

Electronic Supplementary Information

Effect of emerging contaminants on soil microbial community composition, soil enzyme activity, and strawberry plant growth in polyethylene microplastic-containing soils

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Table S1. Chemical contaminants stock solution.

Compound	Mass [mg]
acetaminophen	14.8
caffeine	41.1
carbamazepine	69.8
gemfibrozil	30.0
ibuprofen	100.5
sulfamethoxazole	677.6
sulfanilamide	199.3
triclosan	1493.5
acetone	15831.8

Table S2. Spiked chemical contaminant concentration [mg per kg fresh soil] in C and CP100 pots and their octanol-water partition coefficient ($\log K_{ow}$), molar mass, and chemical structure.

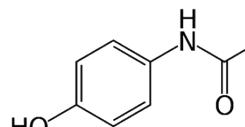
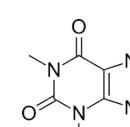
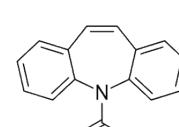
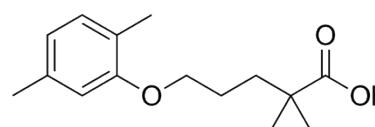
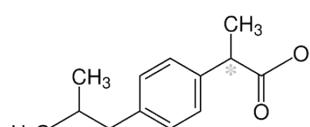
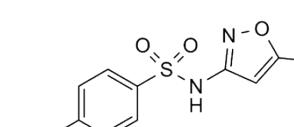
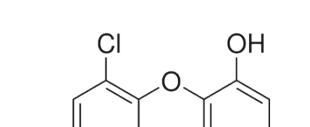
Compound	Concentration [mg/kg]	$\log K_{ow}$	Molar mass [g/mol]	Chemical structure
acetaminophen	0.03	0.46	151.2	
caffeine	0.09	-0.07	194.2	
carbamazepine	0.15	2.45	236.3	
gemfibrozil	0.06	4.24	250.3	
ibuprofen	0.21	3.97	206.3	
sulfamethoxazole	1.42	0.89	253.3	
sulfanilamide	0.42	-0.62	172.2	
triclosan	3.14	4.76	289.5	

Table S3. The composition of the fertilizer used on soils.

Concentrate	Fertilizer	Concentration [mg/L]
A	CaNO ₃	7.1
	NH ₄ NO ₃ (34%)	0.375
	Mg(NO ₃) ₂	3.62
	Fe (EDTA)	0.18
B	monopotassium phosphate (MKP)	1.6
	K ₂ SO ₄	1.87
	KNO ₃	0.62
C	Mn(SO ₄) ₂ (32.5%)	0.115
	ZnSO ₄ (23%)	0.031
	Borax (11.3%)	0.058
	CuSO ₄ (24%)	0.002
	Mo (39.6%)	0.011

Table S4. Detection parameters of chemical contaminants via LC/MS. Recovery was measured at 50 ng/mL (N = 2). RT = retention time.

Compound	Quantifier [m/z]	Ionization	RT [min]	Detection limits [ng/mL]	r ²	Recovery [mean ± SD]
acetaminophen	152.07116	[M+H] ⁺	2.970	0.026	0.9948	101 ± 1.2%
caffeine	195.08822	[M+H] ⁺	3.128	0.151	0.9988	82 ± 0.7%
carbamazepine	237.1028	[M+H] ⁺	4.337	0.066	0.9943	105 ± 0.6%
gemfibrozil	249.1491	[M-H] ⁻	5.195	0.678	0.9995	98 ± 3.0%
ibuprofen	205.12287	[M-H] ⁻	5.013	0.788	0.9983	70 ± 5.4%
sulfamethoxazole	254.05995	[M+H] ⁺	3.666	0.031	0.9989	89 ± 0.1%
sulfanilamide	173.03849	[M+H] ⁺	2.614	0.580	0.9989	122 ± 7.6%
triclosan	286.94335	[M-H] ⁻	5.211	0.212	0.9986	118 ± 0.2%

Table S5. Enzyme and substrate pairings. (MUB = 4-methylumbelliferyl)

Enzyme	EC number	Substrate
β -glucosidase	3.2.1.21	MUB β -D-glucopyranoside
chitinase	3.2.1.14	MUB N-acetyl- β -D-glucosaminide
xylan 1,4- β -xylosidase	3.2.1.37	MUB- β -D-xylopyranoside
phosphatase	3.1.3.x	MUB phosphate (free acid)

Table S6. Weighted UniFrac distance of the soil microbial communities' amplicon sequence variants pairwise PERMANOVA results for control (CTRL), 10 mg HDPE microplastic kg⁻¹ soil (P10), 100 mg HDPE microplastic kg⁻¹ soil (P100), chemical contaminants + 100 mg HDPE microplastic kg⁻¹ soil (CP100), and chemical contaminants (C) treated soils. PRE (pre-treatment) is a composite sample of the uncontaminated soils. q < 0.05 is significant.

Group 1	Group 2	Sample size	Permutations	pseudo-F	p-value	q-value
C	CP100	30	1000000	1.95	0.0110	0.0329
C	CTRL	30	1000000	2.55	0.0020	0.0076
C	P10	30	1000000	1.76	0.0265	0.0517
C	P100	30	1000000	3.06	0.0003	0.0050
C	PRE	18	1000000	3.70	0.0012	0.0062
CP100	CTRL	30	1000000	1.31	0.1366	0.1862
CP100	P10	30	1000000	1.26	0.1636	0.2036
CP100	P100	30	1000000	1.64	0.0315	0.0517
CP100	PRE	18	1000000	2.88	0.0012	0.0062
CTRL	P10	30	1000000	0.58	0.9699	0.9699
CTRL	P100	30	1000000	1.02	0.3933	0.4214
CTRL	PRE	18	1000000	1.92	0.0330	0.0517
P10	P100	30	1000000	1.23	0.1764	0.2036
P10	PRE	18	1000000	2.02	0.0282	0.0517
P100	PRE	18	1000000	1.95	0.0345	0.0517

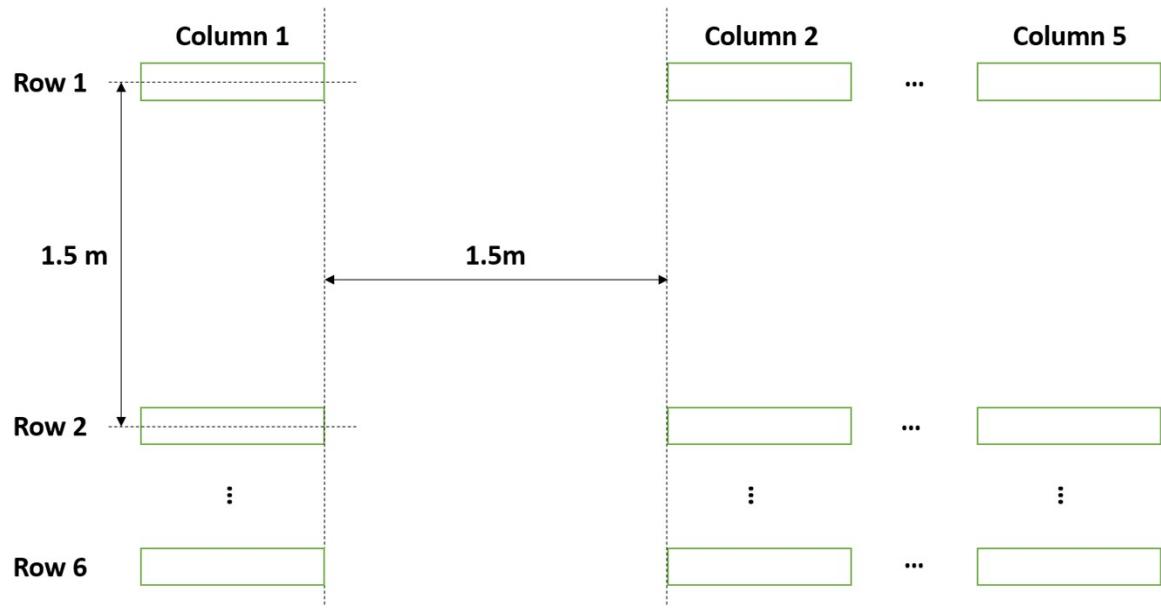


Figure S1. Top-down view of outdoor layout of strawberry pots (green rectangles).

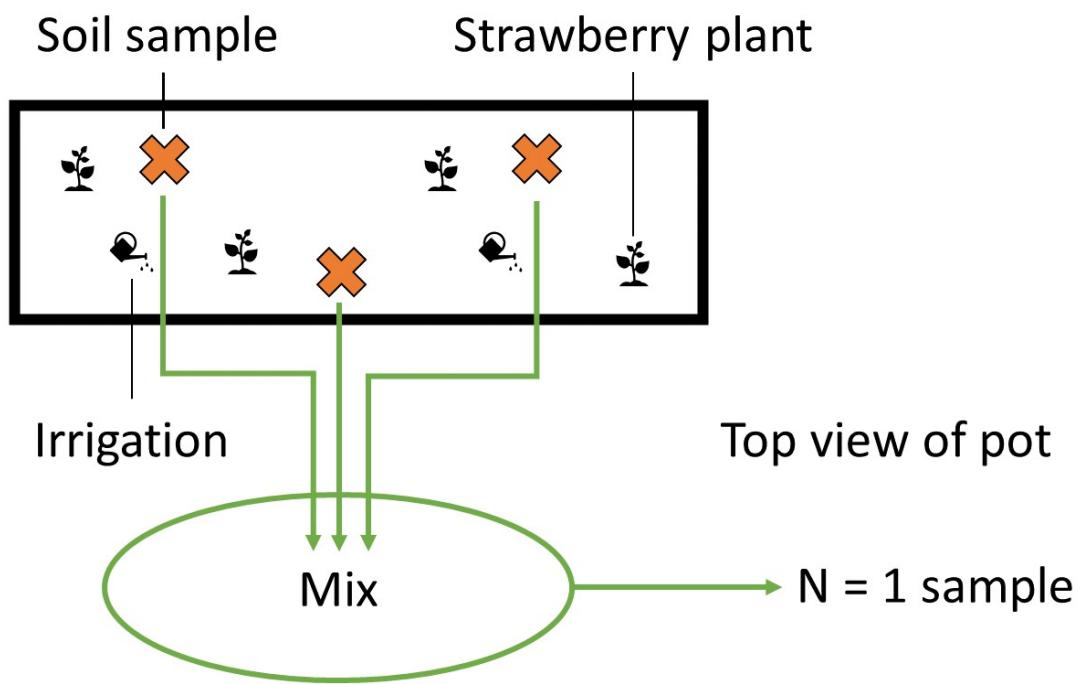


Figure S2. Approximate location of strawberry plants, irrigation, and soil sampling in each pot.

The mixing was performed in an aluminum tray near the pot, before being transferred to glassware for transportation and further analysis in a lab setting.

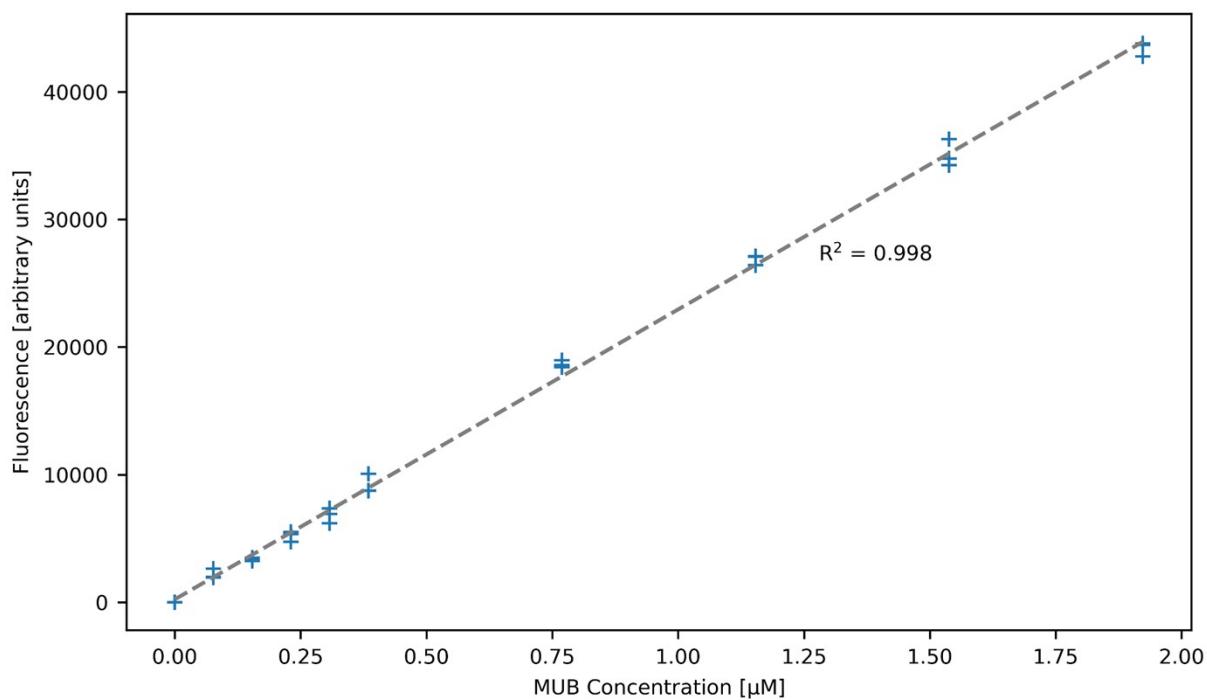


Figure S3. Fluorescence of 4-methylumbelliflone (MUB) in buffer at various concentrations and after NaOH addition. This plot shows that the fluorescence of MUB is linearly correlated to its concentration when the MUB concentration is below 1.92 μM . The highest concentration shown here is equivalent to Well C in Table S7.

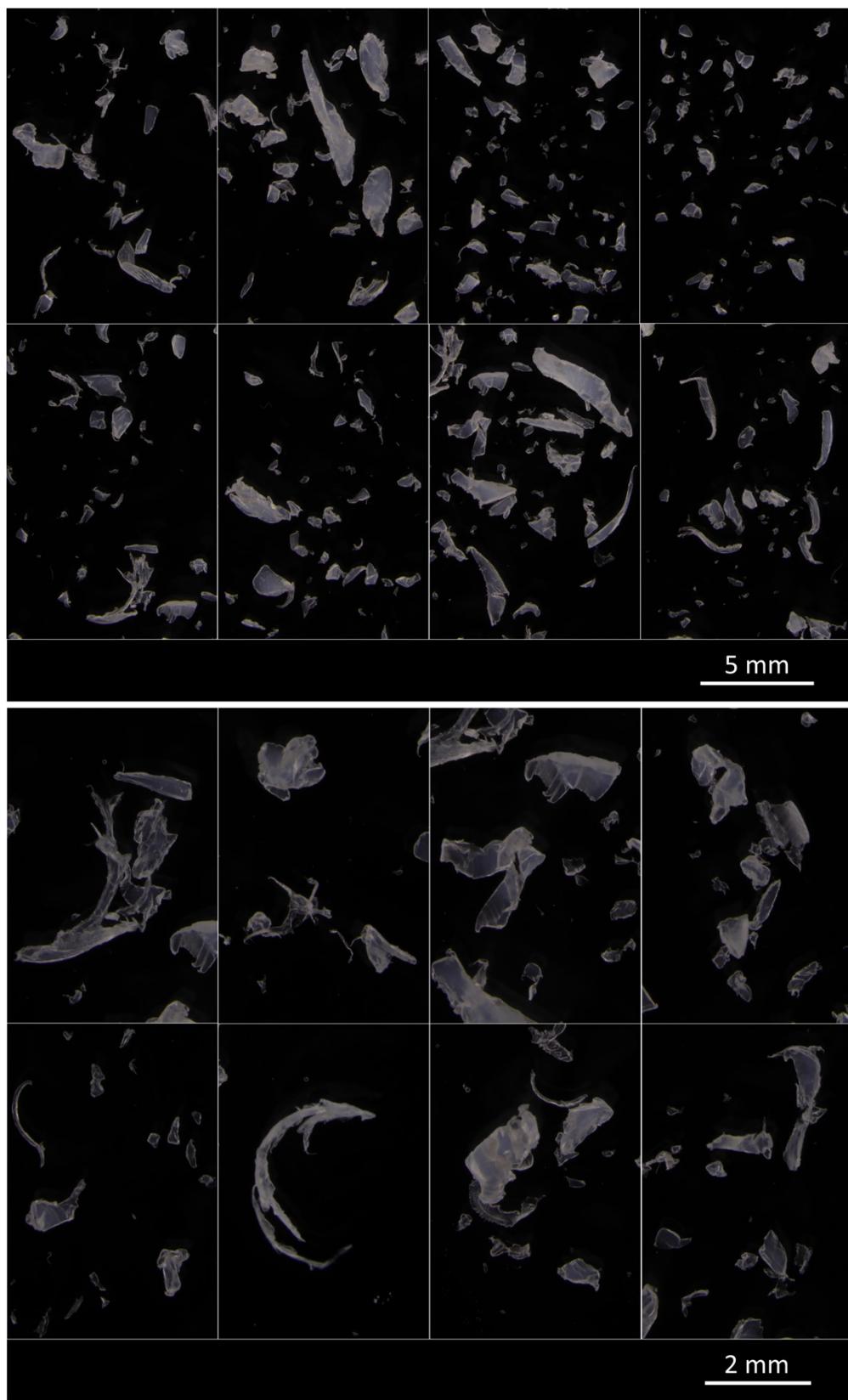


Figure S4. Images of HDPE microplastics taken under a stereomicroscope in dark-field mode.

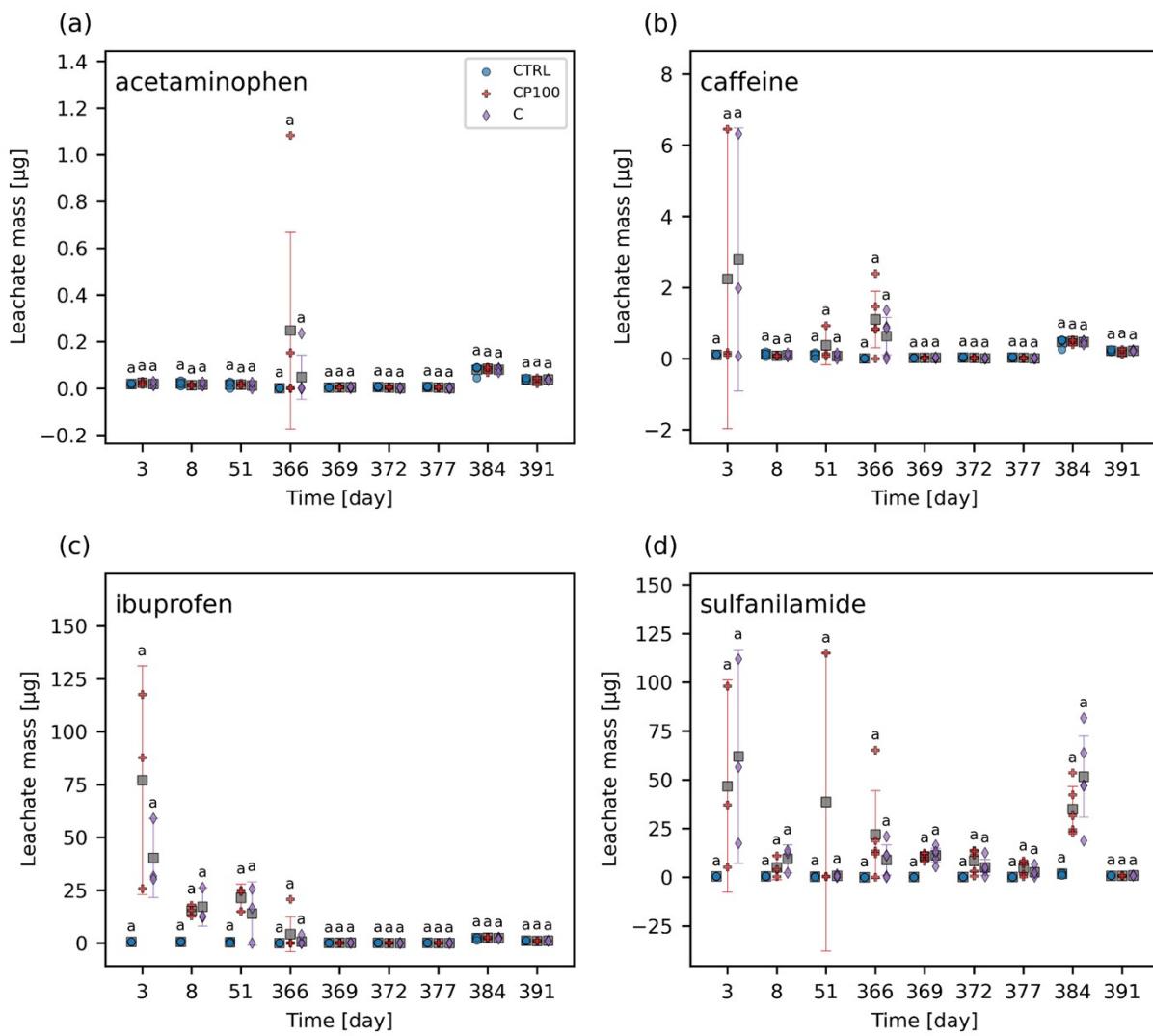


Figure S5. Leachate mass data for (a) acetaminophen, (b) caffeine, (c) ibuprofen, and (d) sulfanilamide in control (CTRL), chemical contaminants + 100 mg HDPE microplastic kg^{-1} soil (CP100), and chemical contaminants (C) treated soils. No leachate data was collected for the intervals including day 52 – 363, 392 – 456. For each timepoint, treatments with the same letter have no significant difference between them. Markers represent individual samples. Grey squares represent the means. Error bars show $2 \times$ the standard error of the mean.

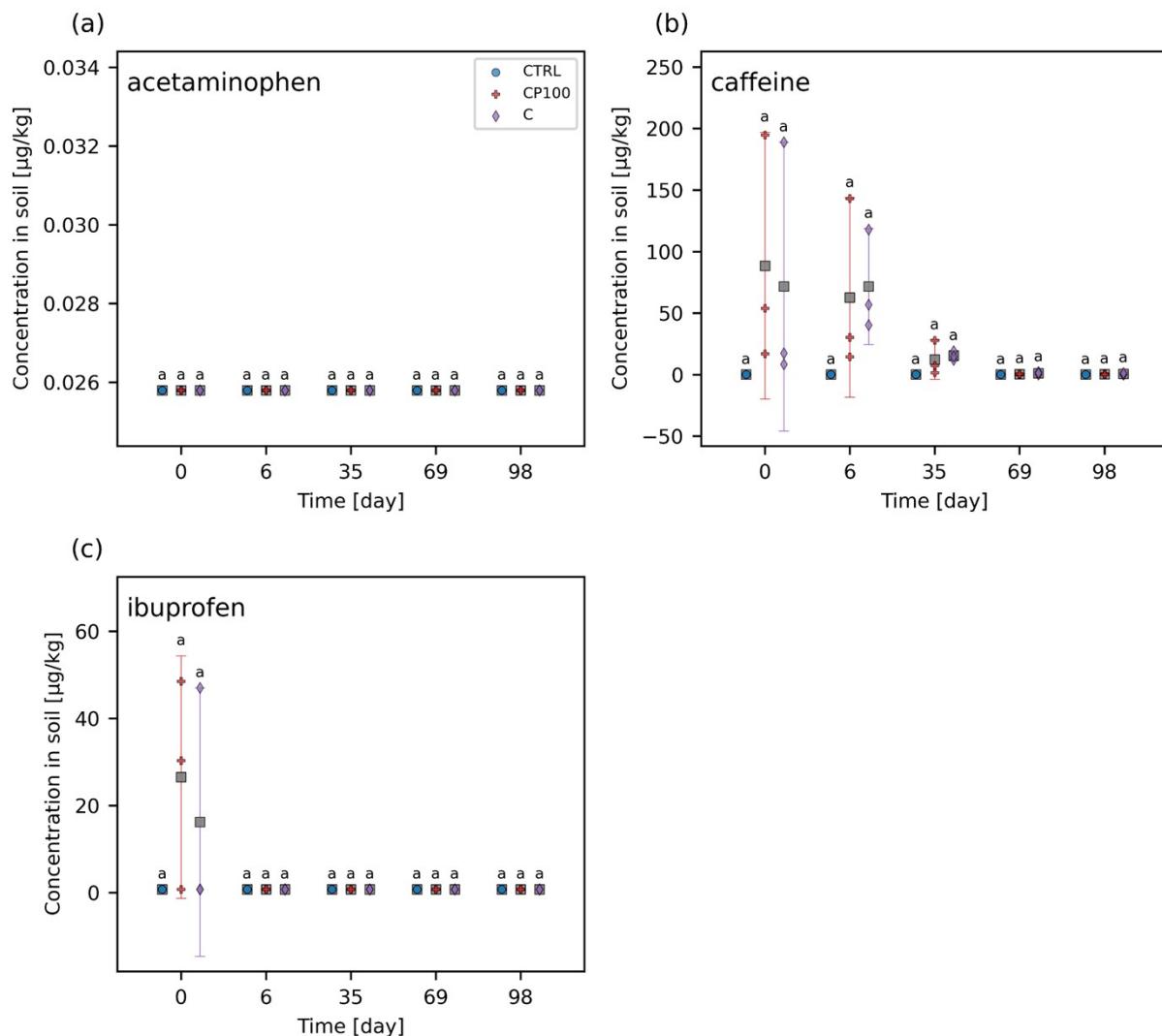


Figure S6. Concentration of (a) acetaminophen, (b) caffeine, and (c) ibuprofen in control (CTRL), chemical contaminants + 100 mg HDPE microplastic kg⁻¹ soil (CP100), and chemical contaminants (C) treated soils. For each timepoint, treatments with the same letter have no significant difference between them. Markers represent individual samples. Grey squares represent the means. Error bars show 2 × the standard error of the mean.

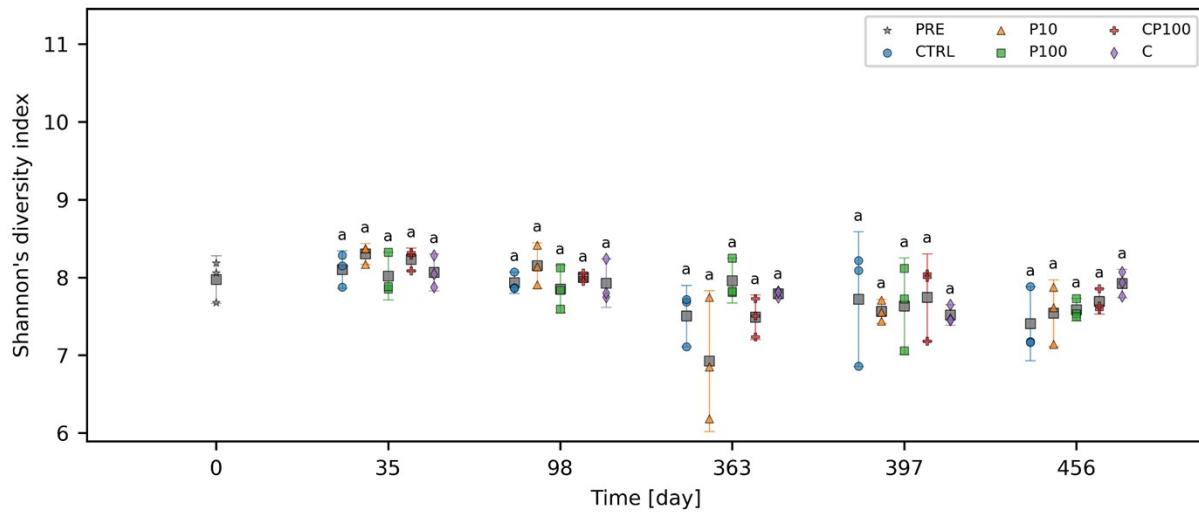


Figure S7. Shannon's diversity index of control (CTRL), 10 mg HDPE microplastic kg⁻¹ soil (P10), 100 mg HDPE microplastic kg⁻¹ soil (P100), chemical contaminants + 100 mg HDPE microplastic kg⁻¹ soil (CP100), and chemical contaminants (C) treated soils. PRE (pre-treatment) is a composite sample of the uncontaminated soils. For each timepoint, treatments with the same letter have no significant difference between them. No letters are present for PRE, since there were no other treatments to compare it to on day 0. Markers represent individual samples. Grey squares represent the means. Error bars show 2 × the standard error of the mean.

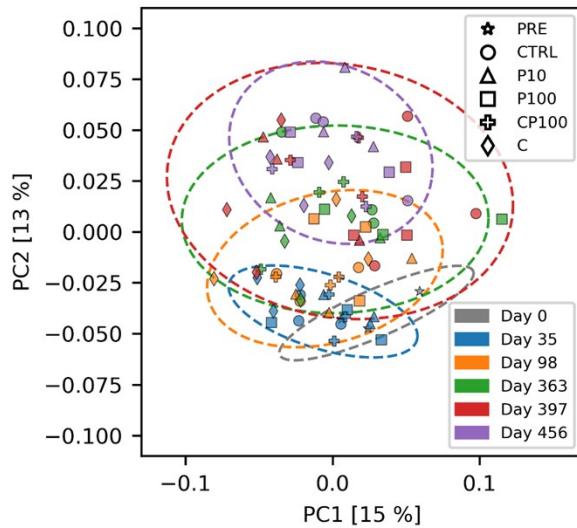


Figure S8. Principal coordinate analysis (PCoA) of the weighted UniFrac distance of the soil microbial communities' amplicon sequence variants in control (CTRL), 10 mg HDPE microplastic kg⁻¹ soil (P10), 100 mg HDPE microplastic kg⁻¹ soil (P100), chemical contaminants + 100 mg HDPE microplastic kg⁻¹ soil (CP100), and chemical contaminants (C) treated soils. PRE (pre-treatment) is a composite sample of the uncontaminated soils. Ellipses represent the zone where 95 % of samples from a bivariate normal distribution with the same mean and covariance matrix as the respective treatment would be drawn from. Numbers in brackets indicate the amount of variance captured by that principal coordinate (PC). This figure shows the same data as Figure 4, but with markers representing treatments and colour representing time to better illustrate the temporal effects on the microbial community composition.

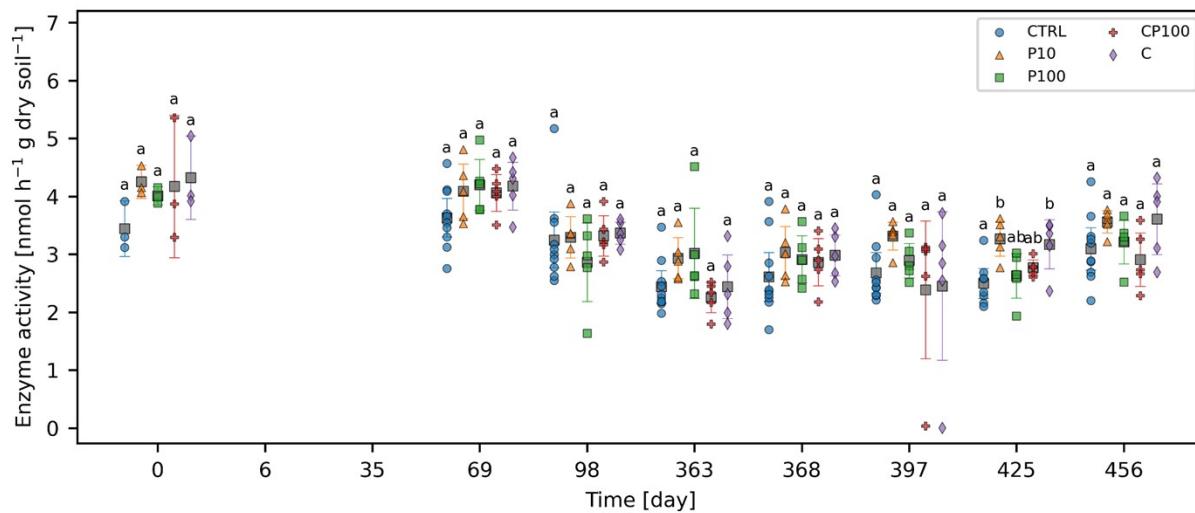


Figure S9. Phosphatase activity over the course of two growing seasons in control (CTRL), 10 mg HDPE microplastic kg⁻¹ soil (P10), 100 mg HDPE microplastic kg⁻¹ soil (P100), chemical contaminants + 100 mg HDPE microplastic kg⁻¹ soil (CP100), and chemical contaminants (C) treated soils. Day 6 and 35 data were omitted due to a potential contamination of the MUB-linked substrates. For each timepoint, treatments with the same letter have no significant difference between them. Markers represent individual samples. Grey squares represent the means. Error bars show 2 × the standard error of the mean.

Calculation of enzyme activity

Enzyme activity was calculated by measuring the fluorescence from 4-methylumbelliferon (MUB). When a 4-methylumbeliferyl-linked substrate (MUB-substrate) is exposed to soil extract, the enzymes extracted from the soil begin to catalyze the MUB-substrate, releasing MUB in the process. To isolate the fluorescence originating from the MUB, the background fluorescence and/or fluorescence quenching from the soil extract and the MUB-substrate after the 2-hour incubation period would have to be measured and subtracted. However, this is not possible, given that the MUB-substrate readily reacts in soil extract. Therefore, a quench coefficient (q) must be calculated which estimates the degree to which the soil extract quenches fluorescence. The net fluorescence from the produced MUB and the remaining unreacted MUB-substrate after the 2-hour incubation period ($F_B - F_F$) is then corrected using the quench coefficient to estimate what the fluorescence of this mixture would have been without the quenching effect of the soil extract. This value can then be subtracted by the net fluorescence of the MUB-linked substrate in buffer ($F_A - F_E$). The data is then normalized as outlined below.

Table S7. Each plate contained the following types of wells.

Well	200 µL	50 µL	10 µL	Wells per plate
A	50 mM sodium acetate buffer	200 µM MUB-substrate solution	1 M NaOH	4
B	soil extract	200 µM MUB-substrate solution	1 M NaOH	2
C	50 mM sodium acetate buffer	10 µM MUB solution	1 M NaOH	4
D	soil extract	10 µM MUB solution	1 M NaOH	2
E	50 mM sodium acetate buffer	50 mM sodium acetate buffer	1 M NaOH	4
F	soil extract	50 mM sodium acetate buffer	1 M NaOH	2

The mean fluorescence (F) from these wells was used to calculate the enzyme activity as follows:

$$q = \frac{F_D - F_F}{F_C - F_E}$$

$$e = \frac{F_C - F_E}{0.5 \text{ nmol MUB}}$$

$$E = \frac{\left(\frac{F_B - F_F}{q} - (F_A - F_E) \right)}{e \times t \times c \times v}$$

Where:

- E is the enzyme activity [(nmol MUB)/(h × (g soil))]
- q is the quench coefficient
- e is the emission coefficient [(net fluorescence) / (nmol MUB)]
- t is the reaction time (~2 h, exact value recorded from timestamps)
- c is the soil in buffer concentration (16 g soil / L buffer)
- v is the soil extract volume (200×10^{-6} L)

Qiime2 commands

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--output-path paired-end-demux.qza \
--input-format PairedEndFastqManifestPhred33V2

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--o-visualization paired-end-demux.qzv

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--p-trim-left-r 6 \
--p-trunc-len-f 244 \
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--o-denoising-stats denoising-stats.qza

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--o-masked-alignment masked-alignedrep-seqs.qza \
--o-tree unrooted-tree.qza \
--o-rooted-tree rooted-tree.qza

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--output-dir core-metrics-results

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--output-path exported-data/weighted_unifrac_pcoa_results
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```

Significance tests

Table S8. Significance testing for contaminants leachate samples found in Figures 1 and S5. HB = Holm-Bonferroni.

Dependent variable	Time	Test	Statistic	p-value	α with HB correction	Is $p < \alpha$?
Acetaminophen	3	One-way ANOVA	0.51	0.623	0.013	FALSE
Acetaminophen	8	One-way ANOVA	0.52	0.619	0.010	FALSE
Acetaminophen	51	One-way ANOVA	0.11	0.895	0.025	FALSE
Acetaminophen	366	Kruskal-Wallis H-test	1.24	0.538	0.008	FALSE
Acetaminophen	369	One-way ANOVA	0.11	0.900	0.050	FALSE
Acetaminophen	372	One-way ANOVA	4.32	0.039	0.006	FALSE
Acetaminophen	377	One-way ANOVA	4.32	0.039	0.006	FALSE
Acetaminophen	384	Kruskal-Wallis H-test	1.85	0.396	0.007	FALSE
Acetaminophen	391	One-way ANOVA	0.33	0.733	0.017	FALSE
Caffeine	3	Kruskal-Wallis H-test	1.16	> 0.1	0.025	FALSE
Caffeine	8	One-way ANOVA	0.52	0.619	0.010	FALSE
Caffeine	51	Kruskal-Wallis H-test	0.97	> 0.1	0.050	FALSE
Caffeine	366	Kruskal-Wallis H-test	4.88	0.087	0.007	FALSE
Caffeine	369	One-way ANOVA	0.11	0.900	0.017	FALSE
Caffeine	372	One-way ANOVA	4.32	0.039	0.006	FALSE
Caffeine	377	One-way ANOVA	4.32	0.039	0.006	FALSE
Caffeine	384	Kruskal-Wallis H-test	1.85	0.396	0.008	FALSE
Caffeine	391	One-way ANOVA	0.33	0.733	0.013	FALSE
Carbamazepine	3	Kruskal-Wallis H-test	5.42	0.086	0.025	FALSE
Carbamazepine	8	Kruskal-Wallis H-test	5.96	0.029	0.013	FALSE
Carbamazepine	51	Kruskal-Wallis H-test	4.36	> 0.1	0.050	FALSE
Carbamazepine	366	Kruskal-Wallis H-test	9.78	0.008	0.006	FALSE
Carbamazepine	369	Kruskal-Wallis H-test	9.98	0.007	0.006	FALSE
Carbamazepine	372	Kruskal-Wallis H-test	9.62	0.008	0.007	FALSE
Carbamazepine	377	Kruskal-Wallis H-test	9.42	0.009	0.008	FALSE
Carbamazepine	384	Kruskal-Wallis H-test	9.40	0.009	0.010	FALSE
Carbamazepine	391	Kruskal-Wallis H-test	5.96	0.029	0.017	FALSE
Gemfibrozil	3	Kruskal-Wallis H-test	5.42	0.086	0.007	FALSE
Gemfibrozil	8	One-way ANOVA	0.89	0.457	0.013	FALSE
Gemfibrozil	51	One-way ANOVA	0.11	0.895	0.025	FALSE
Gemfibrozil	366	Kruskal-Wallis H-test	2.54	0.281	0.008	FALSE
Gemfibrozil	369	One-way ANOVA	0.11	0.900	0.050	FALSE
Gemfibrozil	372	One-way ANOVA	4.32	0.039	0.006	FALSE
Gemfibrozil	377	One-way ANOVA	4.32	0.039	0.006	FALSE
Gemfibrozil	384	Kruskal-Wallis H-test	1.85	0.396	0.010	FALSE
Gemfibrozil	391	One-way ANOVA	0.33	0.733	0.017	FALSE
Ibuprofen	3	Kruskal-Wallis H-test	5.60	0.050	0.007	FALSE
Ibuprofen	8	Kruskal-Wallis H-test	5.42	0.086	0.008	FALSE
Ibuprofen	51	Kruskal-Wallis H-test	3.47	> 0.1	0.050	FALSE

Ibuprofen	366	Kruskal-Wallis H-test	1.24	0.538	0.013	FALSE
Ibuprofen	369	One-way ANOVA	0.11	0.900	0.025	FALSE
Ibuprofen	372	One-way ANOVA	4.32	0.039	0.006	FALSE
Ibuprofen	377	One-way ANOVA	4.32	0.039	0.006	FALSE
Ibuprofen	384	Kruskal-Wallis H-test	1.85	0.396	0.010	FALSE
Ibuprofen	391	One-way ANOVA	0.33	0.733	0.017	FALSE
Sulfamethoxazole	3	Kruskal-Wallis H-test	5.60	0.050	0.025	FALSE
Sulfamethoxazole	8	Kruskal-Wallis H-test	6.49	0.029	0.013	FALSE
Sulfamethoxazole	51	Kruskal-Wallis H-test	1.42	> 0.1	0.050	FALSE
Sulfamethoxazole	366	Kruskal-Wallis H-test	9.50	0.009	0.008	FALSE
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Sulfamethoxazole	372	Kruskal-Wallis H-test	9.62	0.008	0.006	FALSE
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Sulfamethoxazole	391	Kruskal-Wallis H-test	5.96	0.029	0.017	FALSE
Sulfanilamide	3	Kruskal-Wallis H-test	5.60	0.050	0.013	FALSE
Sulfanilamide	8	Kruskal-Wallis H-test	4.36	> 0.1	0.025	FALSE
Sulfanilamide	51	Kruskal-Wallis H-test	0.47	> 0.1	0.050	FALSE
Sulfanilamide	366	Kruskal-Wallis H-test	8.34	0.015	0.010	FALSE
Sulfanilamide	369	Kruskal-Wallis H-test	9.42	0.009	0.008	FALSE
Sulfanilamide	372	Kruskal-Wallis H-test	9.62	0.008	0.007	FALSE
Sulfanilamide	377	Kruskal-Wallis H-test	9.78	0.008	0.006	FALSE
Sulfanilamide	384	Kruskal-Wallis H-test	10.00	0.007	0.006	FALSE
Sulfanilamide	391	One-way ANOVA	0.33	0.733	0.017	FALSE
Triclosan	3	Kruskal-Wallis H-test	3.20	> 0.1	0.050	FALSE
Triclosan	8	One-way ANOVA	0.52	0.619	0.013	FALSE
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Triclosan	366	Kruskal-Wallis H-test	4.74	0.093	0.008	FALSE
Triclosan	369	Kruskal-Wallis H-test	8.18	0.017	0.006	FALSE
Triclosan	372	Kruskal-Wallis H-test	5.16	0.076	0.007	FALSE
Triclosan	377	One-way ANOVA	4.32	0.039	0.006	FALSE
Triclosan	384	Kruskal-Wallis H-test	1.85	0.396	0.010	FALSE
Triclosan	391	One-way ANOVA	0.33	0.733	0.017	FALSE

Table S9. Significance testing for contaminants soil samples found in Figures 2 and S6. HB = Holm-Bonferroni.

Dependent variable	Time	Test	Statistic	p-value	α with HB correction	Is $p < \alpha$?
Acetaminophen	0	None (All values equal)	None (All values equal)	1.000	0.010	FALSE
Acetaminophen	6	None (All values equal)	None (All values equal)	1.000	0.013	FALSE
Acetaminophen	35	None (All values equal)	None (All values equal)	1.000	0.017	FALSE
Acetaminophen	69	None (All values equal)	None (All values equal)	1.000	0.025	FALSE
Acetaminophen	98	None (All values equal)	None (All values equal)	1.000	0.050	FALSE
Caffeine	0	Kruskal-Wallis H-test	5.79	0.029	0.010	FALSE
Caffeine	6	Kruskal-Wallis H-test	5.79	0.029	0.013	FALSE
Caffeine	35	Kruskal-Wallis H-test	5.79	0.029	0.017	FALSE
Caffeine	69	Kruskal-Wallis H-test	3.23	> 0.1	0.025	FALSE
Caffeine	98	Kruskal-Wallis H-test	2.10	> 0.1	0.050	FALSE
Carbamazepine	0	Kruskal-Wallis H-test	5.61	0.050	0.025	FALSE
Carbamazepine	6	Kruskal-Wallis H-test	5.79	0.029	0.010	FALSE
Carbamazepine	35	Kruskal-Wallis H-test	5.61	0.050	0.050	FALSE
Carbamazepine	69	Kruskal-Wallis H-test	5.79	0.029	0.013	FALSE
Carbamazepine	98	Kruskal-Wallis H-test	5.79	0.029	0.017	FALSE
Gemfibrozil	0	Kruskal-Wallis H-test	5.79	0.029	0.010	FALSE
Gemfibrozil	6	Kruskal-Wallis H-test	6.16	0.029	0.013	FALSE
Gemfibrozil	35	Kruskal-Wallis H-test	2.88	> 0.1	0.017	FALSE
Gemfibrozil	69	None (All values equal)	None (All values equal)	1.000	0.025	FALSE
Gemfibrozil	98	None (All values equal)	None (All values equal)	1.000	0.050	FALSE
Ibuprofen	0	Kruskal-Wallis H-test	2.54	> 0.1	0.010	FALSE
Ibuprofen	6	None (All values equal)	None (All values equal)	1.000	0.013	FALSE
Ibuprofen	35	None (All values equal)	None (All values equal)	1.000	0.017	FALSE
Ibuprofen	69	None (All values equal)	None (All values equal)	1.000	0.025	FALSE
Ibuprofen	98	None (All values equal)	None (All values equal)	1.000	0.050	FALSE
Sulfamethoxazole	0	Kruskal-Wallis H-test	6.16	0.029	0.010	FALSE
Sulfamethoxazole	6	Kruskal-Wallis H-test	5.79	0.029	0.013	FALSE
Sulfamethoxazole	35	Kruskal-Wallis H-test	2.00	> 0.1	0.017	FALSE
Sulfamethoxazole	69	None (All values equal)	None (All values equal)	1.000	0.025	FALSE
Sulfamethoxazole	98	None (All values equal)	None (All values equal)	1.000	0.050	FALSE
Triclosan	0	Kruskal-Wallis H-test	5.61	0.050	0.017	FALSE
Triclosan	6	Kruskal-Wallis H-test	7.45	0.004	0.010	TRUE
Triclosan	35	Kruskal-Wallis H-test	5.61	0.050	0.025	FALSE
Triclosan	69	Kruskal-Wallis H-test	5.61	0.050	0.050	FALSE
Triclosan	98	Kruskal-Wallis H-test	6.16	0.029	0.013	FALSE

Table S10. Post hoc analysis for contaminants soil samples found in Figure 2.

Dependent variable	Time	Treatment 1	Treatment 2	Test	α	Is $p < \alpha$?
Triclosan	6	C	CP100	Mann-Whitney U test	0.017	FALSE
Triclosan	6	CTRL	C	Mann-Whitney U test	0.017	FALSE
Triclosan	6	CTRL	CP100	Mann-Whitney U test	0.017	FALSE

Table S11. Significance testing for enzyme activities found in Figures 5 and S9. HB = Holm-Bonferroni.

Dependent variable	Time	Test	Statistic	p-value	α with HB correction	Is $p < \alpha$?
chitinase	0	One-way ANOVA	0.24	0.912	0.050	FALSE
chitinase	6	One-way ANOVA	7.75	0.000	0.005	TRUE
chitinase	35	Kruskal-Wallis H-test	10.10	0.039	0.006	FALSE
chitinase	69	Kruskal-Wallis H-test	1.73	0.786	0.017	FALSE
chitinase	98	One-way ANOVA	0.68	0.615	0.013	FALSE
chitinase	363	Kruskal-Wallis H-test	5.23	0.265	0.010	FALSE
chitinase	368	Kruskal-Wallis H-test	5.97	0.201	0.008	FALSE
chitinase	397	Kruskal-Wallis H-test	6.96	0.138	0.007	FALSE
chitinase	425	One-way ANOVA	3.64	0.019	0.006	FALSE
chitinase	456	One-way ANOVA	0.40	0.808	0.025	FALSE
β -glucosidase	0	Kruskal-Wallis H-test	3.03	> 0.05	0.050	FALSE
β -glucosidase	6	One-way ANOVA	2.12	0.111	0.008	FALSE
β -glucosidase	35	One-way ANOVA	3.66	0.018	0.006	FALSE
β -glucosidase	69	Kruskal-Wallis H-test	2.90	0.575	0.025	FALSE
β -glucosidase	98	One-way ANOVA	1.74	0.173	0.010	FALSE
β -glucosidase	363	Kruskal-Wallis H-test	7.59	0.108	0.007	FALSE
β -glucosidase	368	Kruskal-Wallis H-test	7.75	0.101	0.006	FALSE
β -glucosidase	397	Kruskal-Wallis H-test	3.65	0.456	0.013	FALSE
β -glucosidase	425	One-way ANOVA	5.02	0.005	0.005	TRUE
β -glucosidase	456	One-way ANOVA	0.82	0.525	0.017	FALSE
phosphatase	0	One-way ANOVA	1.06	0.426	0.017	FALSE
phosphatase	69	One-way ANOVA	1.83	0.154	0.010	FALSE
phosphatase	98	Kruskal-Wallis H-test	2.85	0.584	0.025	FALSE
phosphatase	363	Kruskal-Wallis H-test	9.34	0.053	0.007	FALSE
phosphatase	368	One-way ANOVA	0.71	0.591	0.050	FALSE
phosphatase	397	Kruskal-Wallis H-test	7.20	0.126	0.008	FALSE
phosphatase	425	One-way ANOVA	4.89	0.005	0.006	TRUE
phosphatase	456	One-way ANOVA	1.78	0.163	0.013	FALSE
xylan 1,4- β -xylosidase	0	One-way ANOVA	0.50	0.738	0.017	FALSE
xylan 1,4- β -xylosidase	6	One-way ANOVA	4.20	0.011	0.005	FALSE
xylan 1,4- β -xylosidase	35	Kruskal-Wallis H-test	6.11	0.191	0.007	FALSE
xylan 1,4- β -xylosidase	69	Kruskal-Wallis H-test	1.86	0.761	0.025	FALSE
xylan 1,4- β -xylosidase	98	Kruskal-Wallis H-test	2.95	0.565	0.010	FALSE
xylan 1,4- β -xylosidase	363	Kruskal-Wallis H-test	9.76	0.045	0.006	FALSE
xylan 1,4- β -xylosidase	368	One-way ANOVA	0.54	0.710	0.013	FALSE
xylan 1,4- β -xylosidase	397	Kruskal-Wallis H-test	0.74	0.947	0.050	FALSE
xylan 1,4- β -xylosidase	425	One-way ANOVA	3.17	0.032	0.006	FALSE
xylan 1,4- β -xylosidase	456	One-way ANOVA	1.60	0.206	0.008	FALSE

Table S12. Post hoc analysis for enzyme activities found in Figures 5 and S9.

Dependent variable	Time	Treatment 1	Treatment 2	Test	α	Is $p < \alpha$?
chitinase	6	C	CP100	Tukey's HSD	0.05	FALSE
chitinase	6	C	CTRL	Tukey's HSD	0.05	FALSE
chitinase	6	C	P10	Tukey's HSD	0.05	FALSE
chitinase	6	C	P100	Tukey's HSD	0.05	FALSE
chitinase	6	CP100	CTRL	Tukey's HSD	0.05	FALSE
chitinase	6	CP100	P10	Tukey's HSD	0.05	TRUE
chitinase	6	CP100	P100	Tukey's HSD	0.05	FALSE
chitinase	6	CTRL	P10	Tukey's HSD	0.05	TRUE
chitinase	6	CTRL	P100	Tukey's HSD	0.05	FALSE
chitinase	6	P10	P100	Tukey's HSD	0.05	FALSE
β -glucosidase	425	C	CP100	Tukey's HSD	0.05	FALSE
β -glucosidase	425	C	CTRL	Tukey's HSD	0.05	TRUE
β -glucosidase	425	C	P10	Tukey's HSD	0.05	FALSE
β -glucosidase	425	C	P100	Tukey's HSD	0.05	FALSE
β -glucosidase	425	CP100	CTRL	Tukey's HSD	0.05	TRUE
β -glucosidase	425	CP100	P10	Tukey's HSD	0.05	FALSE
β -glucosidase	425	CP100	P100	Tukey's HSD	0.05	FALSE
β -glucosidase	425	CTRL	P10	Tukey's HSD	0.05	FALSE
β -glucosidase	425	CTRL	P100	Tukey's HSD	0.05	FALSE
β -glucosidase	425	P10	P100	Tukey's HSD	0.05	FALSE
phosphatase	425	C	CP100	Tukey's HSD	0.05	FALSE
phosphatase	425	C	CTRL	Tukey's HSD	0.05	TRUE
phosphatase	425	C	P10	Tukey's HSD	0.05	FALSE
phosphatase	425	C	P100	Tukey's HSD	0.05	FALSE
phosphatase	425	CP100	CTRL	Tukey's HSD	0.05	FALSE
phosphatase	425	CP100	P10	Tukey's HSD	0.05	FALSE
phosphatase	425	CP100	P100	Tukey's HSD	0.05	FALSE
phosphatase	425	CTRL	P10	Tukey's HSD	0.05	TRUE
phosphatase	425	CTRL	P100	Tukey's HSD	0.05	FALSE
phosphatase	425	P10	P100	Tukey's HSD	0.05	FALSE

Table S13. Significance testing for plant growth found in Figure 6. HB = Holm-Bonferroni, LUT = Lookup table.

Dependent variable	Time	Test	Statistic	p-value	α with HB correction	Is $p < \alpha$?
dry biomass	1	One-way ANOVA	0.56	0.698	0.025	FALSE
dry biomass	2	None (LUT unavailable)	None (LUT unavailable)	N/A	0.050	FALSE
flower stalks	1	One-way ANOVA	1.42	0.283	0.050	FALSE
flower stalks	2	One-way ANOVA	2.28	0.089	0.025	FALSE
leaves	1	One-way ANOVA	1.35	0.305	0.025	FALSE
leaves	2	One-way ANOVA	0.55	0.700	0.050	FALSE
strawberries	1	One-way ANOVA	0.83	0.529	0.025	FALSE
strawberries	2	One-way ANOVA	0.15	0.960	0.050	FALSE

Table S14. Significance testing for Shannon entropy found in Figure S7. HB = Holm-Bonferroni.

Dependent variable	Time	Test	Statistic	p-value	α with HB correction	Is $p < \alpha$?
Shannon entropy	35	One-way ANOVA	1.14	0.392	0.017	FALSE
Shannon entropy	98	One-way ANOVA	0.85	0.523	0.025	FALSE
Shannon entropy	363	One-way ANOVA	2.72	0.091	0.010	FALSE
Shannon entropy	397	One-way ANOVA	0.12	0.970	0.050	FALSE
Shannon entropy	456	One-way ANOVA	1.51	0.272	0.013	FALSE

Table S15. Significance testing for phylum-level relative abundance found in Figure 3. HB = Holm-Bonferroni.

Dependent variable	Time	Test	Statistic	p-value	α with HB correction	Is $p < \alpha$?
Acidobacteria	35	One-way ANOVA	0.88	0.507	0.050	FALSE
Acidobacteria	98	One-way ANOVA	1.00	0.451	0.025	FALSE
Acidobacteria	363	One-way ANOVA	4.39	0.026	0.010	FALSE
Acidobacteria	397	One-way ANOVA	1.44	0.292	0.017	FALSE
Acidobacteria	456	One-way ANOVA	1.50	0.275	0.013	FALSE
Actinobacteria	35	One-way ANOVA	2.92	0.077	0.013	FALSE
Actinobacteria	98	One-way ANOVA	0.42	0.789	0.025	FALSE
Actinobacteria	363	Kruskal-Wallis H-test	4.77	> 0.05	0.050	FALSE
Actinobacteria	397	One-way ANOVA	7.92	0.004	0.010	TRUE
Actinobacteria	456	One-way ANOVA	1.08	0.415	0.017	FALSE
Chloroflexi	35	One-way ANOVA	0.74	0.587	0.017	FALSE
Chloroflexi	98	One-way ANOVA	2.44	0.115	0.013	FALSE
Chloroflexi	363	Kruskal-Wallis H-test	3.93	> 0.05	0.025	FALSE
Chloroflexi	397	One-way ANOVA	4.91	0.019	0.010	FALSE
Chloroflexi	456	Kruskal-Wallis H-test	6.57	> 0.05	0.050	FALSE
Crenarchaeota	35	One-way ANOVA	0.47	0.754	0.050	FALSE
Crenarchaeota	98	One-way ANOVA	2.29	0.131	0.010	FALSE
Crenarchaeota	363	One-way ANOVA	2.26	0.134	0.013	FALSE
Crenarchaeota	397	One-way ANOVA	1.36	0.315	0.025	FALSE
Crenarchaeota	456	One-way ANOVA	1.74	0.217	0.017	FALSE
Firmicutes	35	One-way ANOVA	1.24	0.353	0.013	FALSE
Firmicutes	98	One-way ANOVA	0.76	0.577	0.050	FALSE
Firmicutes	363	One-way ANOVA	0.85	0.524	0.025	FALSE
Firmicutes	397	One-way ANOVA	4.37	0.027	0.010	FALSE
Firmicutes	456	One-way ANOVA	1.06	0.427	0.017	FALSE
Gemmatimonadetes	35	One-way ANOVA	1.15	0.390	0.017	FALSE
Gemmatimonadetes	98	One-way ANOVA	0.36	0.830	0.050	FALSE
Gemmatimonadetes	363	One-way ANOVA	4.60	0.023	0.010	FALSE
Gemmatimonadetes	397	One-way ANOVA	1.30	0.335	0.013	FALSE
Gemmatimonadetes	456	One-way ANOVA	0.53	0.720	0.025	FALSE

Nitrospirae	35	Kruskal-Wallis H-test	9.37	0.050	0.010	FALSE
Nitrospirae	98	One-way ANOVA	0.56	0.699	0.025	FALSE
Nitrospirae	363	One-way ANOVA	0.74	0.587	0.017	FALSE
Nitrospirae	397	One-way ANOVA	0.23	0.915	0.050	FALSE
Nitrospirae	456	One-way ANOVA	0.95	0.475	0.013	FALSE
Other	35	One-way ANOVA	0.94	0.478	0.013	FALSE
Other	98	One-way ANOVA	0.32	0.861	0.017	FALSE
Other	363	Kruskal-Wallis H-test	4.77	> 0.05	0.050	FALSE
Other	397	One-way ANOVA	3.31	0.057	0.010	FALSE
Other	456	One-way ANOVA	0.31	0.866	0.025	FALSE
Planctomycetes	35	One-way ANOVA	2.21	0.140	0.013	FALSE
Planctomycetes	98	One-way ANOVA	0.59	0.675	0.050	FALSE
Planctomycetes	363	One-way ANOVA	2.51	0.108	0.010	FALSE
Planctomycetes	397	One-way ANOVA	2.11	0.154	0.017	FALSE
Planctomycetes	456	One-way ANOVA	1.53	0.266	0.025	FALSE
Proteobacteria	35	One-way ANOVA	6.10	0.009	0.010	TRUE
Proteobacteria	98	One-way ANOVA	4.12	0.031	0.013	FALSE
Proteobacteria	363	One-way ANOVA	1.26	0.349	0.025	FALSE
Proteobacteria	397	One-way ANOVA	3.90	0.037	0.017	FALSE
Proteobacteria	456	One-way ANOVA	0.69	0.616	0.050	FALSE
Verrucomicrobia	35	Kruskal-Wallis H-test	1.73	> 0.05	0.025	FALSE
Verrucomicrobia	98	One-way ANOVA	0.17	0.950	0.017	FALSE
Verrucomicrobia	363	One-way ANOVA	1.64	0.239	0.010	FALSE
Verrucomicrobia	397	Kruskal-Wallis H-test	6.40	> 0.05	0.050	FALSE
Verrucomicrobia	456	One-way ANOVA	0.59	0.676	0.013	FALSE
WS3	35	One-way ANOVA	1.73	0.221	0.010	FALSE
WS3	98	One-way ANOVA	0.41	0.801	0.050	FALSE
WS3	363	One-way ANOVA	1.52	0.270	0.013	FALSE
WS3	397	One-way ANOVA	1.27	0.343	0.017	FALSE
WS3	456	One-way ANOVA	0.41	0.801	0.025	FALSE

Table S16. Post hoc analysis for phylum-level relative abundance found in Figure 3.

Dependent variable	Time	Treatment 1	Treatment 2	Test	α	Is $p < \alpha$?
Actinobacteria	397	C	CP100	Tukey's HSD	0.05	FALSE
Actinobacteria	397	C	CTRL	Tukey's HSD	0.05	TRUE
Actinobacteria	397	C	P10	Tukey's HSD	0.05	FALSE
Actinobacteria	397	C	P100	Tukey's HSD	0.05	FALSE
Actinobacteria	397	CP100	CTRL	Tukey's HSD	0.05	TRUE
Actinobacteria	397	CP100	P10	Tukey's HSD	0.05	FALSE
Actinobacteria	397	CP100	P100	Tukey's HSD	0.05	FALSE
Actinobacteria	397	CTRL	P10	Tukey's HSD	0.05	TRUE
Actinobacteria	397	CTRL	P100	Tukey's HSD	0.05	FALSE
Actinobacteria	397	P10	P100	Tukey's HSD	0.05	FALSE
Proteobacteria	35	C	CP100	Tukey's HSD	0.05	FALSE
Proteobacteria	35	C	CTRL	Tukey's HSD	0.05	TRUE
Proteobacteria	35	C	P10	Tukey's HSD	0.05	TRUE
Proteobacteria	35	C	P100	Tukey's HSD	0.05	FALSE
Proteobacteria	35	CP100	CTRL	Tukey's HSD	0.05	FALSE
Proteobacteria	35	CP100	P10	Tukey's HSD	0.05	FALSE
Proteobacteria	35	CP100	P100	Tukey's HSD	0.05	FALSE
Proteobacteria	35	CTRL	P10	Tukey's HSD	0.05	FALSE
Proteobacteria	35	CTRL	P100	Tukey's HSD	0.05	FALSE
Proteobacteria	35	P10	P100	Tukey's HSD	0.05	FALSE