

Supplementary Material

Title: Microplastic fiber and drought effects on plants and soil are only slightly modified by arbuscular mycorrhizal fungi

Running title: AM fungi, microplastic and drought

Anika Lehmann^{1,2,†,*}, **Eva F. Leifheit**^{1,2,*}, **Linshan Feng**¹, **Joana Bergmann**^{1,2,3}, **Anja Wulf**¹, **Matthias C. Rillig**^{1,2}

¹ Freie Universität Berlin, Institut für Biologie, 14195 Berlin, Germany;

² Berlin-Brandenburg Institute of Advanced Biodiversity Research, 14195 Berlin, Germany

³ Leibniz Centre for Agricultural Landscape Research (ZALF), Müncheberg, Germany

† Correspondence: lehmann.anika@googlemail.com; Tel.: +49-30-838-53145

* These authors contributed equally to this work.

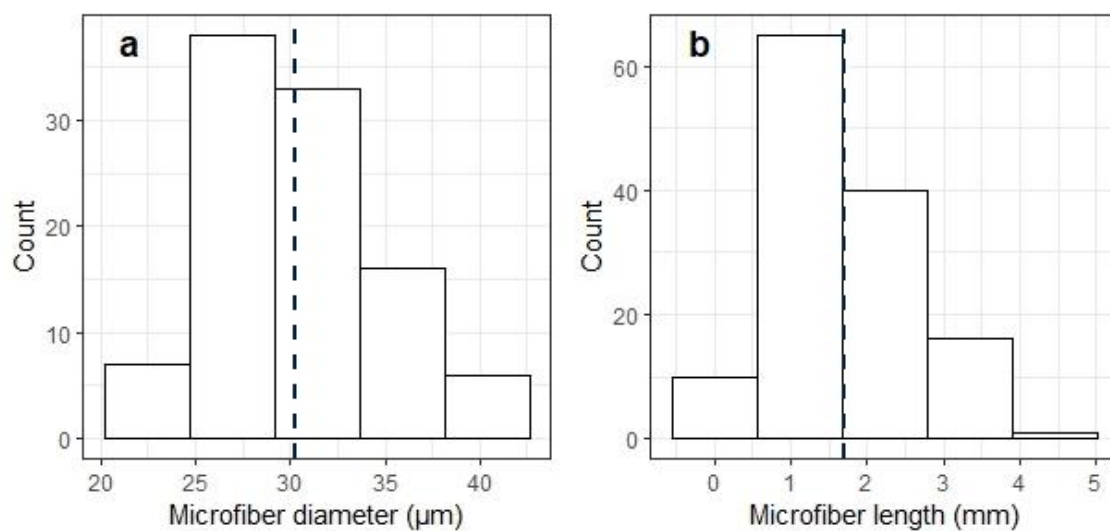


Fig. S1. Histograms depicting the diameter and length distribution of manually processed polyester microfibers. On average, fiber fragments had a diameter of $30\mu\text{m}$ and a length of 1.7mm .

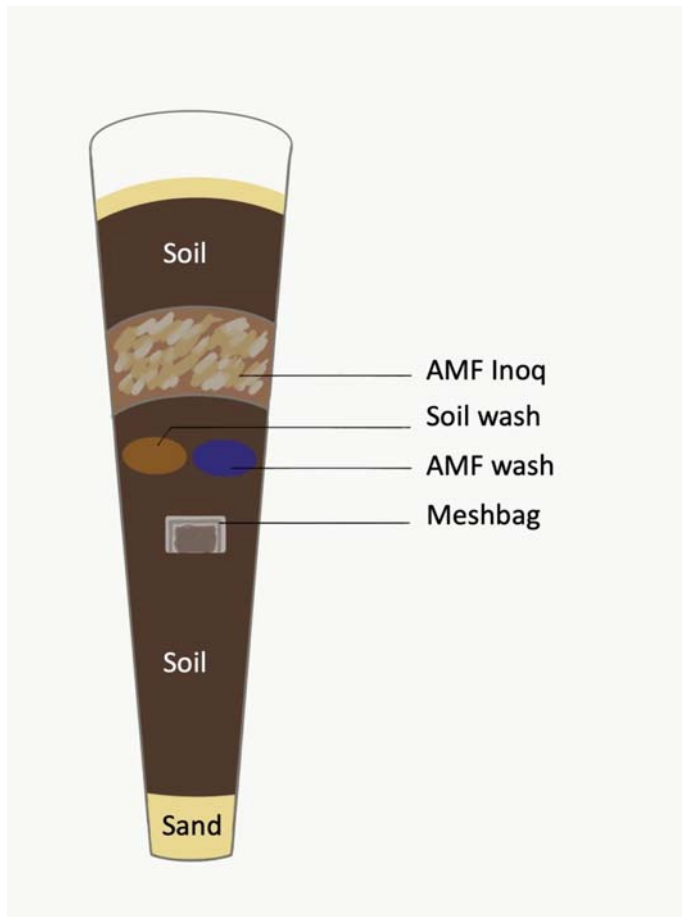


Fig. S2 Set-up of the growth systems used in our experiment. At the bottom a layer of sand covered a sheet of black gardening mesh. The pots were filled with 150g of soil-sand mixture in which we embedded a tea filled mesh bag (with 5mm distance to the pot wall) and added a soil and microbial wash and the AM fungal inoculum band.

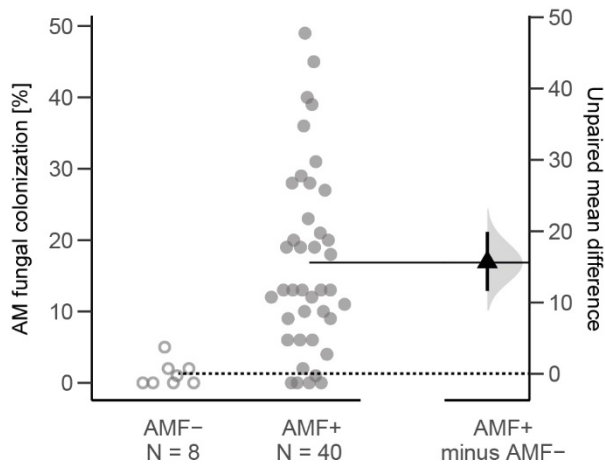


Fig. S3 Colonization of onion roots by AM fungal structures in %. A subset of 8 samples was tested for the controls. Raw data are depicted via swarmplot, while the difference between the treatment levels (AMF present – yes/no; AMF+ depicted by filled and A- with empty circles) is presented by unpaired mean differences. When the CI is not overlapping zero then a higher colonization of the respective AM fungal structure was detected in the AMF+ treatment (depicted by an upward oriented arrow head).

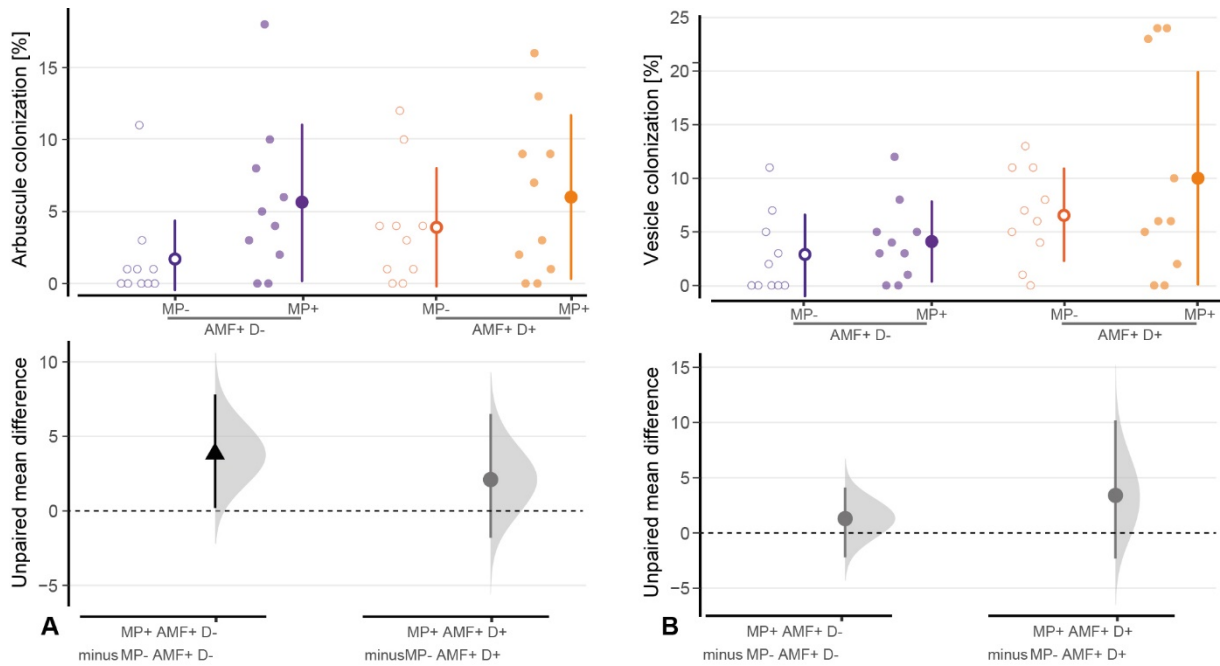


Fig. S4. Response of the percental portion of AM fungal structures (arbuscules (A) and vesicles (B)) to application of microfibers, AM fungi and drought. Data distributions are aligned with corresponding mean and standard deviation ($n=10$ for each group). Blueish colors represent well-watered and reddish drought conditions. Empty circles depict absence and filled circles presence of microfiber contamination. The multi-group estimation plots (second row) depict the unpaired mean differences of the microplastic- and AM fungal-only and microplastic-AM fungal combination under well-watered and drought scenario. Samples without AM fungi and microplastic were used as control group for well-watered and drought conditions, separately. Circles and triangles represent the effect size mean (unpaired mean; effect magnitude) and the vertical lines the corresponding confidence intervals (effect precision). The sampling error distribution is presented as a grey curve. Positive (arrow head up) or negative (arrow head down) effect sizes and corresponding CIs of treatment compared to control group are depicted in black while neutral effects (circle) are colored in grey; neutral effects occur when the CIs overlap the dashed zero line (line of no effect). Model outcomes of raw data analyses are presented in Table S1.

Table S1. Outcomes of the generalized least square models for percentage root colonization over all structures and specifically for vesicles and arbuscules (each in %, n=40) for the treatments (AMF= arbuscular mycorrhizal fungi, MP= microplastic, D= drought). P < 0.05 was considered significant and marked in bold. Degrees of freedom (df), F- and p-value for each variable are presented.

Treatments	Vesicles		Arbuscules		AMF colonization		
	df	F-value	p-value	F-value	p-value	F-value	p-value
MP	1, 36	1.24258	0.2724	4.42132	0.0425	2.93962	0.095
Drought	1, 36	5.86629	0.0206	1.18144	0.2843	1.6576	0.2061
MP:Drought	1, 36	0.11646	0.7349	1.00667	0.3224	0.66364	0.4206

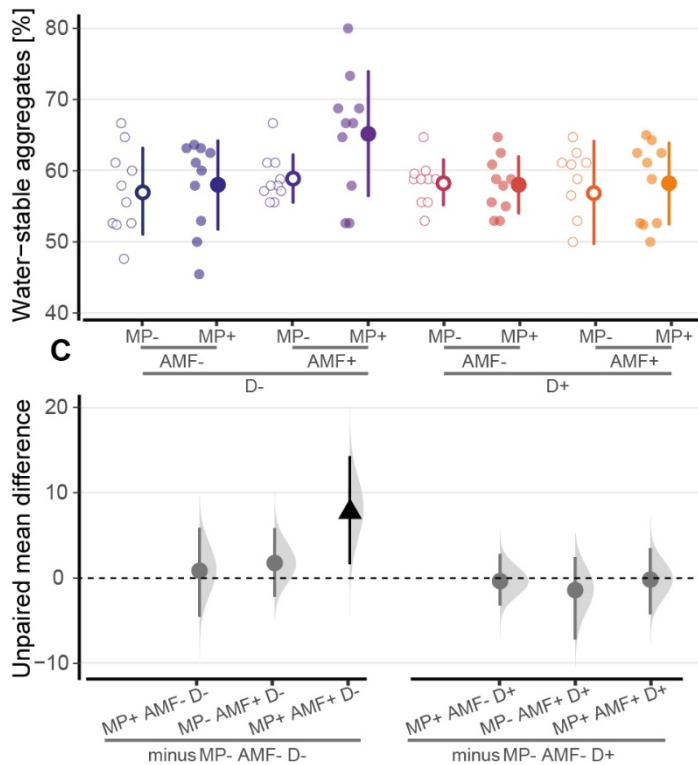


Fig. S5 Water-stability of soil aggregates in % as affected by the application of microfibers, AM fungi and drought. Data distributions are aligned with corresponding mean and standard deviation (n=10 for each group). Blueish colors represent well-watered and reddish drought conditions. Empty circles depict absence and filled circles presence of microfiber contamination. The multi-group estimation plots (second row) depict the unpaired mean differences of the microplastic- and AM fungal-only and microplastic-AM fungal combination under well-watered and drought scenario. Samples without AM fungi and microplastic were used as control group for well-watered and drought conditions, separately. Circles and triangles represent the effect size mean (unpaired mean; effect magnitude) and the vertical lines the corresponding confidence intervals (effect precision). The sampling error distribution is presented as a grey curve. Positive (arrow head up) or negative (arrow head down) effect sizes and corresponding CIs of treatment compared to control group are depicted in black while neutral effects (circle) are colored in grey; neutral effects occur when the CIs overlap the dashed zero line (line of no effect). Model outcomes of raw data analyses are presented in Table S2.

Table S2. Outcomes of the generalized least square models for water-stable aggregates (WSA in %, n=80) for the treatments (AMF= arbuscular mycorrhizal fungi, MP= microplastic, D= drought). Degrees of freedom (df), F- and p-value for each variable are presented.

Treatments	WSA		
	df	F-value	p-value
AMF	1, 70	1.244	0.2685
MP	1, 70	1.758	0.1891
D	1, 70	2.157	0.1463
AMF : MP	1, 70	1.495	0.2254
AMF : D	1, 70	3.805	0.055
MP : D	1, 70	1.446	0.2331
AMF : MP : D	1, 70	0.541	0.4643