DO TERRESTRIAL ORGANISMS, ISOPODS *PORCELLIO SCABER* AND EARTHWORMS *EISENIA ANDREI*, AVOID MICROPLASTIC CONTAMINATED SOIL?

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INTRODUCTION AND AIM: Microplastics can potentially enter the terrestrial environment by fragmentation of plastic wastes, via microplastic contaminated sewage sludge deposition on agricultural land, or as a result of extreme agricultural praxes where plastic bags are used as mulch. Despite the potential presence of microplastics in terrestrial environments, data regarding the effects of microplastic on terrestrial organisms are very scarce. In this study, we investigated if terrestrial isopods *Porcellio scaber* and earthworms *Eisenia andrei* avoid soil contaminated with microplastic. Namely, by avoiding contaminated soil their habitat function is reduced.

METHODS:

***MICROPLASTICS*:** facial scrub (microbeads) (**Fig .1**) and plastic bag (**Fig. 2**)

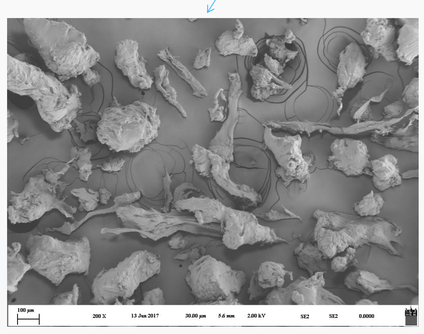

Fig 1. Microbeads from facial crub. Average diameter cca. 200-300 um)

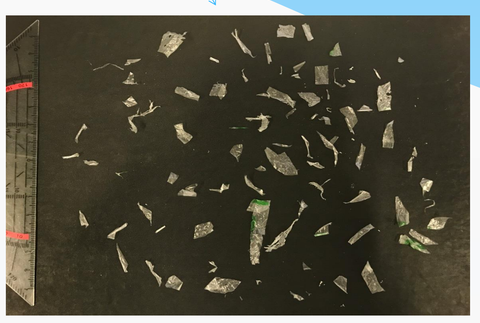

Fig 2. Median diameter value 4 mm; 60% of particles below 5 mm.

ISOPODS:

Test with isopods were done according to [1]. Microplastics (final concentration 4 mg/g dry weight) was mixed with soil and moistened with dH_2O to reach a final moisture content equivalent to 40% of the water holding capacity of the Lufa 2.2 soil. One vessels was filled with 20 g of moist soil microplastics mixture, and the other with clean soil (**Fig. 3**). In individual test the soil selection test was run with 10 replicates per treatment, each replicate contained 1 animal. The group exposure was done in 5 replicates per treatment, each replicate contained 10 animals. In individual exposures the vessels were monitored 10 times at regular time intervals and the location of the individual animal was recorded (control vs. mixture side). In group experiments the number of animals on each side was recorded after 48 h of exposure only.

EARTHWORMS:

The earthworm avoidance test was performed in accordance with the ISO 7512-1:2008 [2]. Rectangular polypropylene vessels with a volume of 1.5 L were divided into two equal sections by a vertically introduced divider (**Fig. 3**). One half of the vessel was filled with the contaminated soil (4 mg MP/g dry weight) and the other with control soil. The exposure was done in 3 replicates per treatment, each replicate contained 10 animals. At the end of the test period (48 h) the number of worms in each section was counted.

Fig. 3: Experimental set- up with isopods (left) and earthworms (right).

RESULTS: equal share of animals was found in microplastic contaminated soil and uncontaminated soil. No avoidance response of isopods or earthworms toward microplastic contaminated soil was found (**Fig. 4**).

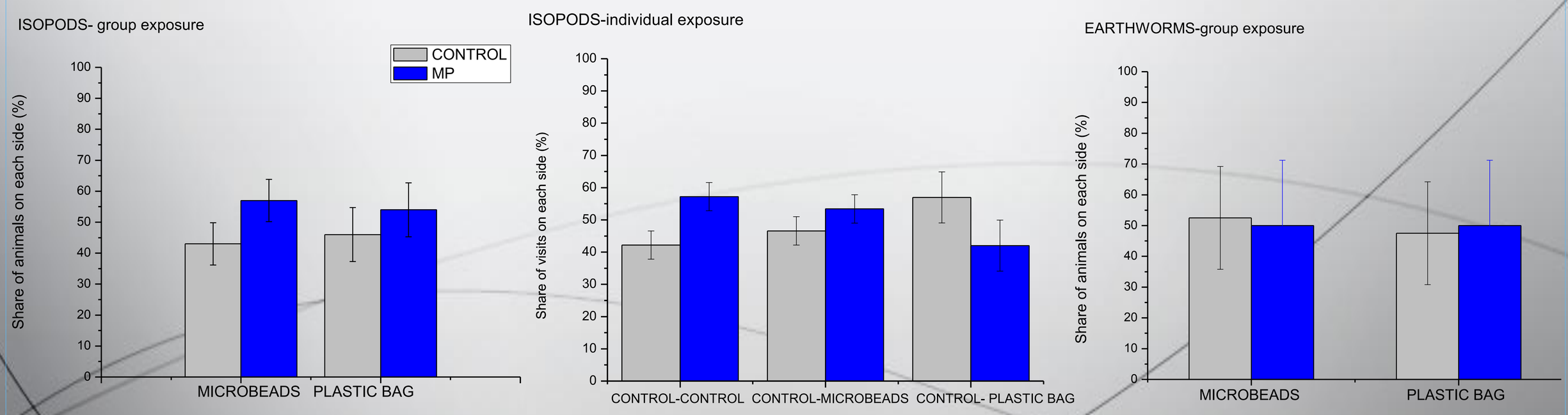

Fig. 4: Results of soil selection tests with isopods (left) and earthworms (right). Mean values and SD are shown. For isopods, results are shown for individual and group exposures. MP-microplastic.

CONCLUSIONS: Our results revealed no avoidance response of isopods and earthworms toward microplastic contaminated soil. It remains to be investigated how longer exposures to microplastic would affect the behaviour of terrestrial organisms. Also it is of interest how environmentally aged microplastic (e.g. coated with biofilm) would affect the organisms. Knowledge in this field is important to assess the potential hazard of microplastic released to soil.

REFERENCES:

1. Škarková, P, Kos, M, Drobne, D, Vávrová, M., Jemec Kokalj A., 2016. Effects of food salinization on terrestrial crustaceans *Porcellio scaber*. Applied Soil Ecology 100, 1-7.
2. ISO 17512-1:2008. Soil quality -- Avoidance test for determining the quality of soils and effects of chemicals on behaviour -- Part 1: Test with earthworms (*Eisenia fetida* and *Eisenia andrei*)

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