Supplementary Information

1 Preparation

1.1 Pre-trials

Pre-trials for testing the efficiency of the procedure have been carried out beforehand. To determine the loss of particles in the nylon washing bags, aluminium silicate (200 µm – 500 µm) and polyethylene microbeads (150 µm - 800 µm) were used. Due to the mesh size, the inner washing bag retains particles ≥ 300 µm (Figure S1A), the outer washing bag particles ≥ 100 µm (Figure S1B). Therefore, particles smaller than 100 µm were excluded from the analysis. Using three washing bags five times, each time freshly filled with aluminium silicate tested the loss of particles. These pre-trial bags were weight before and after the washing procedure. An average loss of 6.29 g (14.79 %) was determined. This high loss is assumed to result from the sharp form of the aluminium silicate fractions, which damages the nylon fabrics (Figure S1D). The loss of 0.358 g (2.99 %) was much less while using the less sharp microbeads instead of aluminium silicate. Here, no damage could be recognised. Since microbeads are more comparable to actual microplastic in their characteristics, an average loss of 3 % have to be taken into account per sample. Sewing the bags additionally in a cross stitch helps to keep the loose fibres of the nylon cloth together, and further secures the closeness.

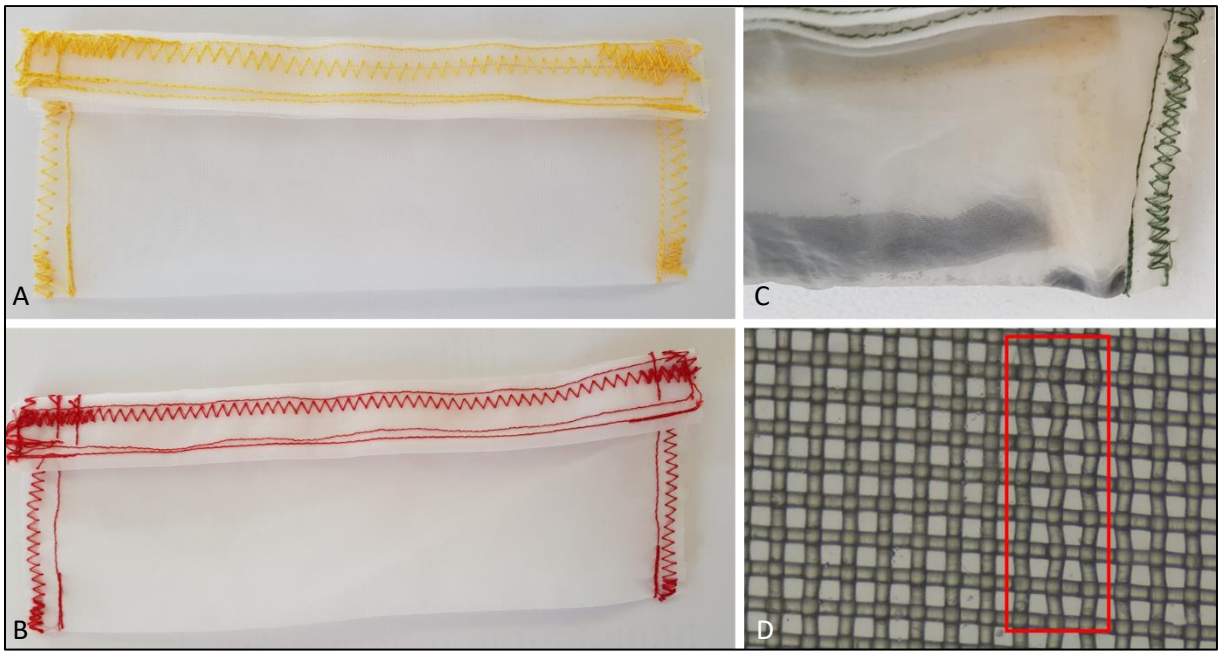


Figure S1. The washing bag design used for the pre-trials using pre-sewed bags closed with stitched hook and loop fastener (A & B). The size fragmentation based on the nylon cloths operates successful during the washing procedure, what approved the method of different cloths (C). These pre-trials were conducted with aluminium silicate (200 µm – 500 µm).

Figure S2. The second circle of pre-trials were conducted with microbeads (ca. 150 µm – 800 µm; see B & C). Measurements was conducted beforehand to check if the used mesh sizes are suitable to keep the particles in the washing sachets. Oil pellets (green) were removed during washing procedure (see A); the remaining microbeads were used for determining the loss of particles.

1.2 Working in an Acrylic Hood

The sample handling was conducted in an acrylic hood. This closed environment decreases the risk of contaminating the samples. Holes for the hands, the electric cable and hoses as well as fixing equipment, were sawn out (Figure S4).

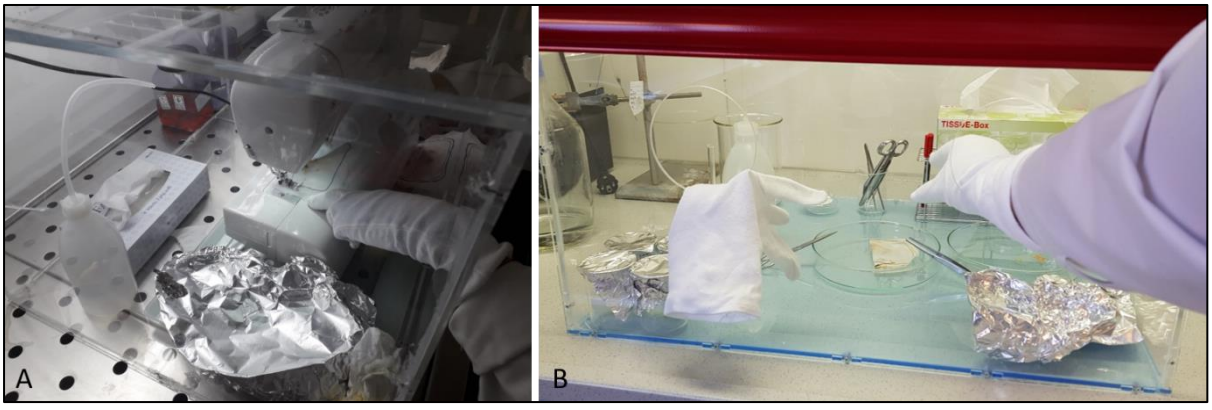


Figure S3. Work process in an acrylic box. A) Sewing the samples into nylon washing bags. B) Processing the samples with cotton gloves within an acrylic box situated under a fume hood.

2 Intestinal and Faecal Samples

2.1 New design of the washing bags

For analysing the intestine and faeces samples, a new design of the washing bags was established (

Figure S4). The hook and loop fastener as seen in Figure S1 was discarded, to reduce the potential of hold back particles. Furthermore, the samples are directly sewed into both cloths to decrease the contamination and it turned out to be a faster method. For distinguishing samples, rinsed metal paperclips where added.

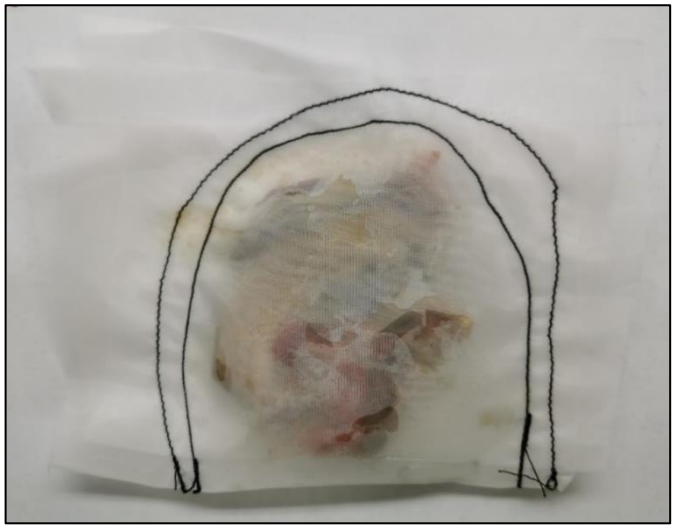


Figure S4. Sliced intestine sample sewed in the nylon washing bags. Washing bag consists of two nylon cloths (inner bag: 300 µm and outer bag: 100 µm). Overhanging nylon cloth is cropped before conducting the washing cycle.

2.2 Prevention of overcounting

Lost fibres of the nylon washing bags can easily be identified, since they are showing a distinct fibre pattern (Figure S5). Black cotton yarn was used for sewing, since the Nile Red staining does not stain it (Figure S6). Therefore, these nylon fibres and the black cotton were not included in this study. All particles <100 µm are discarded due to the mesh size of the outer bag (100 µm) to avoid counting particles entering the sample as contamination.

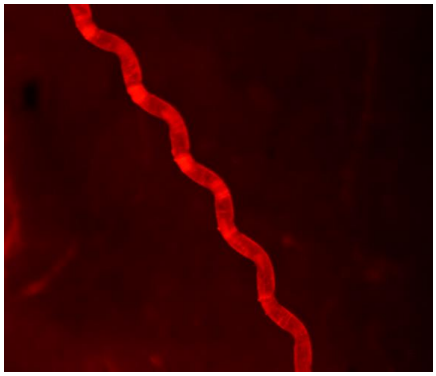


Figure S5. Nylon fibres of the washing bag cloth with an incomparable zigzag shape and reddish colour.

3 Identification Catalogue

Particles are assumed as MP in the presented study after the following criteria:

- the particle is self-contained
- the structure distinguishes from those associated with a biogenic one (e.g. chitin structure, plant-based or fish-related ones)
- the particle is fluorescent (yellow to whitish colour)
- the location of the particle is ca. 3 mm distant from the filter edge (avoidance of contamination)

The following figures underpin the beforehand described list of issues and can help for distingishing MP from biogenic particles.

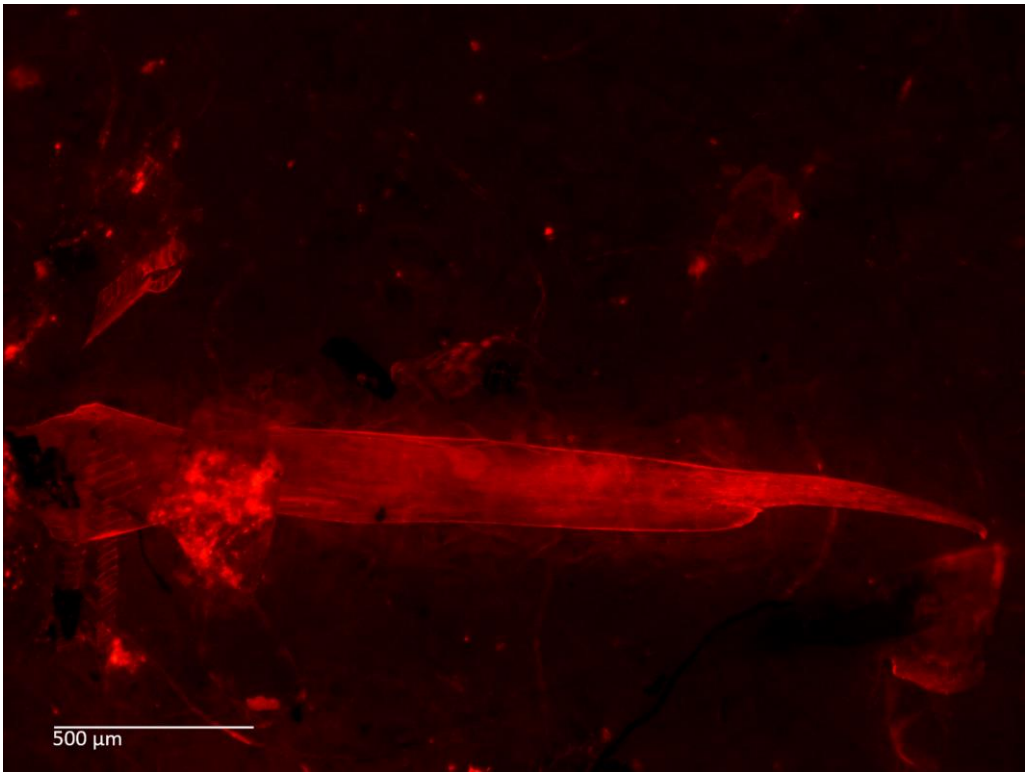


Figure S6. Fish bone.

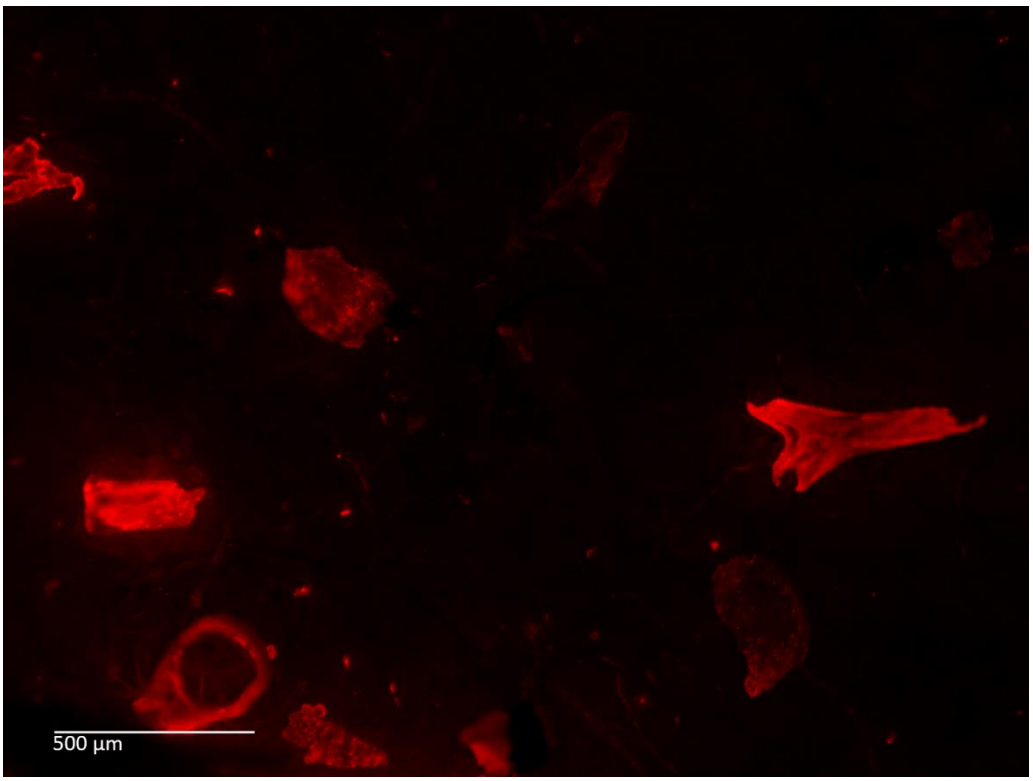


Figure S7. Fish bones and plant-based fragments.

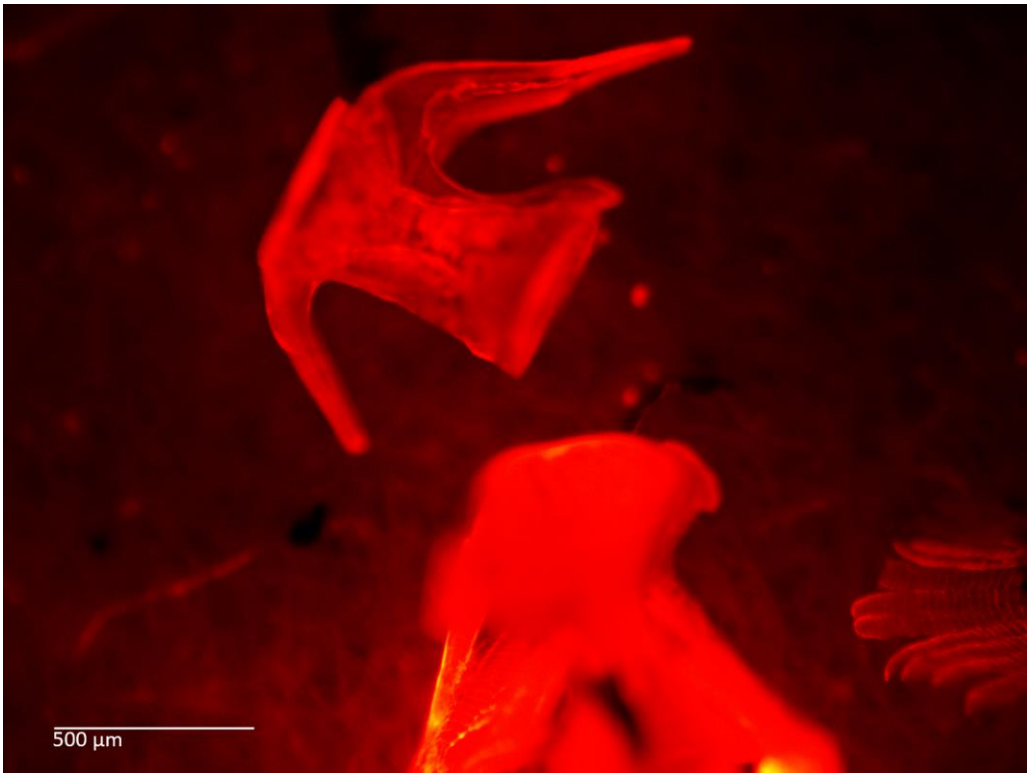


Figure S8. Fish vertebrae.

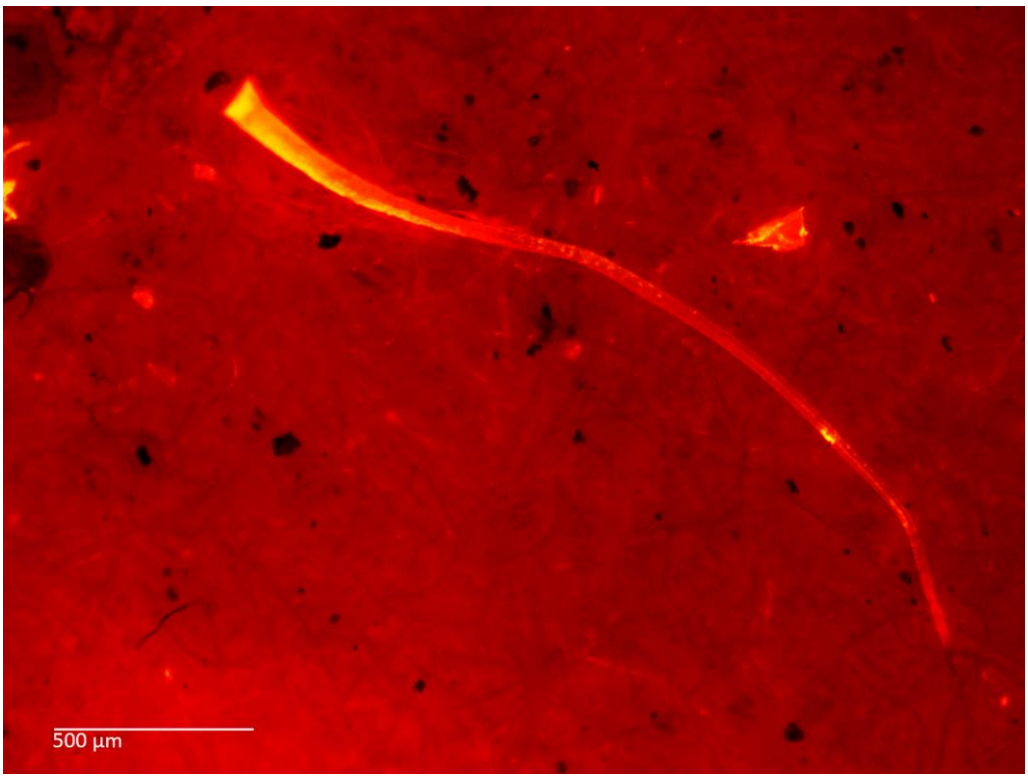


Figure S9. Fishbone

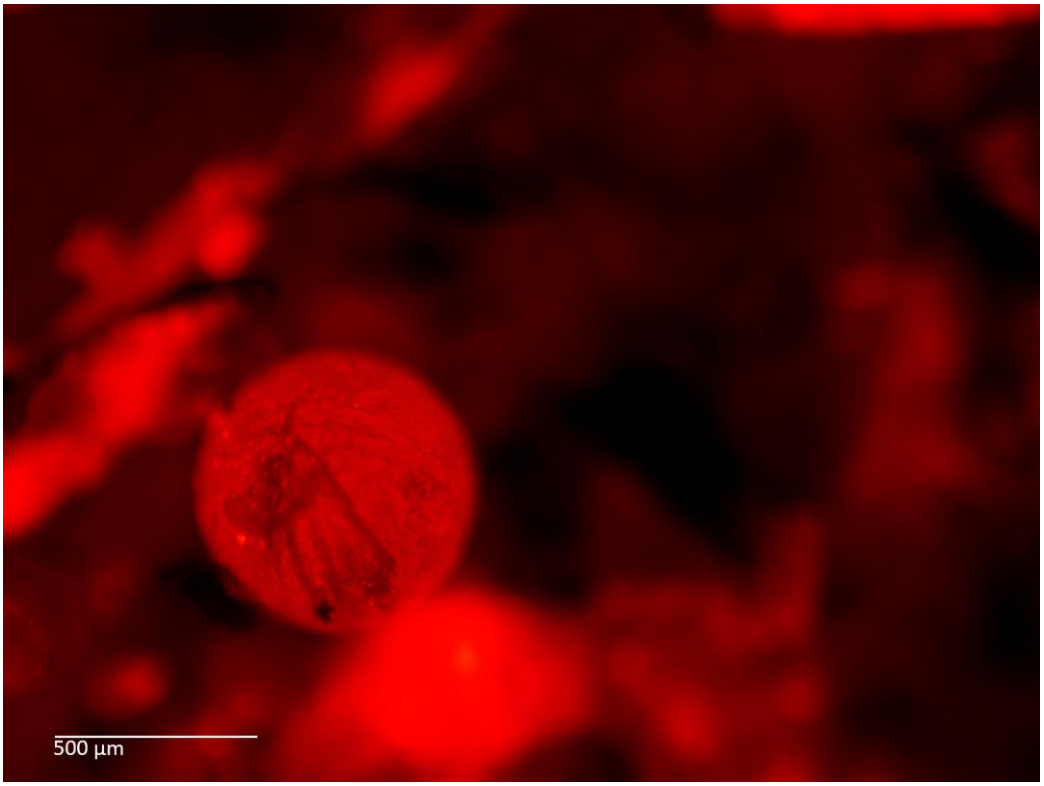


Figure S10. Biogenic fragment (e.g. plant-based seed or a big fish lens). Certainly, no microplastic bead.

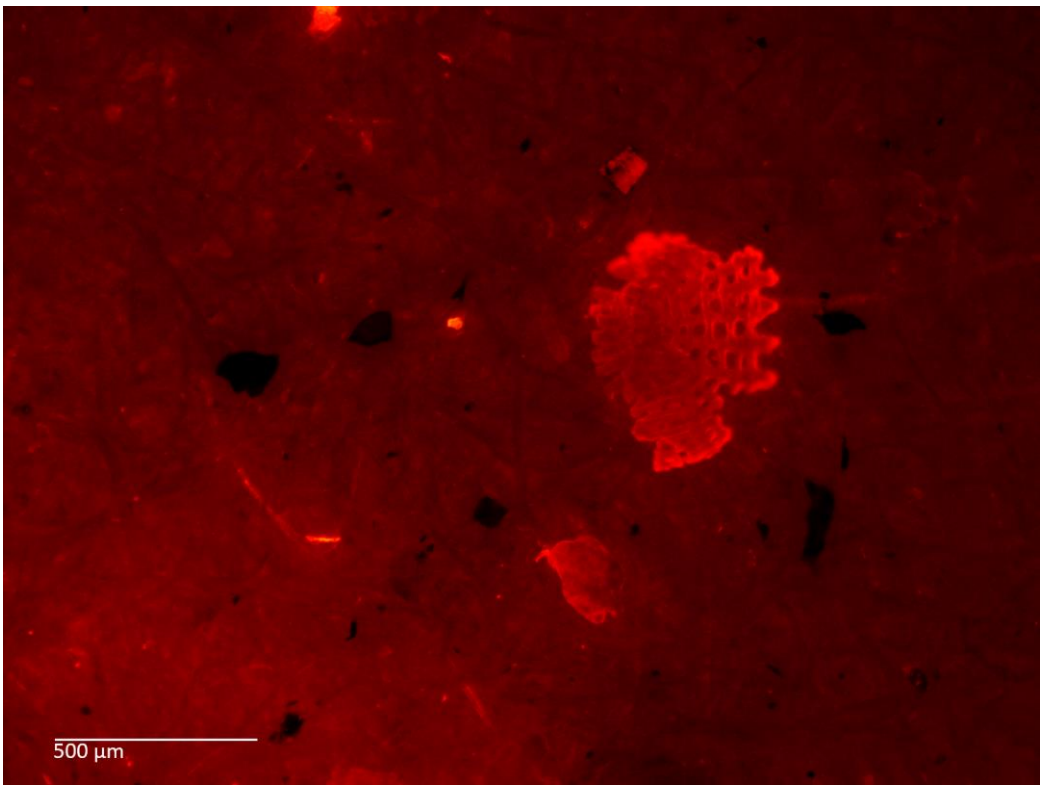


Figure S11. Biogenic fragment.

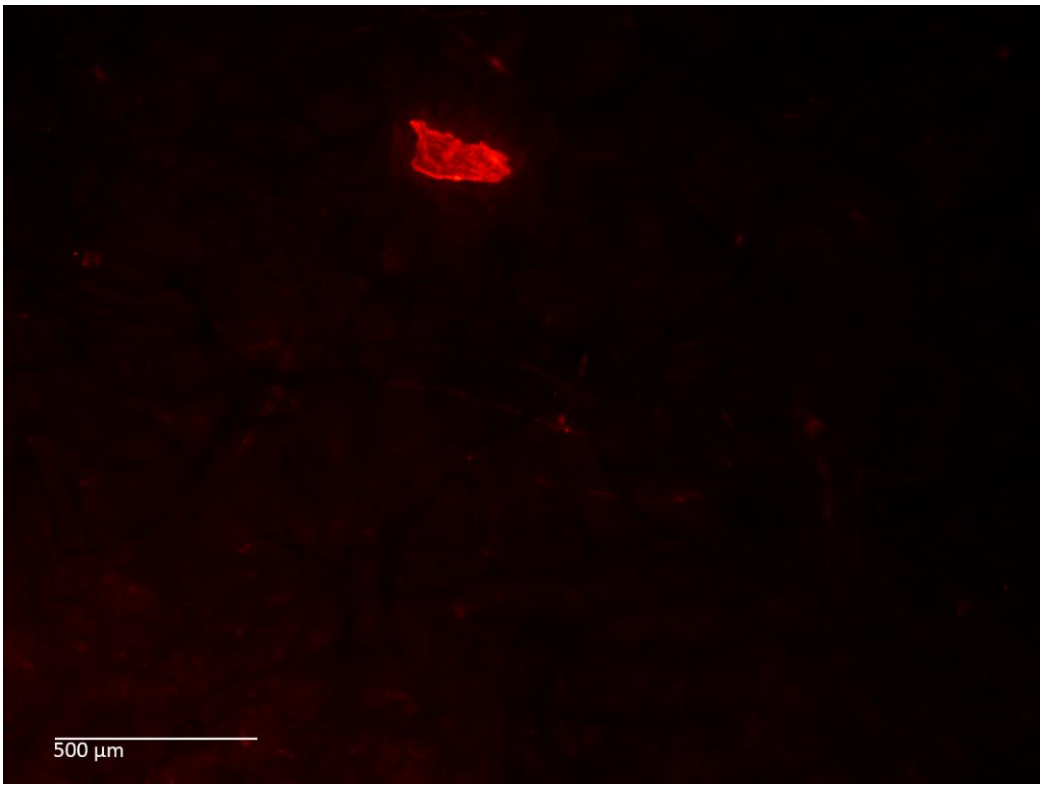


Figure S12. Plant-based fragment.

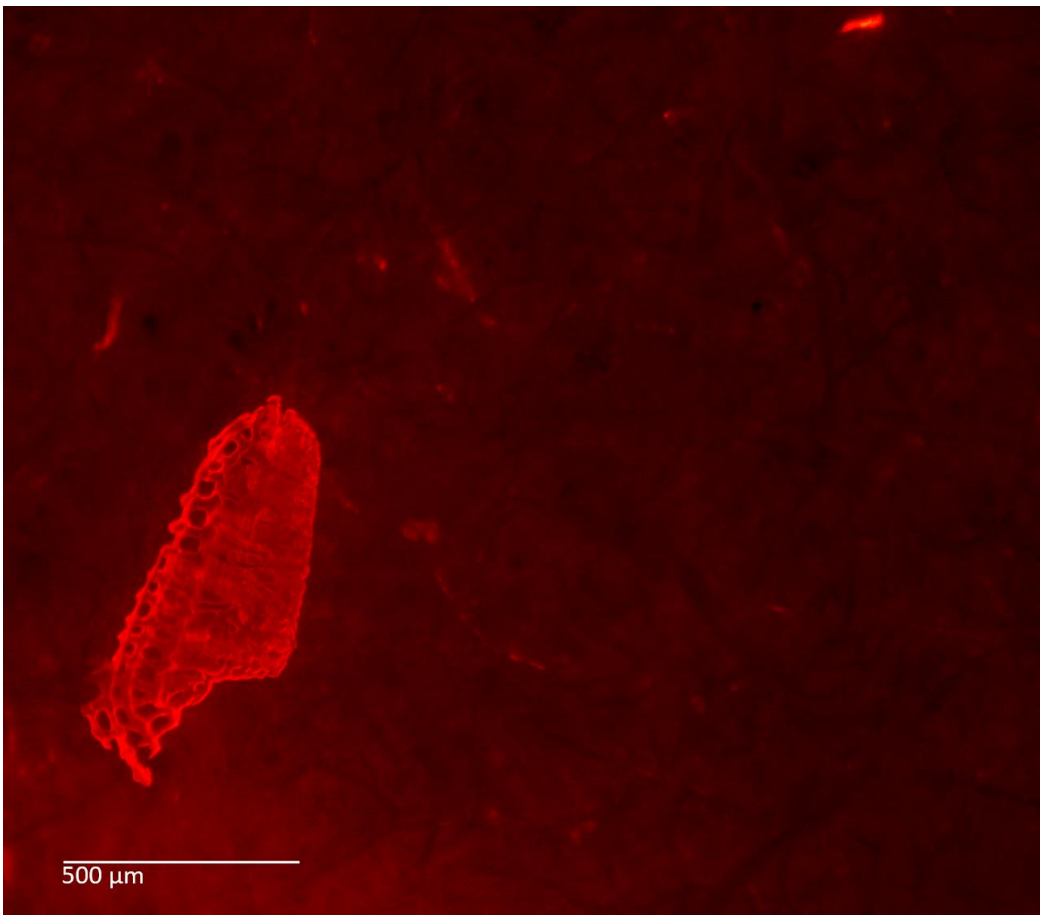


Figure S13. Biogenic fragment.

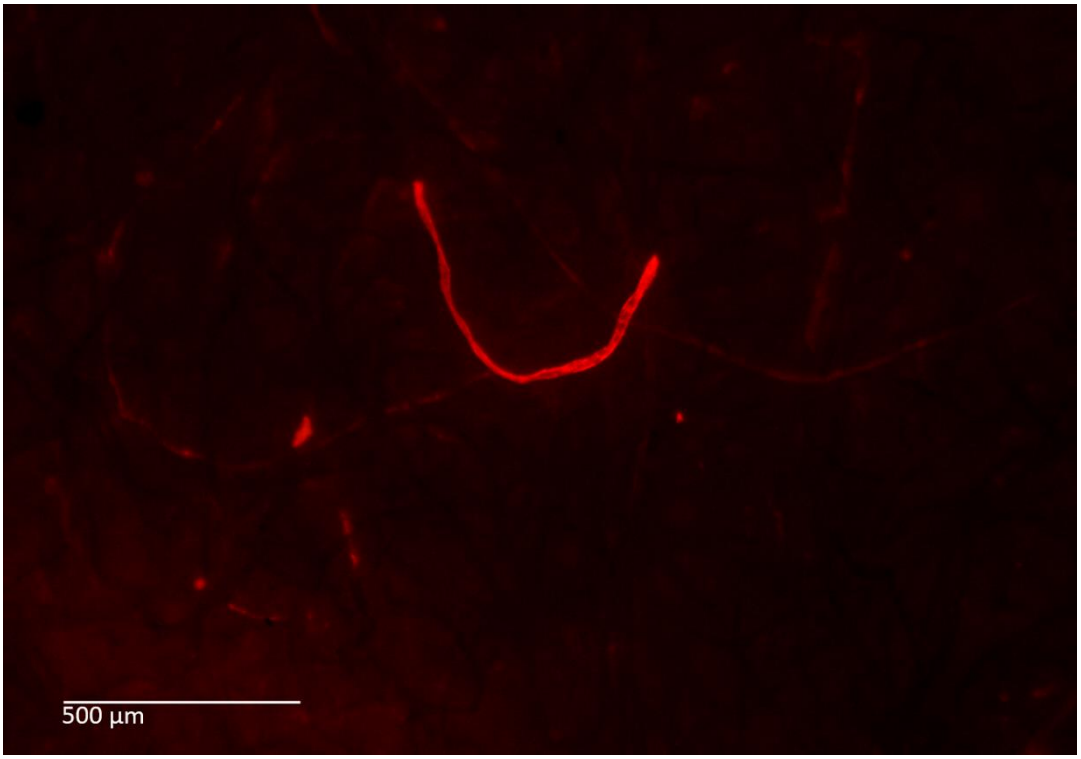


Figure S14. Cellulose fibre.

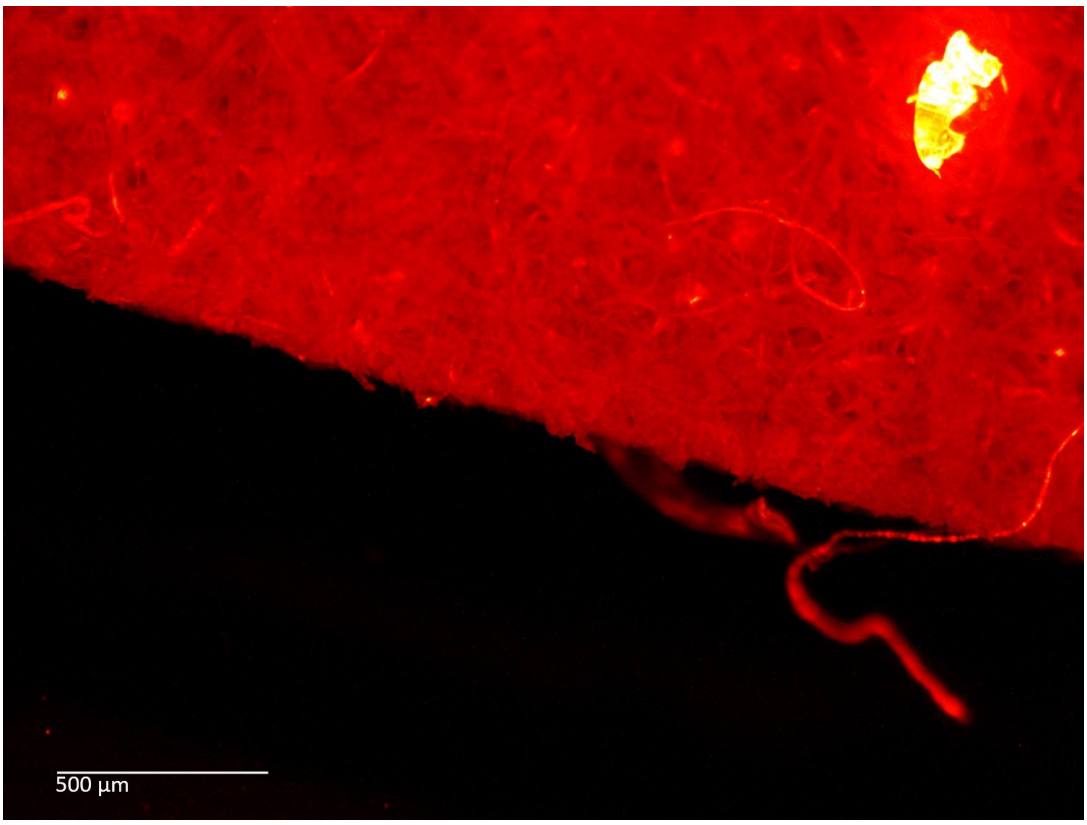


Figure S15. Microplastic at the edge of a filter. This fragment is to close located to the edge, thus it was not counted.

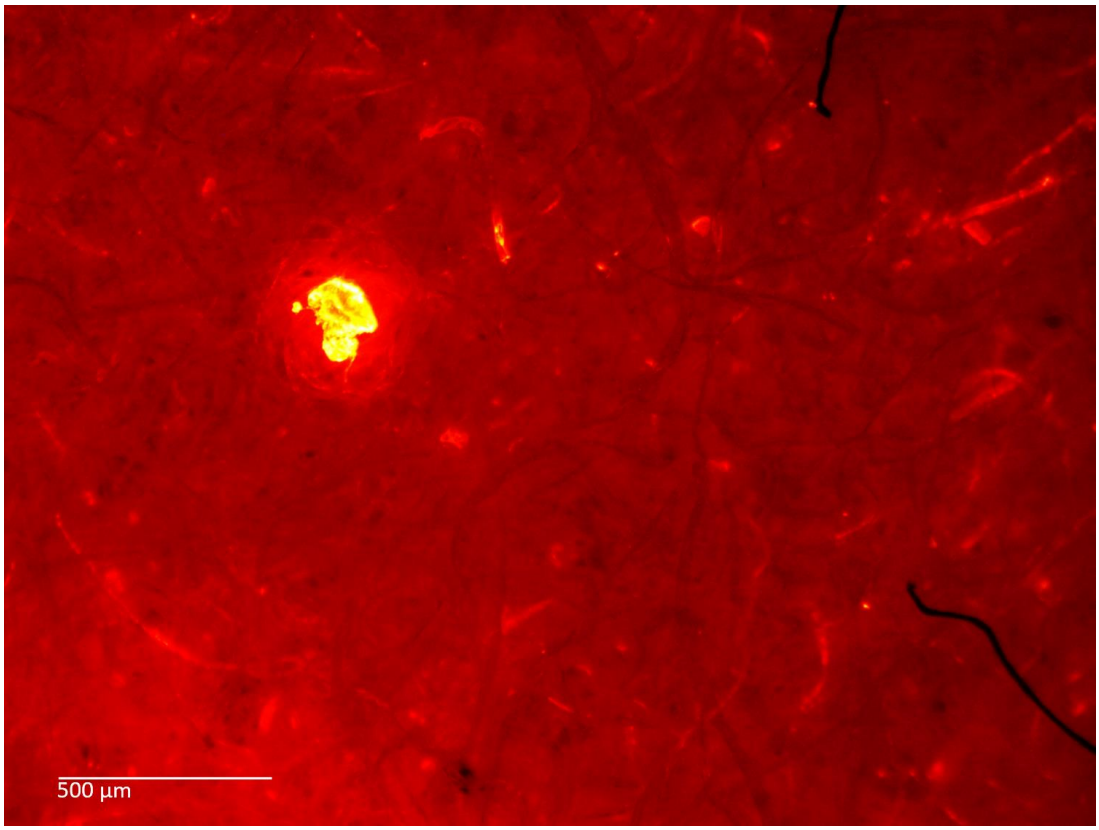


Figure S16. Countable microplastic fragment.

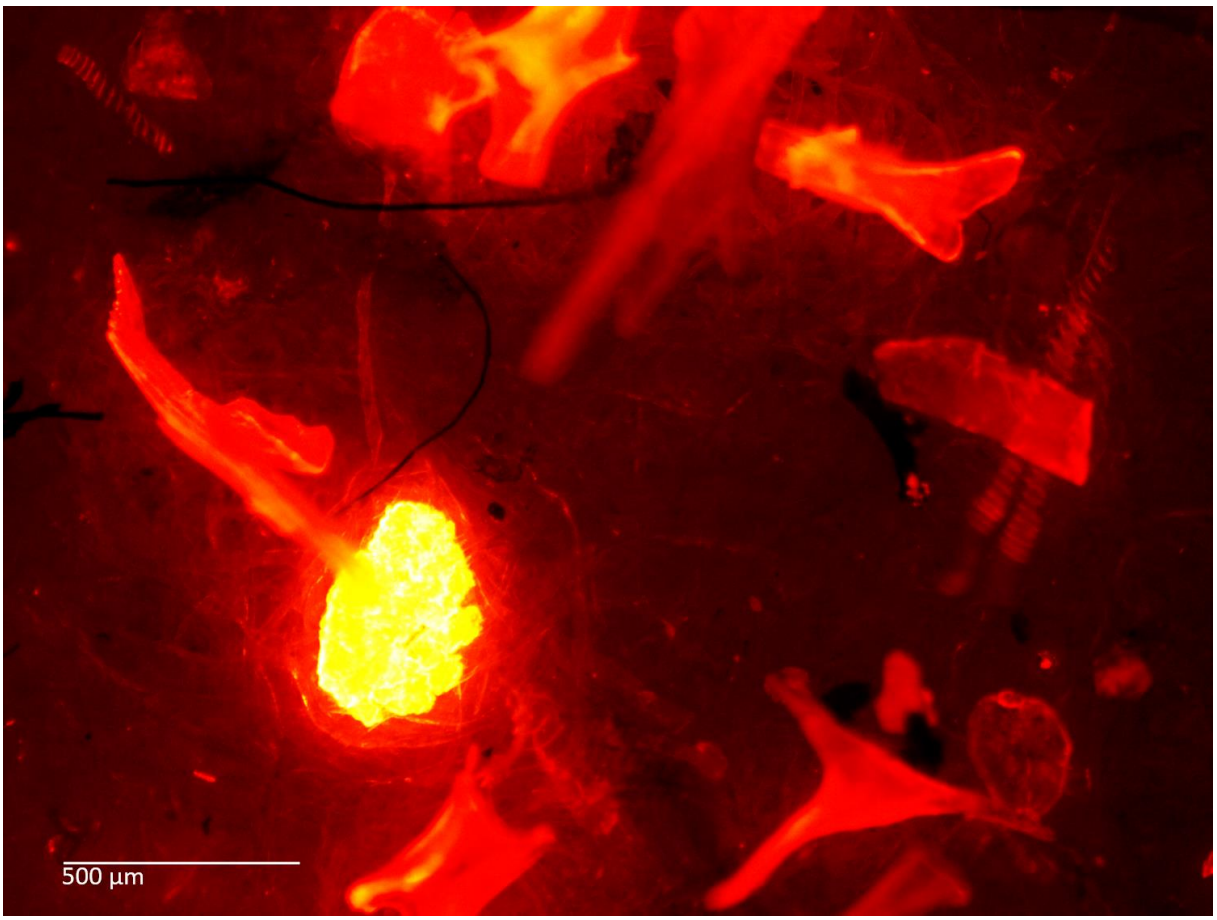


Figure S17. Countable microplastic fragment surrounded by fish bones.

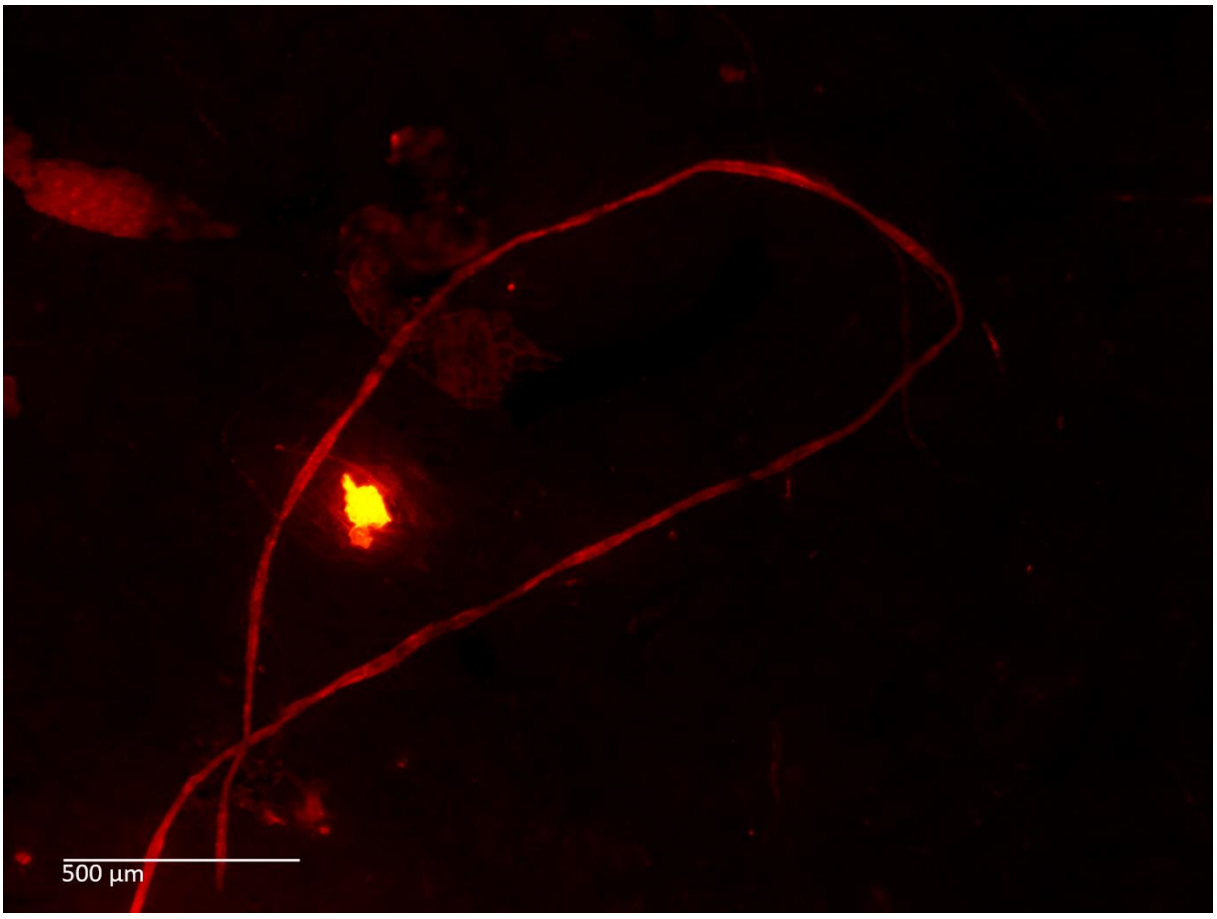
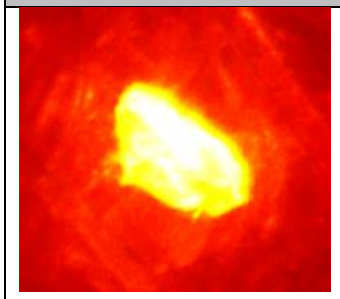
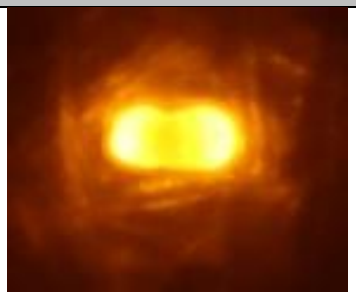
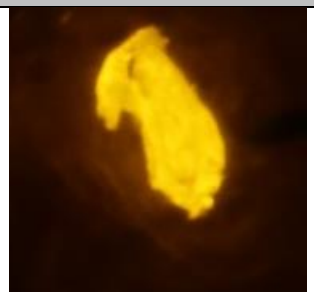
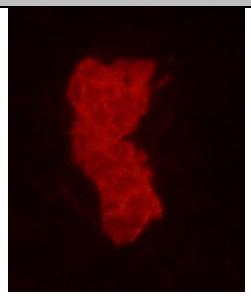
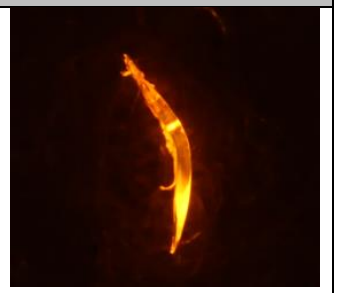


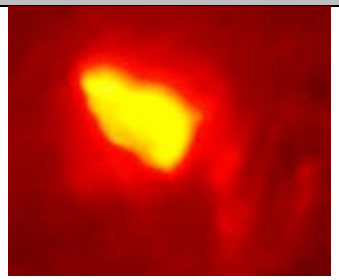
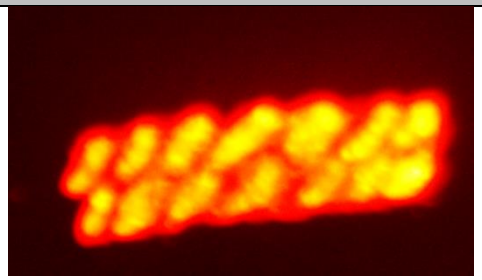
Figure S18. Countable microplastic fragment surrounded by a cellulose fibre.

Categorization of parameters:

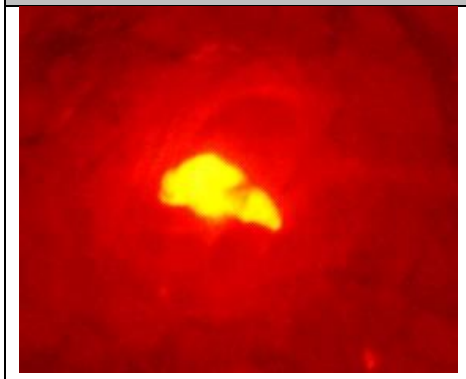
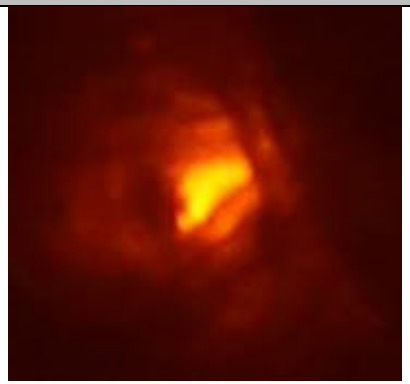
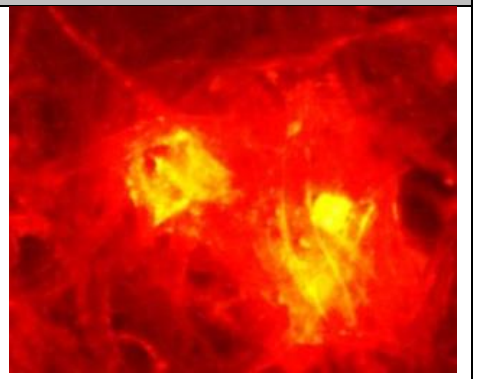
Fluorescence intensity

White	**White yellow**	**Yellow**	**Orange**	**Red**	**Multicolour**
Most parts bright whitish	Predominantly yellow with white spots	Predominantly yellow	Predominantly orange	Predominantly red	More than one colour
					

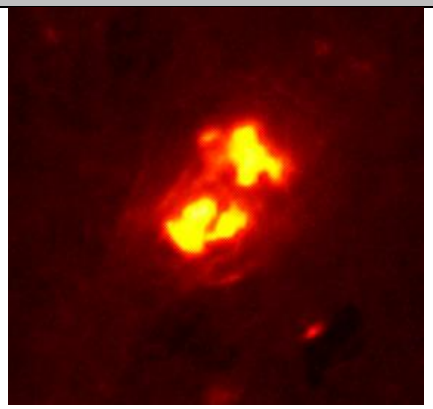
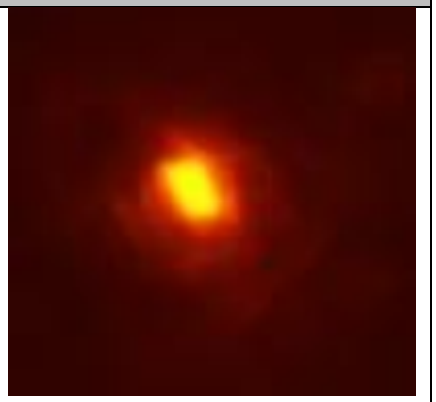
Surface

Plain: No shadows and structures visible	**Irregular:** Regular structures and different colours visible
	

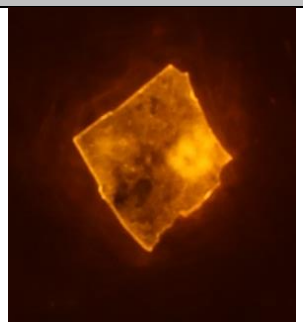
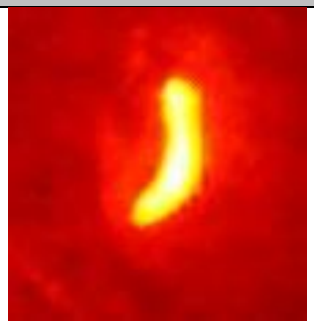
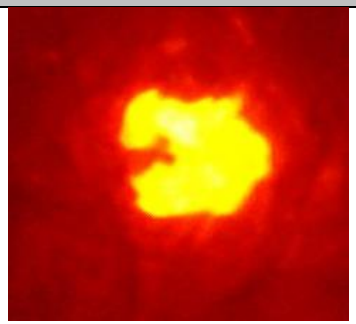
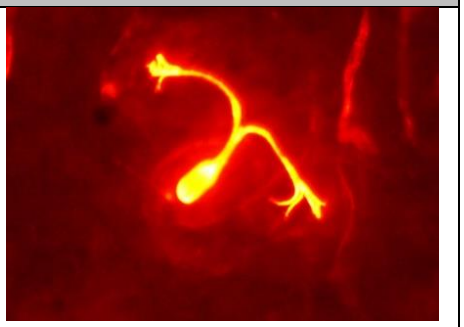
Appearance

Self-contained	Blurred	Melted
		

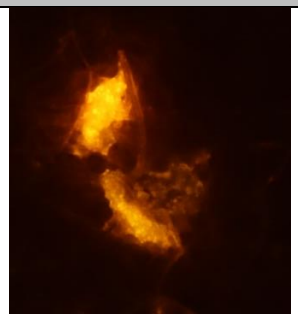
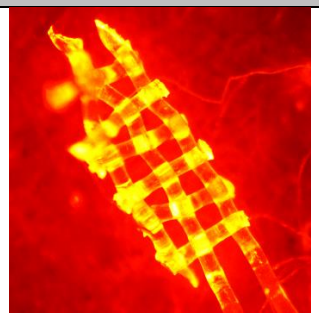
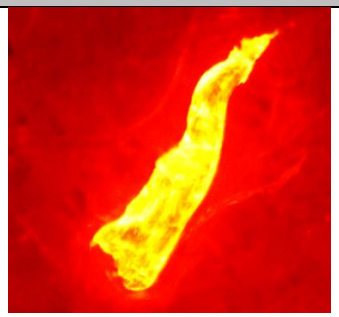
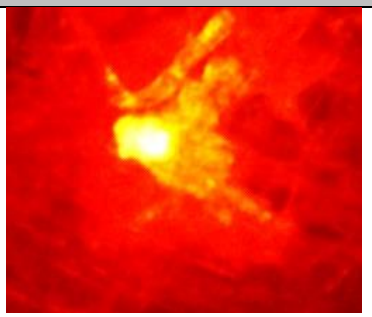
Completeness

Scattered	In one piece / undamaged
More than one part but clearly one particle	Particle in one part
	

Surroundings

Sharp-edged: Clear boundaries and separated from filter structure	**No notches:** Particle complete without any coves	**Notched:** Clear coves available	**Fringed:** Particle with extensions
			

Organic matter

Yes	**No**	**Possible**	**Not available**
Structure reminds of known organic material	Regular structure as usually occurs with synthetic polymers	Both (Yes/No) can't be excluded	Structure not identifiable (also due to low resolution)
			

4 Results

All microplastic particles (≥ 100µm) found in intestinal and faecal samples of seals from German waters are shown in Table S1. Labelling is made up of species initials (Pv = *Phoca vitulina*; Hg = *Halichoerus grypus*) and sampling location (e.g. Intestine). All samples are archived in glass jars.

Faecal samples archived in plastic bags since 2012, are named as "LDPE_Bag_2012". Faecal samples of seals (no information on the species) were halved and stored in glass jars or plastic bags for different time periods, which is also stated in the label ("Glass_3_months" or "Plastic_3_months", etc.). The column "identified polymer structures" gives information on the identified polymers regardless the number of particles analysed. NA (no information available) refers to samples in which no particles could be further identified by µRaman spectroscopy.

The findings of fragments and fibres, in addition to the identified polymer structure should only give information on the usefulness of the applied and presented protocol.

Table S1. Results of microplastic analysis.

Sample	**total amount of particles (≥ 100 µm)**	**amount of fibres**	**amount of fragments**	**identified polymers**
Pv_Intestine_1	**25**	6	19	PE
Pv_Intestine_2	**14**	3	11	EVA, PE
Pv_Intestine_3	**90**	35	55	na
Pv_Intestine_4	**19**	6	13	PET
Pv_Intestine_5	**17**	4	13	na
Hg_Intestine_1	**20**	3	17	PE, PET, PP, PA
Hg_Intestine_2	**13**	4	9	na
Hg_Intestine_3	**6**	0	6	na
Hg_Intestine_4	**25**	4	21	PET
Hg_Intestine_5	**26**	5	21	na
LDPE Bag_2012_1	**17**	4	13	na

LDPE Bag_2012_2	**30**	5	25	na
LDPE Bag_2012_3	**7**	1	6	na
LDPE Bag_2012_4	**20**	3	17	na
LDPE Bag_2012_5	**29**	8	21	na
Glas_3 months_1	**15**	3	12	PE
Plastic_3 months_1	**22**	3	19	na
Glas_3 months_2	**8**	1	7	PE
Plastic_3 months_2	**26**	6	20	na
Glas_3 months_3	**12**	4	8	na
Plastic_3 months_3	**34**	3	31	na
Glas_6 months_1	**8**	1	7	na
Plastic_6 months_1	**33**	12	21	na
Glas_6 months_2	**10**	6	4	na
Plastic_6 months_2	**18**	7	11	PE, EVA
Glas_6 months_3	**14**	6	8	na
Plastic_6 months_3	**13**	6	7	na

Glas_12 months_1	**12**	2	13	na
Plastic_12 months_1	**12**	3	9	na
Glas_12 months_2	**11**	3	8	na
Plastic_12 months_2	**17**	4	13	EVA
Glas_12 months_3	**10**	5	5	na
Plastic_12 months_3	**14**	1	13	PE, PET