal: A Review*. Journal of Toxicology and Risk Assessment, 5(2).
- Duis, K., & Coors, A. (2016). *Microplastics in the aquatic and terrestrial environment: sources (with a specific focus on personal care products), fate and effects.* Environmental Sciences Europe, 28(1), 1–25

Gohla, J., Bračun, S., Gretschel, G., Koblmüller, S., Maximilian, W., & Pacher, C. (2021). *Potassium Carbonate (K_2CO_3) – A cheap, non-toxic and high-density floating solution for microplastic isolation from beach sediments*. Marine Pollution Bulletin, 170.