# Supporting Information

## Plastic products leach chemicals that induce *in vitro* toxicity under realistic use conditions

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## Supplementary methods

### S1. Determination of migration conditions

In order to determine migration conditions, 60 g of PVC 4 were placed in 2 L-glass bottle containing 1.5 L ultrapure water and incubated for two, five or ten days (d) in a climate chamber (Thermotec GmbH & Co. KG, Weilburg) at 40 °C in the dark. Three mL of the aqueous solution were stored in clean brown glass vials with PFTE caps at 8 °C until application to the bioassay. The remaining volume was transferred into new 2-L glass bottles for solid-phase extraction (SPE). Of procedural and SPE blanks, only some induced a very low baseline toxicity. Neither effects of aqueous migrates nor migrates (SPE-extracted) on unspecific and endocrine endpoints correlated with the leaching time (Table S1). Instead, effect concentrations (ECs) and levels (ELs) rather depended on the endpoint and whether the sample was extracted by SPE or not. Exemplary, after 10 d of migration the aqueous migrate induced a stronger baseline toxicity than after 5 d whereas it was vice versa for the SPE-extracted migrate. Thus, we chose the test setup recommended by the European Commission regulation on plastic FCMs of 10 d.[1]

### S2. Databases for *in silico* fragmentation

We generated three databases for the tentative identification of plastic chemicals using the Metascope algorithm in Progenesis QI:

1) the mapped version of the database of Chemicals associated with Plastic Packaging (CPPdb) containing chemicals likely[2] and possibly[3] associated with plastic packaging,

2) the chemicals registered under the REACH regulation in 2020 (ECHAdb) falling in all product categories except PC 11 (Explosives), PC 12 (Fertilizers), PC 14 (Metal surface treatment products), PC 23 (Leather treatment products), PC 25 (Metal working fluids), PC 27 (Plant protection products), PC 29 (Pharmaceuticals), PC 33 (Semiconductors), PC 36 (Water softeners), PC 37 (Water treatment chemicals), PC 38 (Welding and soldering products, flux products), PC 42

(Electrolytes for batteries). We downloaded the lists from https://echa.europa.eu/information-on-chemicals/registered-substances on 13.12.2020, combined all available CAS numbers and removed duplicates,

3) the chemicals (pre)registered under the REACH regulation in 2017 (NORMANdb) as provided by the NORMAN Suspect List Exchange.[4] We combined all available SMILES codes and removed duplicates.

To generate molecular structures for *in silico* fragmentation, we annotated the available CAS numbers of the chemicals in the ECHAdb with SMILES codes using the CompTox Chemicals Dashboard (https://comptox.epa.gov/dashboard/dsstoxdb/batch_search). For the CPPdb and the NORMANdb, we used the SMILES codes available in the respective list. We converted the SMILES to CID numbers using PubChem's Identifier Exchange Service (https://pubchem.ncbi.nlm.nih.gov/idexchange) and downloaded the structural information as individual sdf file per database using PubChem Entrez (https://pubchem.ncbi.nlm.nih.gov). Each annotation step resulted in the loss of compounds that either had no SMILES codes, CID or structural information (Table S2). The resulting structural databases contained 2680 (CPPdb), 7092 (ECHAdb), and 65,738 chemicals (NORMANdb) for *in silico* fragmentation.

## Tables and Figures

**Table S1**. Determination of migration conditions using PVC 4.

| Sample | Test | Endpoint | Migrate (SPE-extracted) | | | Aqueous Migrate | |
|---|---|---|---|---|---|---|---|
| | | | Day 2 | Day 5 | Day 10 | Day 5 | Day 10 |
| PVC 4 | Microtox | $EC_{20}$ [mg plastic well$^{-1}$] | 4.17 | 2.05 | 4.17 | 0.71 | 0.43 |
| | | $EC_{50}$ [mg plastic well$^{-1}$] | 17.10 | 9.46 | 14.01 | — | 1.66 |
| | | LI [%] | 100 .0 | 100.0 | 100.0 | 45.70 | 59.55 |
| | AREc32 | $EC_{IR2}$ [mg plastic well$^{-1}$] | 10.33 | 10.90 | 11.93 | n.a. | n.a |
| | | IR [%] | 77.26 | 77.76 | 60.56 | n.a. | n.a |
| | | Cytotoxic concentration [mg plastic well$^{-1}$] | n.a. | n.a | 200.0 | 200.0 | 20 0 |
| | YES | Relative estrogenic activity [%] | — | — | — | n.a. | 96.35 |
| | | Cytotoxicity, $EC_{20}$ [mg plastic well$^{-1}$] | yes | - | 31.84 | 52.44 | 98.94 |
| | YAAS | Relative antiandrogenic activity [%] | 60.68 | 50.11 | 50.10 | — | — |
| | | $EC_{50}$ [mg plastic well$^{-1}$] | 39.51 | 46.15 | 50.34 | n.a. | n.a. |
| | | Cytotoxicity, $EC_{20}$ [mg plastic well$^{-1}$] | — | — | 70.68 | 88.11 | 80.45 |
| PB | Microtox | $EC_{20}$ [mg plastic well$^{-1}$] | 503.8 | 145.9 | 289.8 | — | — |
| | | $EC_{50}$ [mg plastic well$^{-1}$] | — | — | 501.8 | | |
| | | LI [%] | 21.82 | 42.77 | 53.35 | — | — |
| | AREc32 | $EC_{IR2}$ [mg plastic well$^{-1}$] | — | — | — | n.a. | n.a |
| | | IR [%] | 1.55 | 1.57 | 1.62 | n.a. | n.a |
| | | Cytotoxic concentration [mg plastic well$^{-1}$] | n.a. | n.a | — | — | — |
| | YES | Relative estrogenic activity [%] | — | — | — | n.a. | n.a. |
| | | Cytotoxicity, $EC_{20}$ [mg plastic well$^{-1}$] | yes | yes | — | — | — |
| | YAAS | Relative antiandrogenic activity [%] | — | — | — | — | — |
| | | Cytotoxicity, $EC_{20}$ [mg plastic well$^{-1}$] | — | — | — | — | — |
| SPE blank | Microtox | $EC_{20}$ [mg plastic well$^{-1}$] | — | 423.3 | — | — | — |
| | | $EC_{50}$ [mg plastic well$^{-1}$] | — | — | — | — | — |
| | | LI [%] | 16.63 | 31.32 | 18.83 | 0.50 | — |
| | AREc32 | $EC_{IR2}$ [mg plastic well$^{-1}$] | 1.32 | 1.46 | 1.37 | n.a. | n.a |
| | | IR [%] | — | — | — | n.a. | n.a |
| | | Cytotoxic concentration [mg plastic well$^{-1}$] | n.a. | n.a | — | — | — |
| | YES | Relative estrogenic activity [%] | — | — | — | n.a. | n.a. |
| | | Cytotoxicity, $EC_{20}$ [mg plastic well$^{-1}$] | yes | yes | — | — | — |
| | YAAS | Relative antiandrogenic activity [%] | — | — | — | — | — |
| | | Cytotoxicity, $EC_{20}$ [mg plastic well$^{-1}$] | — | — | — | — | — |

Note: PVC, polyvinyl chloride; PB, procedural blank, SPE, solid-phase-extraction; IR, induction ratio; $EC_{20/50}$, effective concentration inducing 20/50% effect; LI, luminescence inhibition, $EC_{IR2}$, effective concentration leading to a luciferase induction ratio of 2.0 over the control (IR 2); —, no effect at measured concentrations; n.a., not assessed (e.g., due to cytotoxicity).

Number of experiments (*n*) performed and concentrations analyzed in 1:2 serial dilutions/assay:

Microtox: aqueous migrate $n = 1$ in duplicates, 0.018–5.00 mg plastic well$^{-1}$, migrate: $n = 2$–4 in duplicates, 0.018–600 mg plastic well$^{-1}$.

AREc32 assay: migrate $n = 3$–4 in duplicates, 1.56–200 mg plastic well$^{-1}$, IRs are given for the highest measured noncytotoxic concentration.

YES: aqueous migrate: $n = 8$, 3 mg plastic well$^{-1}$, migrate: $n = 8$–16, 0.195–100 mg plastic well$^{-1}$, relative activities are given for the highest measured noncytotoxic concentration.

YAAS: aqueous migrate: $n$ = 8, 3.00 mg plastic well$^{-1}$, migrate: $n$ = 16–24, 0.195–100 mg plastic well$^{-1}$, limit of detection = 57.6, relative activities are given for the highest measured noncytotoxic concentration.

**Table S2.** Number of compounds covered by the three databases used for the tentative identification of plastic chemicals using *in silico* fragmentation.

| Database | CAS numbers | SMILES | CIDs | Structures for *in silico* fragmentation |
|---|---|---|---|---|
| CPPdb | 4256[a] | 2777 | 2702 | 2680 |
| ECHAdb | 10,443 | 7305 | 7099 | 7092 |
| NORMANdb | 68,679[a] | 68,679 | 67,628 | 65,738 |

[a] we used the SMILES codes directly

**Table S3.** Reference compounds used in the *in vitro* bioassays: 3,5-dichlorophenol for luminescence inhibition of *A. fischeri*, 17$\beta$-estradiol for agonistic activity at the hER$\alpha$, and flutamide for antagonistic activity at the hAR as well as *tert*-butylhydroquinone (t-BHQ) for induction of an oxidative stress response. For 3,5-DCP, mean $EC_{50}$ values and $r^2$ are derived from 13 (migrate) and 7 (aqueous migrate), for 17$\beta$-estradiol and flutamide from 19 and 22 independent experiments, and for t-BHQ from one test.

| Reference compound | Grade, supplier, CAS number | Concentration range (mol L$^{-1}$) | $EC_{50}$ (mol L$^{-1}$) | $r^2$ |
|---|---|---|---|---|
| 3,5-DCP (migrate) | 97 %, Sigma-Aldrich, 591-35-5 | $1.7 \times 10^{-6} – 2.9 \times 10^{-4}$ | $3.65 \times 10^{-5}$ | 0.993 |
| 3,5-DCP (aqueous migrate) | 97 %, Sigma-Aldrich, 591-35-5 | $4.6 \times 10^{-6} – 5.9 \times 10^{-4}$ | $8.01 \times 10^{-5}$ | 0.995 |
| 17β-estradiol | > 99 %, Merck, 50-28-2 | $1.0 \times 10^{-12} – 1.0 \times 10^{-8}$ | $7.63 \times 10^{-11}$ | 0.989 |
| Flutamide | > 98 %, Merck, 13311-84-7 | $7.8 \times 10^{-7} – 5.0 \times 10^{-5}$ | $1.24 \times 10^{-5}$ | 0.969 |
| t-BHQ | 97 %, Sigma-Aldrich, 1948-33-0 | $2.0 \times 10^{-6} – 5.6 \times 10^{-5}$ | — | 0.928 |

**Table S4.** Baseline toxicity (mean ± SEM) of procedural (PB) and SPE blanks (SPE) as well as of plastic migrates in the Microtox assay.

| Sample | $EC_{20}$ (mg plastic well$^{-1}$) | $EC_{50}$ (mg plastic well$^{-1}$) | Luminescence inhibition (%)[a] | n[b] |
|---|---|---|---|---|
| PB1 | — | — | 16.1 ± 2.97 | 6 |
| PB2 | — | — | 19.5 ± 1.45 | 6 |
| PB3 | — | — | 17.8 ± 1.54 | 3 |
| PB4 | — | — | 18.5 ± 1.72 | 3 |
| PB 5 | — | — | 4.30 ± 1.61 | 3 |
| PB 6 | — | — | 3.70 ± 1.78 | 3 |
| SPE 1 | — | — | 20.1 ± 1.61 | 6 |
| SPE 2 | — | — | 11.9 ± 1.59 | 3 |
| SPE 3 | — | — | 3.23 ± 2.25 | 3 |
| HDPE 1 | 53.2 ± 13.9 | 271 ± 11.4 | 66.4 ± 1.52 | 3 |
| LDPE 1 | 2.55 ± 0.49 | 8.44 ± 1.37 | 99.5 ± 0.34 | 4 |
| LDPE 2 | 2.83 ± 1.44 | 12.8 ± 2.37 | 100 ± 0.00 | 3 |
| LDPE 3 | 22.6 ± 5.06 | 127 ± 14.78 | 79.5 ± 1.48 | 3 |
| LDPE 4 | 3.38 ± 0.39 | 11.0 ± 1.23 | 100 ± 0.00 | 3 |
| PS 1 | 17.7 ± 1.86 | 56.4 ± 3.72 | 99.4 ± 0.34 | 3 |
| PS 2 | 0.48 ± 0.28 | 3.37 ± 0.79 | 100 ± 0.00 | 3 |
| PS 3 | 322 ± 77.1 | — | 31.7 ± 1.48 | 3 |
| PS 4 | 120 ± 17.1 | — | 47.2 ± 0.14 | 3 |
| PP 1 | 22.2 ± 3.82 | 53.1 ± 3.78 | 100 ± 0.00 | 3 |
| PP 2 | 4.24 ± 0.22 | 31.7 ± 2.29 | 100 ± 0.00 | 3 |
| PET 1 | 10.6 ± 1.55 | 37.8 ± 7.23 | 91.7 ± 1.08 | 2 |
| PVC 1 | 0.56 ± 0.14 | 1.98 ± 0.53 | 100 ± 0.00 | 6 |
| PVC 2 | 0.14 ± 0.06 | 1.59 ± 0.63 | 100 ± 0.00 | 6 |
| PVC 3 | 0.93 ± 0.72 | 5.06 ± 2.46 | 99.3 ± 0.49 | 5 |
| PVC 4 | 0.12 ± 0.03 | 2.72 ± 0.99 | 99.5 ± 0.30 | 6 |
| PUR 1 | 23.2 ± 0.62 | 93.8 ± 3.17 | 89.4 ± 0.74 | 3 |
| PUR 2 | 2.04 ± 0.64 | 11.3 ± 1.67 | 98.4 ± 0.69 | 4 |
| PUR 3 | 32.7 ± 5.29 | 122 ± 13.4 | 86.9 ± 1.69 | 4 |
| PUR 4 | 2.63 ± 1.10 | 15.5 ± 3.48 | 100 ± 0.04 | 4 |
| PLA 1 | 109 ± 7.08 | 247 ± 10.1 | 81.1 ± 0.26 | 3 |
| PLA 2 | 47.4 ± 4.37 | 270 ± 17.8 | 63.7 ± 1.42 | 3 |
| PLA 3 | 3.98 ± 0.62 | 16.7 ± 1.20 | 100 ± 0.00 | 3 |
| PLA 4 | 143 ± 7.66 | — | 46.7 ± 1.96 | 3 |

Note: —, no luminescence inhibition observed (< 20% for $EC_{20}$ or ≤ negative controls for luminescence inhibition) at analyzed concentrations.

[a] Luminescence inhibition induced by migrates from 600 mg plastic well$^{-1}$.

[b] Number of independent experiments performed with two technical replicates, each.

**Table S5.** Oxidative stress response (mean ± SEM) as well as cytotoxicity induced by procedural (PB) and SPE blanks (SPE) as well as plastic migrates in the AREc32 assay.

| Sample | $EC_{IR2}$ (mg plastic well$^{-1}$) | Max. IR[a] | Noncytotoxic conc. (mg plastic well$^{-1}$)[b] | *n*[c] |
|---|---|---|---|---|
| PB1 | — | 0.91 ± 0.11 | 200 | 3 |
| PB2 | — | 0.87 ± 0.09 | 200 | 3 |
| PB3 | — | 1.00 ± 0.04 | 200 | 3 |
| PB4 | — | 1.25 ± 0.11 | 200 | 3 |
| PB 5 | — | 1.02 ± 0.21 | 200 | 3 |
| PB 6 | — | 0.83 ± 0.09 | 200 | 3 |
| SPE 1 | — | 0.84 ± 0.07 | 200 | 3 |
| SPE 2 | — | 1.24 ± 0.01 | 200 | 3 |
| SPE 3 | — | 1.05 ± 0.15 | 200 | 3 |
| HDPE 1 | — | 1.03 ± 0.07 | 6.25 | 3 |
| LDPE 1 | 12.0 ± 1.06 | 58.9 ± 16.0 | 50 | 3 |
| LDPE 2 | 60.6 ± 1.46 | 2.45 ± 0.17 | 100 | 3 |
| LDPE 3 | 64.8 ± 8.38 | 9.81 ± 1.01 | 200 | 3 |
| LDPE 4 | 2.15 ± 0.25 | 46.70± 15.9 | 6.26 | 3 |
| PS 1 | 9.64 ± 0.38 | 80.3 ± 9.58 | 50 | 3 |
| PS 2 | 87.3 ± 141 | 1.91 ± 0.17[d] | 6.25 | 3 |
| PS 3 | 97.2 ± 57.3 | 3.49 ± 0.37[e] | 200 | 3 |
| PS 4 | 107 ± 14.6 | 3.30 ± 0.29 | 200 | 3 |
| PP 1 | 3.20 ± 0.18 | 43.9 ± 3.55 | 12.5 | 3 |
| PP 2 | 7.55 ± 0.34 | 6.18 ± 0.46 | 50 | 3 |
| PET 1 | 131 ± 29.1 | 5.67 ± 1.68 | 200 | 3 |
| PVC 1 | 6.62 ± 2.65 | 24.4 ± 17.6 | 12.5 | 3 |
| PVC 2 | 91.6 ± 79.2 | 10.1 ± 8.39 | 25 | 3 |
| PVC 3 | 74.0 ± 10.0 | 5.84 ± 1.03 | 200 | 3 |
| PVC 4 | 18.8 ± 12.0 | 29.4 ± 13.8 | 50 | 3 |
| PUR 1 | 25.0 ± 0.48 | 10.2 ± 0.97 | 100 | 3 |
| PUR 2 | 12.9 ± 3.41 | 4.57 ± 0.56 | 50 | 3 |
| PUR 3 | 15.2 ± 2.96 | 23.8 ± 1.22 | 100 | 3 |
| PUR 4 | 19.5 ± 3.70 | 9.87 ±0.41 | 100 | 3 |
| PLA 1 | 103.5 ± 11.5 | 4.19 ± 0.49 | 200 | 3 |
| PLA 2 | 110.7 ± 9.08 | 4.56 ± 1.03 | 200 | 3 |
| PLA 3 | 26.9 ± 2.41 | 2.89 ± 0.90 | 50 | 3 |
| PLA 4 | — | 1.36 ± 0.21 | 100 | 3 |

Note: —, no oxidative stress response observed (induction rate < 2) at the analyzed concentrations.

[a] Maximal luciferase induction ratio induced by the highest measured non-cytotoxic concentration.

[b] Highest analyzed concentration at which no cytotoxicity was observed. Concentrations between 1.56 and 200 mg plastic well$^{-1}$ were tested.

[c] Number of independent experiments performed in two technical replicates, each.

[d] IR of two replicates exceeded 2 and thus, the $EC_{IR2}$ was still calculated using all 3*n*.

[e] Only of $n = 2$ since 1*n* was cytotoxic at 200 mg plastic well$^{-1}$ and thus, excluded.

**Table S6.** Estrogenic and antiandrogenic activities (mean ± SEM) of procedural blanks (PB), SPE blanks (SPE) and samples in YES and YAAS as well cytotoxicity.

| | YES | | | | YAAS | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Sample | Cytotox.[a] | rEA (%)[b] | *p* value[c] | *n* | Cytotox.[a] | rAA (%)[b] | $EC_{50}$ (mg plastic)[d] | *p* value[c] | *n* |
| PB1 | — | 0.08 ± 0.05 | n.a. | 24 | — | — | — | n.a. | 32 |
| PB2 | — | 0.15 ± 0.13 | n.a. | 24 | — | 10.4 ± 1.99 | — | n.a. | 32 |
| PB3 | — | 0.44 ± 0.07 | n.a. | 24 | — | 4.76 ± 1.41 | — | n.a. | 32 |
| PB4 | — | 0.18 ± 0.13 | n.a. | 24 | — | 14.5 ± 2.18 | — | n.a. | 24 |
| PB 5 | — | 0.14 ± 0.05 | n.a. | 24 | — | 1.97 ± 1.29 | — | n.a. | 32 |
| PB 6 | — | 0.10 ± 0.04 | n.a. | 24 | — | 7.58 ± 0.98 | — | n.a. | 32 |
| SPE 1 | — | — | n.a. | 24 | — | — | — | n.a. | 32 |
| SPE 2 | — | 0.24 ± 0.21 | n.a | 24 | — | — | — | n.a. | 32 |
| SPE 3 | — | 0.14 ± 0.18 | n.a. | 24 | — | — | — | n.a. | 32 |
| HDPE 1 | — | 0.51 ± 0.97 | n.a. | 16-24 | | 92.1 ± 0.50 | 23.4 | <0.001 | 32 |
| LDPE 1 | 49.0 | — (25.0) | n.a. | 16-24 | 83.9 | 32.6 ± 1.62 (50.0) | — | <0.001 | 24 |
| LDPE 2 | 57.0 | 1.04 ± 0.08 (50.0) | n.a. | 16-24 | 51.8 | — (25.0) | — | n.a. | 24 |
| LDPE 3 | — | 0.75 ± 0.08 | n.a. | 16-24 | — | 71.6 ± 1.49 | 63.5 | <0.001 | 24 |
| LDPE 4 | 100 | 0.50 ± 0.01 (50.0) | n.a. | 16-24 | — | 91.6 ± 0.56 | 36.7 | <0.001 | 24 |
| PS 1 | — | — | n.a. | 16-24 | — | 61.6 ± 2.00 | 72.3 | <0.001 | 32 |
| PS 2 | 3.33 | 0.82 ± 0.20 (3.13) | n.a. | 16-24 | 0.35 | — [f] | — | n.a. | 32 |
| PS 3 | — | 0.14 ± 0.06 | n.a. | 16-24 | — | 0.72 ± 1.21 | — | n.a. | 32 |
| PS 4 | — | 0.84 ± 0.11 | n.a. | 16-24 | — | 7.07 ± 1.73 | — | n.a. | 32 |
| PP 1 | 75.7 | — (50.0) | n.a. | 16-24 | 19.9 | 40.6 ± 1.03 (25.0) | — | <0.001 | 32 |
| PP 2 | 43.0 | — (25.0) | n.a. | 16-24 | 0.03 | — (0.01) | — | n.a. | 24 |
| PET 1 | — | — | n.a. | 16-24 | — | 1.13 ± 1.22 | — | n.a. | 24 |
| PVC 1 | 21.3 | 0.46 ± 0.12 (12.5) | n.a. | 16-24 | 60.0 | 54.0 ± 2.53 (50.0) | 47.0 | <0.001 | 32 |
| PVC 2 | 2.02 | 59.4 ± 2.38 (1.56) [e] | <0.001 | 16-24 | 1.66 | 90.9 ± 1.25 (0.78) | 0.28 | <0.001 | 32 |
| PVC 3 | 75.6 | 0.21 ± 0.16 (50) | n.a. | 16-24 | 3.66 | — (0.31) | — | n.a. | 32 |
| PVC 4 | 41.0 | 0.98 ± (25) | n.a. | 16-24 | 40.1 | 19.3 ± 3.00 (25.0) | — | n.a | 32 |
| PUR 1 | — | 0.01 ± 0.06 | n.a. | 16-24 | — | 45.9 ± 1.68 | — | <0.001. | 32 |
| PUR 2 | — | 0.36 ± 0.03 | n.a. | 16-24 | — | 65.8 ± 3.20 | 70.3 | <0.001 | 32 |
| PUR 3 | — | 0.90 ± 0.33 | n.a. | 16-24 | — | 79.9 ± 2.27 | 60.1 | <0.001 | 32 |
| PUR 4 | — | 0.43 ± 0.04 | n.a. | 16-24 | — | 92.2 ± 0.68 | 20.2 | <0.001 | 32 |
| PLA 1 | 22.1 | — (12.5) | n.a. | 16-24 | — | 42.5 ± 1.45 | — | <0.001. | 32 |
| PLA 2 | 67.4 | — (25) | n.a. | 16-24 | — | 7.85 ± 1.95 | — | n.a. | 32 |
| PLA 3 | 0.10 | — (0.08) | n.a. | 16-24 | 0.02 | 4.51 ± (0.01) | — | n.a. | 32 |
| PLA 4 | — | — | n.a. | 16-24 | — | 12.3 ± 1.46 | — | n.a. | 32 |

Note: rEA, relative estrogenic activity; rAA, relative antiandrogenic activity; —, No effect observed for the respective endpoint; n a., not analyzed.

[a] Cytotoxicity (cytotox.) as $EC_{20}$ (mg plastic well$^{-1}$) of migrates from ≤ 100 mg plastic well$^{-1}$.

[b] rEA or rAA for the highest measured non-cytotoxic concentration (in brackets if not 100 mg well$^{-1}$).

[c] Statistical differences of relative activities were only analyzed if > limit of detection (YES: 1.65%, YAAS: 27.3%). *p* value compared to the control using Kruskal-Wallis with Dunn's post hoc test.

[d] For the antiandrogen activity $EC_{50}$ values were derived if within the measured concentrations.

[e] For the estrogenic activity of PVC 2 a $EC_{50}$ of 0.27 mg plastic well$^{-1}$ was derived.

[f] All measured concentrations (0.20–100 mg plastic well$^{-1}$) were cytotoxic, such that antiandrogenicity could not be assessed

**Table S7.** Comparison of the toxicity of migrates and extracts.

| Sample | $EC_{20}$ baseline toxicity (mg plastic well$^{-1}$) | | $EC_{IR2}$ oxidative stress (mg plastic well$^{-1}$) | | % relative estrogenic activity | | % relative antiandrogenic activity | |
|---|---|---|---|---|---|---|---|---|
| | Extract | Migrate | Extract | Migrate | Extract | Migrate | Extract | Migrate |
| HDPE 1 | 14.6 | — | — | — | 2.75 | (0.34) | 31.53 | (13.7) |
| LDPE 1 | 4.34 | 2.55 | — | — | (0.28) | — | (2.16) | (0.14) |
| LDPE 2 | 1.02 | 2.83 | — | — | (2.21) | (0.31) | (9.30) | — |
| LDPE 3 | — | — | — | — | (1.63) | (0.01) | (11.1) | (0.35) |
| LDPE 4 | 2.63 | 3.38 | 0.48 | 2.15 | (0.71) | — | (12.3) | (4.84) |
| PS 1 | — | 17.7 | — | — | 3.82 | — | — | — |
| PS 2 | 1.30 | 0.48 | 2.79 | — | (1.13) | — | — | — |
| PS 3 | 22.3 | — | x | — | (0.37) | (0.16) | — | — |
| PS 4 | 18.3 | — | x | — | (1.17) | — | (1.16) | — |
| PP 1 | — | 22.2 | 3.83 | 3.20 | — | — | 47.84 | (0.31) |
| PP 2 | 3.63 | 4.24 | 0.99 | — | — | — | — | — |
| PET 1 | — | 10.6 | 6.69 | — | (1.22) | (0.05) | (16.3) | — |
| PVC 1 | 0.23 | 0.56 | 6.64 | 6.62 | — | — | (25.5) | (6.05) |
| PVC 2 | 1.22 | 0.14 | 2.14 | — | 27.1 | 60.28 | 40.1 | 90.9 |
| PVC 3 | 1.80 | 0.93 | 2.42 | — | — | — | 34.6 | — |
| PVC 4 | 0.49 | 0.12 | 1.16 | — | 6.91 | — | 48.4 | (2.47) |
| PUR 1 | 3.02 | — | 1.13 | — | (0.59) | — | 55.8 | (0.08) |
| PUR 2 | 2.73 | 2.04 | 0.51 | — | — | (0.02) | 69.0 | (4.37) |
| PUR 3 | 3.56 | — | 0.47 | — | (0.36) | (0.14) | 82.3 | (1.65) |
| PUR 4 | 10.2 | 2.63 | 1.82 | — | (2.26) | (0.07) | 30.4 | (10.7) |
| PLA 1 | 2.12 | — | — | — | (0.02) | — | — | (1.36) |
| PLA 2 | 6.21 | — | — | — | (0.34) | — | (13.2) | — |
| PLA 3 | 0.01 | 3.98 | 0.98 | — | — | — | — | — |
| PLA 4 | 1.32 | — | — | — | — | (0.14) | (15.3) | (8.02) |

Note: –, no effect at concentrations analyzed or value not calculable.

Bioassay results of migrates and extracts at concentration ranges or points assessed for both leaching conditions: Baseline toxicity: 0—22.5 mg plastic well$^{-1}$, oxidative stress response induction: 0—7.5 mg plastic well$^{-1}$, estrogenic and antiandrogenic activities: 3.75 mg plastic well$^{-1}$ (exception: PVC 2: 0.94 (YES) and 0.78 mg plastic well$^{-1}$ (YAAS)). Relative activities < LOD are given in brackets and correspond to 1.65 % (migrates) and 2.33 % (extracts) for estrogenic as well as to 27.3 % (migrates) and 29.2 % (extracts) for antiandrogenic activities.

**Table S8.** Comparison of the toxicity of migrates (with SPE) with aqueous migrates (without SPE).

| Sample | $EC_{20}$ baseline toxicity (mg plastic well$^{-1}$) | | LI baseline toxicity (mg plastic well$^{-1}$) | | % relative antiandrogenic activity | |
|---|---|---|---|---|---|---|
| | Migrate | Aqueous migrate | Migrate | Aqueous migrate | Migrate | Aqueous migrate |
| HDPE 1 | — | — | 2.75 | 3.85 | (11.4) | (7.18) |
| LDPE 1 | 2.55 | 2.88 | 36.1 | 41.1 | <LOD | >LOD |
| LDPE 2 | 2.83 | 3.84 | 32.5 | 29.6 | — | — |
| LDPE 3 | — | — | 7.45 | — | (0.23) | — |
| LDPE 4 | 3.38 | 3.24 | 28.8 | 39.1 | (3.74 | (16.0) |
| PS 1 | — | 4.83 | 5.46 | 21.0 | (0.01) | (0.15) |
| PS 2 | 0.48 | 1.10 | 56.3 | 61.7 | — | (21.8) |
| PS 3 | — | — | 2.76 | 4.63 | — | — |
| PS 4 | — | — | 1.84 | 1.48 | — | (1.85) |
| PP 1 | — | — | 2.85 | 9.85 | (0.13) | 29.4 |
| PP 2 | 4.24 | 3.51 | 21.9 | 30.2 | — | cytotoxic |
| PET 1 | — | 4.37 | 10.1 | 23.6 | — | — |
| PVC 1 | 0.56 | 0.46 | 68.5 | 86.1 | (5.02) | (3.38) |
| PVC 2 | 0.14 | 0.42 | 67.6 | 72.6 | cytotoxic | cytotoxic |
| PVC 3 | 0.93 | 0.80 | 59.9 | 47.4 | — | cytotoxic |
| PVC 4 | 0.12 | 0.77 | 60.1 | 50.9 | (1.55) | (17.9) |
| PUR 1 | — | — | 5.18 | — | (0.05) | (0.39) |
| PUR 2 | 2.04 | 3.37 | 35.6 | 30.5 | (3.61) | (3.33) |
| PUR 3 | — | — | 3.84 | 16.1 | (1.23) | (1.48) |
| PUR 4 | 2.63 | 1.43 | 30.5 | 44.6 | (8.31) | (16.9) |
| PLA 1 | — | — | 0.15 | 2.58 | (1.05) | — |
| PLA 2 | — | — | 4.06 | 7.23 | — | — |
| PLA 3 | 3.98 | 0.78 | 24.0 | 70.2 | — | — |
| PLA 4 | — | — | 1.35 | 18.5 | (7.87) | (0.38) |

Note: Note: –, no effect at concentrations analyzed or value not calculable.

Bioassay results of migrates and extracts at concentration ranges or points assessed for both leaching conditions: Baseline toxicity: $EC_{20}$ at 0—5.0 mg plastic well$^{-1}$ and luminescence inhibition (LI) at 3 mg plastic well$^{-1}$, antiandrogenic activities: 3.00 mg plastic well$^{-1}$ (exception: PVC 2: 0.78 mg plastic well$^{-1}$). Relative antiandrogenic activities < LOD are given in brackets and correspond to 27.3 % (migrates) and 29.2 % (extracts)..

**Table S9.** The ten most frequently identified compounds across all plastic samples. Note that the compounds have been identified multiple times in one sample, indicating the presence of isomers or false-positive annotations.

| PubChem CID | No. IDs | Common name | IUPAC name | Use according to PubChem | Formula | m/z | Detected in samples |
|---|---|---|---|---|---|---|---|
| 175956 | 77 | Mono(2-acryloyloxyethyl) succinate | 4-oxo-4-(2-prop-2-enoyloxyethoxy)butanoic acid | adhesive | C9H12O6 | 455.1163 | PLA 1, PLA 2, PLA 3, PLA 4 |
| 17083 | 40 | – | 11-aminoundecanoic acid | used in plastics[a] | C11H23NO2 | 184.1703 | HDPE 1, LDPE 1, LDPE 3, PET 1, PLA 3, PLA 4, PP 2, PS 2, PUR 1, PUR 2, PUR 3, PUR 4, PVC 2, PVC 4 |
| 62551 | 40 | Pentaethylene glycol | 2-[2-[2-[2-(2-hydroxyethoxy)ethoxy] ethoxy]ethoxy]ethanol | processing aid and additive[b] | C10H22O6 | 221.1381 | LDPE 3, LDPE 4, PLA 2, PLA 3, PP 1, PS 2, PUR 4, PVC 1, PVC 4 |
| 9988 | 39 | – | ethyl 2-fluoroacetate | no information | C4H7FO2 | 107.0500 | HDPE 1, LDPE 1, LDPE 2, LDPE 3, LDPE 4, PLA 2, PP 2, PS 2, PS 4, PUR 1, PUR 2, PUR 3, PUR 4, PVC 1, PVC 3, PVC 4 |
| 69661 | 37 | 12-Aminolauric acid | 12-aminododecanoic acid | used in plastics[a] | C12H25NO2 | 198.1857 | HDPE 1, LDPE 1, LDPE 3, PET 1, PLA 3, PLA 4, PP 3, PS 4, PUR 1, PUR 2, PUR 3, PUR 4, PVC 2 |
| 242516 | 37 | 6-Deoxy-D-mannono-4-lactone | 3,4-dihydroxy-5-(1-hydroxyethyl)oxolan-2-one | no information | C6H10O5 | 145.0499 | PLA 1, PLA 2, PLA 3, PLA 4, PS 2, PUR 1 |
| 20836325 | 34 | – | (2E,6Z)-dodeca-2,6-dien-1-ol | no information | C12H22O | 165.1642 | HDPE 1, LDPE 1, LDPE 2, LDPE 3, PET 1, PLA 1, PLA 2, PP 1, PS 1, PS 2, PS 4, PUR 1, PVC 1, PVC 2, PVC 3, PVC 4 |
| 7528 | 33 | Solketal | (2,2-dimethyl-1,3-dioxolan-4-yl) methanol | solvent[b] | C6H12O3 | 265.1641 | LDPE 3, LDPE 4, PLA 3, PP 1, PP 2, PVC 4 |
| 13690 | 32 | Laurolactam | Azacyclotridecan-2-one | used in plastics[a] | C12H23NO | 198.1858 | HDPE 1, LDPE 2, LDPE 3, PET 1, PP 2, PS 2, PUR 1, PUR 4, PVC 2, PVC 3 |
| 67796 | 31 | – | hexanoyl fluoride | no information | C6H11FO | 119.0868 | HDPE 1, LDPE 2, LDPE 3, PET 1, PLA 1, PLA 4, PP 2, PS 2, PUR 4, PVC 2, PVC 4 |

[a] according to EPA CPDat Chemical and Product Categories**,** [b] according to EPA Safer Choice

**Table S10.** Tentatively identified compounds out of the ten features with the highest abundance in the migrate of each plastic sample.

| Sample | PubChem CID | Formula | Score | Description | Neutral mass (Da) | m/z | Retention time (min) |
|---|---|---|---|---|---|---|---|
| HDPE 1 | none identified | | | | | | |
| LDPE 1 | 72822400 | C29H36O8 | 41.3 | 6-[2-(11,17-dihydroxy-10,13-dimethyl-3-oxo-7,8,9,11,12,14,15,16-octahydro-6H-cyclopenta[a]phenanthren-17-yl)-2-oxoethoxy]carbonylcyclohex-3-ene-1-carboxylic acid | 512.2415 | 535.2307 | 24.97 |
| LDPE 2 | 548230 | C14H28O6 | 41.1 | 2-(hydroxymethyl)-6-octoxyoxane-3,4,5-triol | | 315.1783 | 20.06 |
| LDPE 2 | 88669 | C16H20N4O2 | 40.1 | 2-[4-[(4-aminophenyl)diazenyl]-N-(2-hydroxyethyl)anilino]ethanol | | 323.1475 | 20.06 |
| LDPE 2 | 103841 | C16H32O6 | 40.8 | 2-[2-[2-(2-hydroxyethoxy)ethoxy]ethoxy]ethyl 2-ethylhexanoate | 320.2209 | 343.2101 | 23.54 |
| LDPE 2 | 92926 | C13H26O4 | 42.6 | 2,3-dihydroxypropyl decanoate | 246.1840 | 269.1732 | 23.73 |
| LDPE 2 | 73743781 | C26H40NO5- | 40.9 | 3-[2-[2-(2-hydroxyethyl)anilino]ethoxycarbonyl]pentadec-5-enoate | 446.2911 | 469.2803 | 25.49 |
| LDPE 2 | 99120788 | C18H36O6 | 42.3 | (2R,3S,4R,5R)-6-dodecoxy-2,3,4,5-tetrahydroxyhexanal | 348.2523 | 371.2416 | 25.56 |
| LDPE 3 | 3218 | C17H26O4 | 45.2 | 2,5-dihydroxy-3-undecylcyclohexa-2,5-diene-1,4-dione | 294.1836 | 277.1804 | 23.88 |
| LDPE 4 | 21149416 | C18H30O7 | 41.3 | 6-(5-carboxy-2,2,3-trimethylpentanoyl)oxy-4,5,5-trimethyl-6-oxohexanoic acid | 358.1991 | 381.1883 | 20.14 |
| LDPE 4[a] | 115157 | C21H32O9 | 42.3 | 2-[2,2-bis(2-prop-2-enoyloxyethoxymethyl)butoxy]ethyl prop-2-enoate | 428.2051 | 451.1943 | 21.97 |
| LDPE 4 | 139595991 | C21H32O8 | 44 | 1-[3-(2-carboxycyclohexanecarbonyl)oxy-2,2-dimethylpropoxy]carbonylcyclohexane-1-carboxylic acid | 412.2109 | 435.2001 | 22.42 |
| PS 1 | 446541 | C17H20O6 | 40.4 | (E)-6-(4-hydroxy-6-methoxy-7-methyl-3-oxo-1H-2-benzofuran-5-yl)-4-methylhex-4-enoic acid | | 353.1582 | 18.88 |
| PS 1[a] | 115157 | C21H32O9 | 42.9 | 2-[2,2-bis(2-prop-2-enoyloxyethoxymethyl)butoxy]ethyl prop-2-enoate | 428.2049 | 451.1941 | 21.98 |
| PS 2 | 74338 | C21H20O3 | 42.2 | [3,4-bis(phenylmethoxy)phenyl]methanol | 320.1407 | 343.1299 | 9.56 |
| PS 2 | 4867 | C18H38O10 | 46.4 | Nonaethylene glycol | 414.2470 | 415.2543 | 9.95 |
| PS 3 | none identified | | | | | | |
| PS 4 | 10180 | C20H24N2OS | 42.2 | 1-[2-(diethylamino)ethylamino]-4-methylthioxanthen-9-one | | 379.1237 | 24.08 |
| PP 1 | 12298194 | C22H28N2O2 | 50.1 | methyl (3S,4R)-4-anilino-3-methyl-1-(2-phenylethyl)piperidine-4-carboxylate | 352.2162 | 353.2235 | 13.84 |
| PP 1 | 16206038 | C15H28O7 | 44.7 | 1-[2,3-bis(2-hydroxypropoxy)propoxy]propan-2-yl prop-2-enoate | | 343.1734 | 15.52 |
| PP 1 | 117007 | C9H14N2O2 | 41 | 4-(2-methoxyethoxy)benzene-1,3-diamine | | 387.1995 | 16.13 |
| PP 1 | 119970 | C14H31NO2 | 41.6 | 1-[2-hydroxypropyl(octyl)amino]propan-2-ol | 245.2358 | 246.2430 | 18.85 |
| PP 1 | 352309 | C16H35NO2 | 50.2 | Lauryldiethanolamine | 273.2675 | 274.2747 | 22.18 |
| PP 1 | 139595991 | C21H32O8 | 42.3 | 1-[3-(2-carboxycyclohexanecarbonyl)oxy-2,2-dimethylpropoxy]carbonylcyclohexane-1-carboxylic acid | 412.2107 | 435.1999 | 22.44 |
| PP 1 | 48194 | C22H27NO3 | 41.7 | ethyl 4-morpholin-4-yl-2,2-diphenylbutanoate | 353.2007 | 354.2079 | 22.51 |
| PP 2 | 113595 | C24H42O13 | 52.8 | 1-[3-[3-[2,3-bis(oxiran-2-ylmethoxy)propoxy]-2-hydroxypropoxy]-2-(oxiran-2-ylmethoxy)propoxy]-3-(oxiran-2-ylmethoxy)propan-2-ol | 538.2632 | 561.2524 | 15.34 |

| Sample | PubChem CID | Formula | Score | Description | Neutral mass (Da) | m/z | Retention time (min) |
|---|---|---|---|---|---|---|---|
| PP 2 | 66197 | C10H16O5 | 43.2 | diethyl 2-acetylbutanedioate | 216.1002 | 455.1896 | 16.47 |
| PET 1 | 5541 | C9H14O6 | 41.8 | Triacetin | 218.0791 | 241.0684 | 10.76 |
| PET 1[a] | 53421640 | C21H33N3O3 | 41.6 | N-[3,5-bis(2,2-dimethylpropanoylamino)phenyl]-2,2-dimethylpropanamide | 375.2520 | 376.2593 | 20.13 |
| PVC 1 | 3024241 | C9H14O4 | 49.9 | 1-[[5-(hydroxymethyl)furan-2-yl]methoxy]propan-2-ol | | 373.1869 | 17.80 |
| PVC 1 | 53461848 | C27H36O6S | 44.4 | ethyl 7-acetylsulfanyl-10,13-dimethyl-2',3-dioxospiro[2,6,7,8,9,11,12,14,15,16-decahydro-1H-cyclopenta[a]phenanthrene-17,5'-oxolane]-3'-carboxylate | 471.2213 | | 17.80 |
| PVC 1 | 3024241 | C9H14O4 | 47.1 | 1-[[5-(hydroxymethyl)furan-2-yl]methoxy]propan-2-ol | | 373.1865 | 19.16 |
| PVC 1 | 19675 | C24H30N2O2S | 40 | 1-[10-[3-[4-(2-hydroxyethyl)piperidin-1-yl]propyl]phenothiazin-2-yl]ethanone | 410.2047 | 843.3986 | 22.53 |
| PVC 2[a] | 81779 | C24H51O10P | 42.5 | tris[2-(2-butoxyethoxy)ethyl] phosphate | 530.3234 | 531.3307 | 25.61 |
| PVC 3 | 12308635 | C29H44O8 | 41.1 | 3-[(3S,5R,8R,9S,10S,13R,14S,17R)-14-hydroxy-10,13-dimethyl-3-[(2R,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxy-1,2,3,4,5,6,7,8,9,11,12,15,16,17-tetradecahydrocyclopenta[a]phenanthren-17-yl]-2H-furan-5-one | 520.3043 | 543.2935 | 23.68 |
| PVC 3 | 81779 | C24H51O10P | 43.7 | tris[2-(2-butoxyethoxy)ethyl] phosphate | 530.3229 | 531.3302 | 25.61 |
| PVC 3 | 11046239 | C18H34O6 | 42.1 | [(2R)-2-[(2R,3R,4S)-3,4-dihydroxyoxolan-2-yl]-2-hydroxyethyl] dodecanoate | 346.2359 | 369.2251 | 25.91 |
| PVC 4 | 6540 | C18H39O7P | 44.1 | Tri(butoxyethyl)phosphate | 398.2438 | 399.2510 | 25.56 |
| PVC 4 | 6455034 | C13H22O | 41.8 | 2-methyl-4-(2,2,3-trimethylcyclopent-3-en-1-yl)butanal | | 227.2012 | 27.25 |
| PVC 4 | 75403 | C24H48O8 | 47.5 | 2-[2-[2-[2-[2-(2-hydroxyethoxy)ethoxy]ethoxy]ethoxy]ethoxy]ethyl dodecanoate | 464.3335 | 487.3227 | 27.27 |
| PUR 1 | none identified | | | | | | |
| PUR 2 | none identified | | | | | | |
| PUR 3 | none identified | | | | | | |
| PUR 4 | 22838325 | C26H47NO6 | 42.3 | (2-hydroxy-3-octadecanoyloxypropyl) (2S)-5-oxopyrrolidine-2-carboxylate | 469.3383 | 492.3275 | 1.14 |
| PLA 1 | 63216 | C20H21N3O3 | 40.3 | 1-(2-morpholin-4-ylacetyl)-3-phenyl-2H-quinazolin-4-one | | 374.1459 | 26.29 |
| PLA 2 | none identified | | | | | | |
| PLA 3 | 37907 | C15H17ClN2O2 | 48.3 | Climbazole | 292.0986 | 293.1059 | 17.18 |
| PLA 3 | 6454788 | C10H16O4 | 43.6 | 6-but-3-enoxy-6-oxohexanoic acid | 200.1048 | 423.1988 | 20.77 |
| PLA 3 | 6454788 | C10H16O4 | 46.4 | 6-but-3-enoxy-6-oxohexanoic acid | 200.1056 | 401.2186 | 20.77 |
| PLA 4 | 20836181 | C19H37NO | 47.4 | (Z)-N-methyloctadec-9-enamide | | 296.2939 | 30.10 |
| PLA 4 | 26396 | C10H18O2 | 43 | 6-ethenyl-2,2,6-trimethyloxan-3-ol | | 341.2676 | 30.16 |

Note: [a] listed in the CPPdb as associated with plastic packaging.

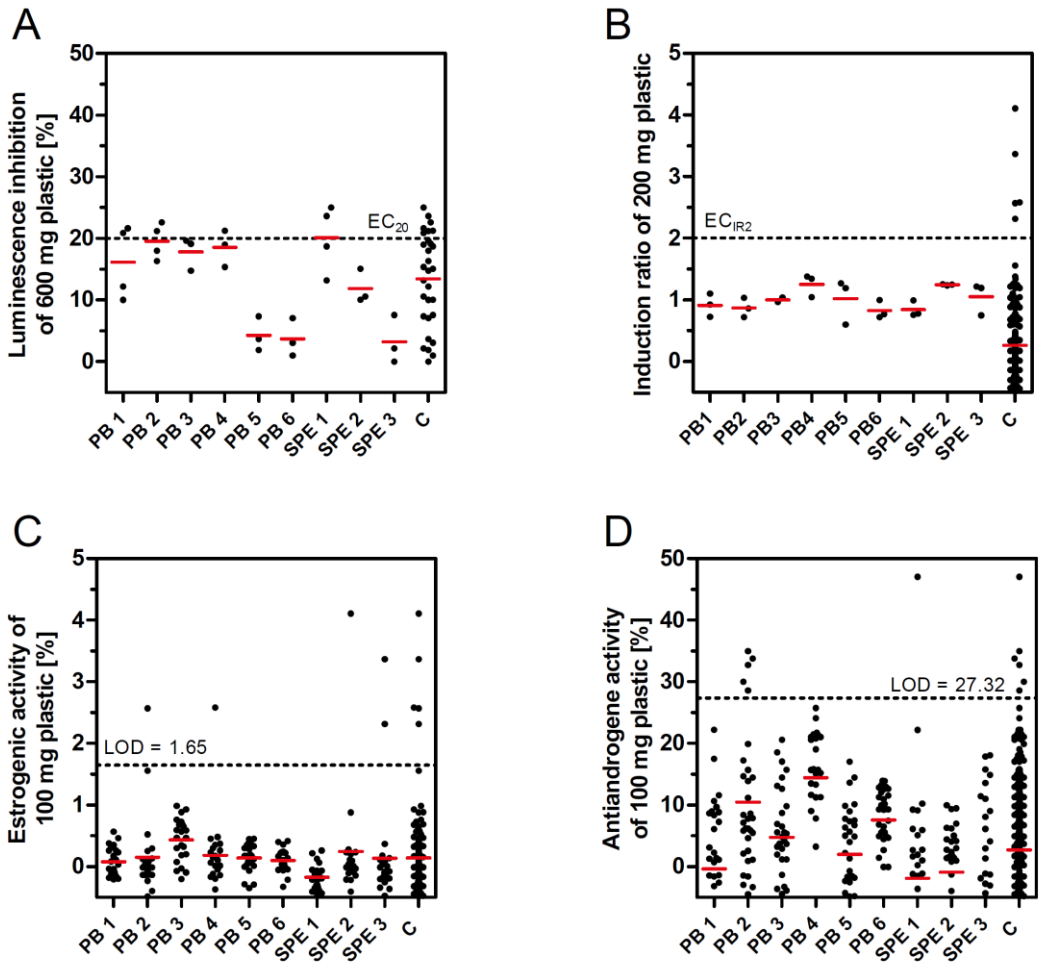



**Figure S1.** Effects of the procedural (PB) and SPE blanks (SPE) of the experiments with migrates (SPE extracted) as well as their pooled values (= controls; C) in the *in vitro* bioassays. PBs, SPEs, and C do not induce effects in the Microtox assay (*A*, $n$ = 3–4 in duplicates), the AREc32 assay (*B*, $n$ = 2–5 in duplicates), the yeast-based reporter-gene assays for estrogenic activity (*C*, $n$ = 24 from three independent experiment) and antiandrogenic activity (*D*, $n$ = 24–80 from ≥ 3 independent experiment). The results are presented as means (line) of $n$ (dots). The corresponding cut-off levels of each assay (effect concentrations (EC) and limits of detection (LOD)) were not exceeded.

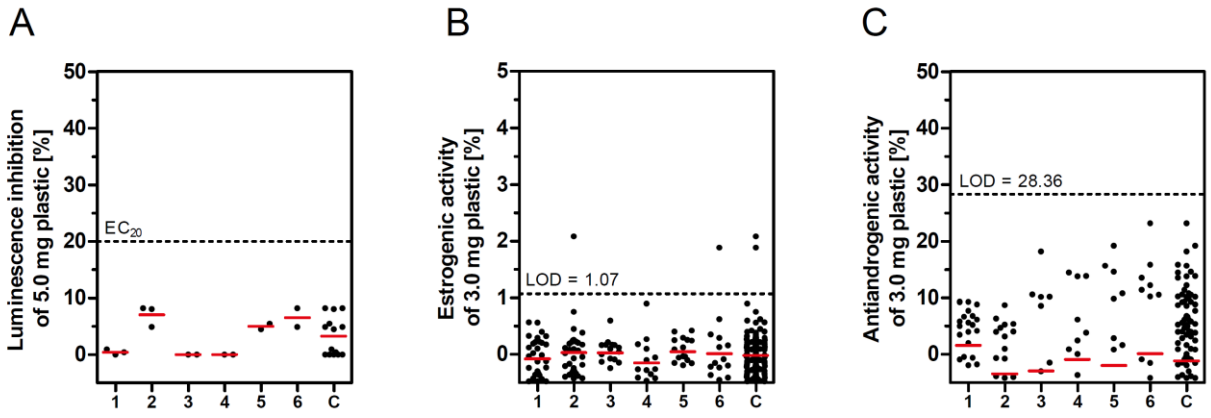



**Figure S2.** Effects of procedural blanks 1–6 of the experiments with aqueous migrates as well as their pooled values (= controls; C) in the *in vitro* bioassays. Procedural blanks and C do not induce effects in the Microtox assay (*A*, $n$ = 2–3 in duplicates), the yeast-based reporter-gene assays for estrogenic activity (*B*, $n$ = 16–32 from three independent experiment) and antiandrogenic activity (*C*, $n$ = 16–24 from ≥ 3 independent experiment). The results are presented as means (line) of $n$ (dots). The corresponding cut-off levels of each assay (effect concentrations (EC) and limits of detection (LOD)) were not exceeded.

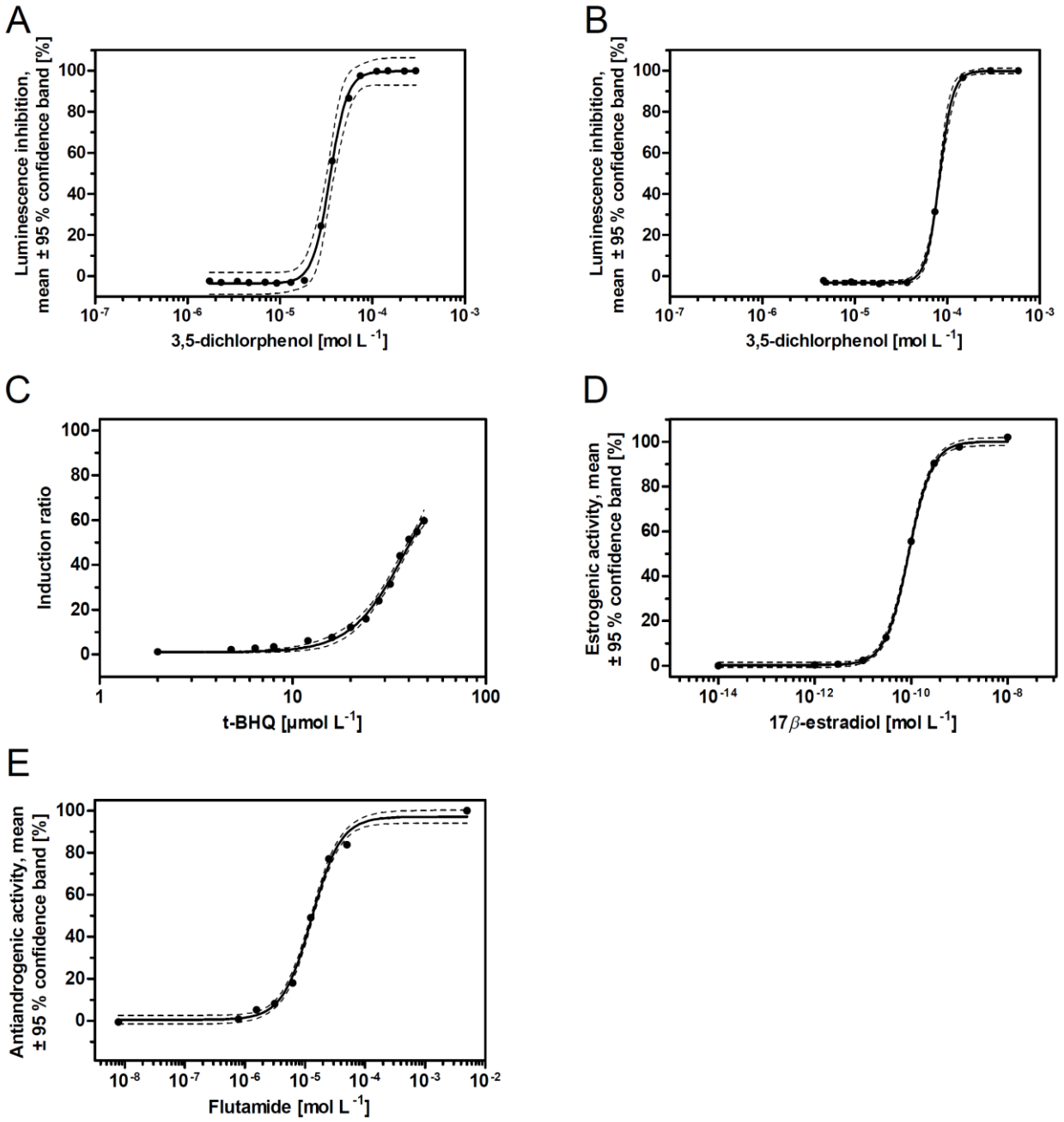



**Figure S3.** Dose-response relationship of the reference compounds in the bioassays. Microtox ($n$ = 3), measured in different concentrations for migrates (*A*) and aqueous migrates (*B*), AREc32 assay (*C*, $n$ = 8), YES (*D*, $n$ = 32), and YAAS (*E*, $n$ = 32). In addition to the full dose-response curve of *tert*-butylhydroquinone (t-BHQ) analyzed in the AREc32 assay once, a serial dilution (1:2) of t-BHQ ($10^{-5}$ M) was included on every 96-well plate in order to ensure a comparable sensitivity of the cells of different passages.

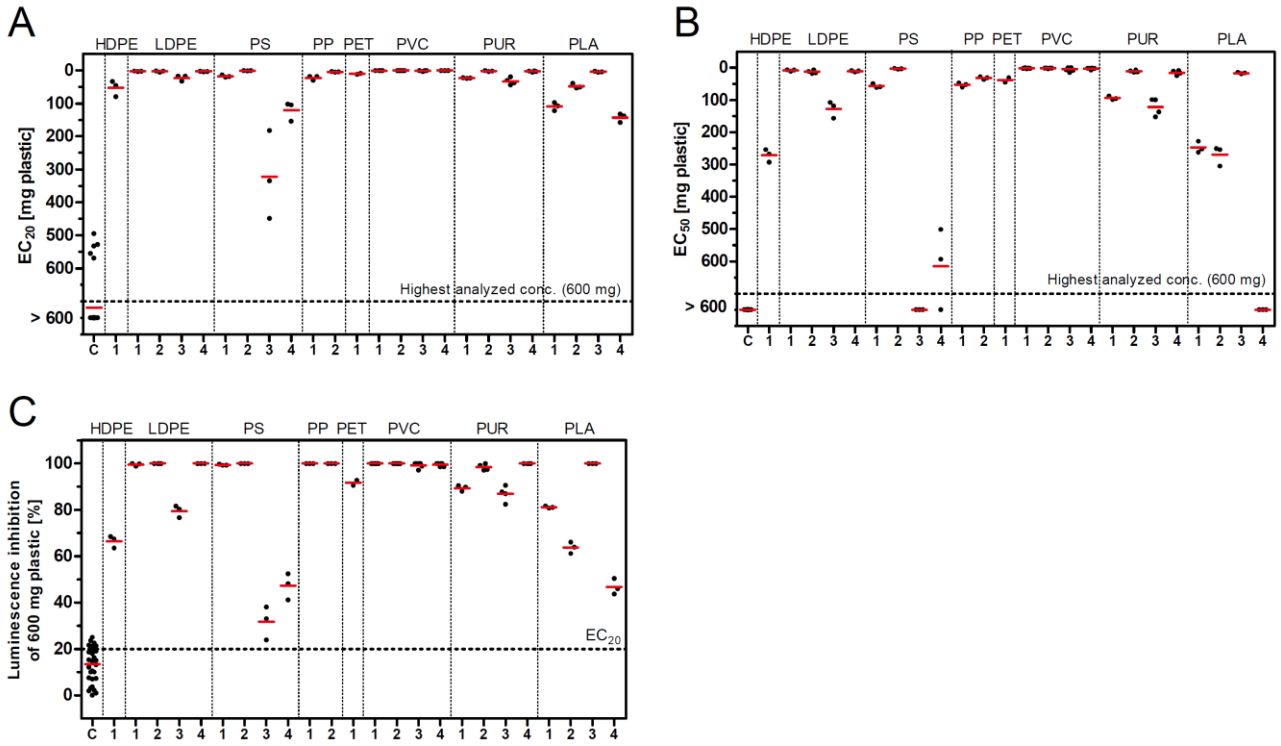



**Figure S4.** Baseline toxicity of plastic migrates in the Microtox assay presented as means (lines) $EC_{20}$ (*A*) and $EC_{50}$ (*B*) baseline toxicity as well as luminescence inhibition (*C*). >600 indicates that the migrate of 600 mg plastic well$^{-1}$ did not inhibit the bioluminescence by >20 or 50 %, respectively.

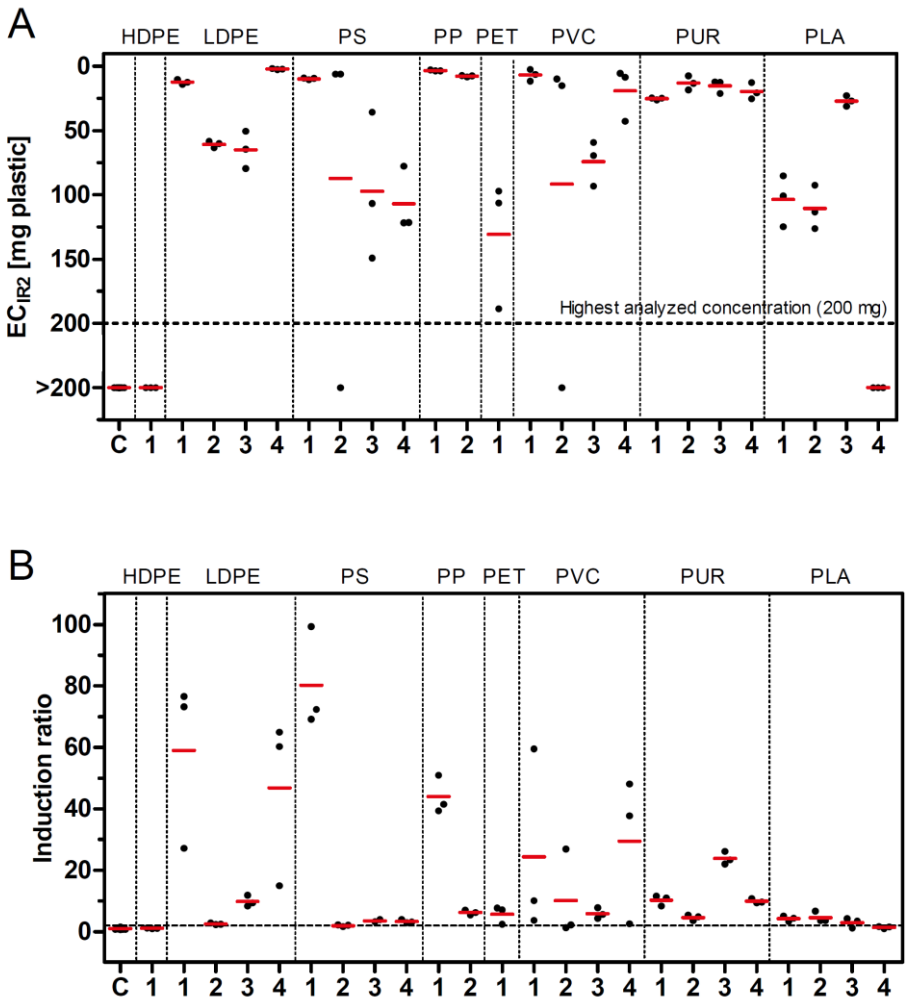



**Figure S5.** Oxidative stress response induced by migrates in the AREc32 assay as mean (lines) $EC_{IR2}$ (*A*) and induction ratios of the highest measured non-cytotoxic concentrations (*B*, see Table S5) >200 indicates that migrates of 200 mg plastic well$^{-1}$ (highest analyzed concentration) did not exceed an induction ratio of 2.

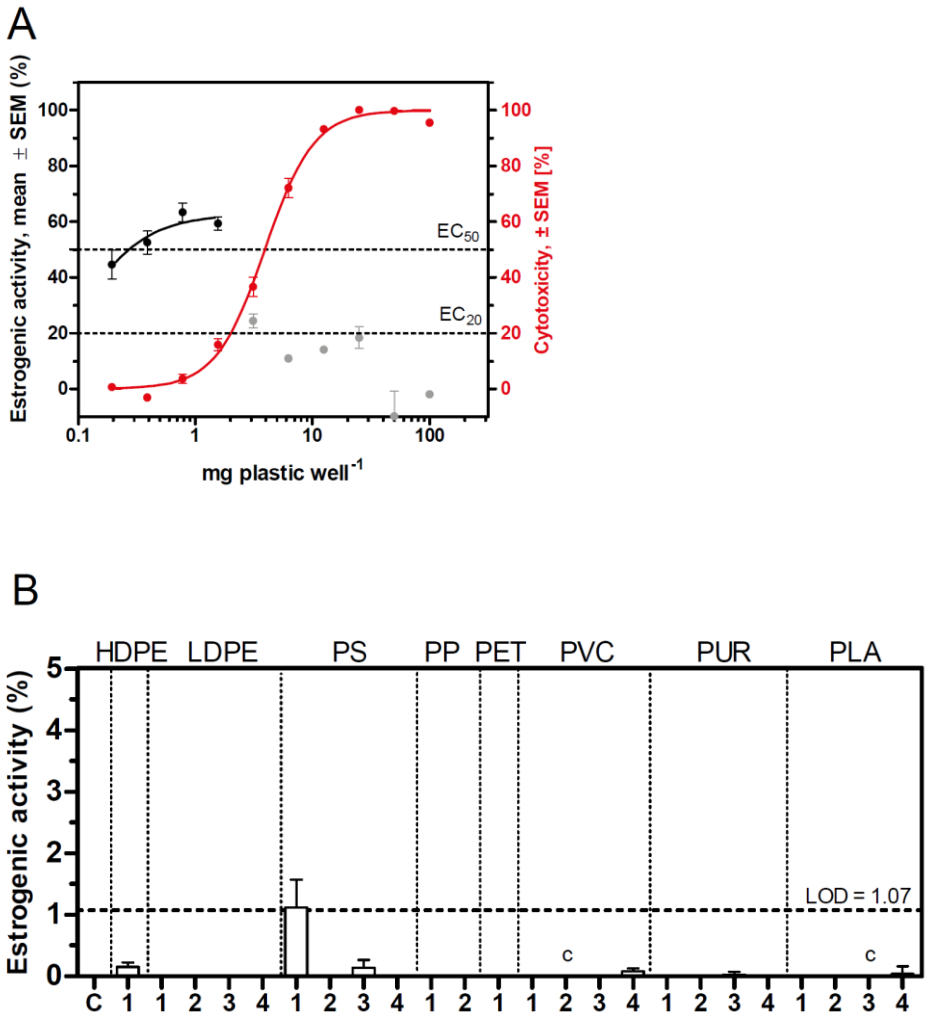
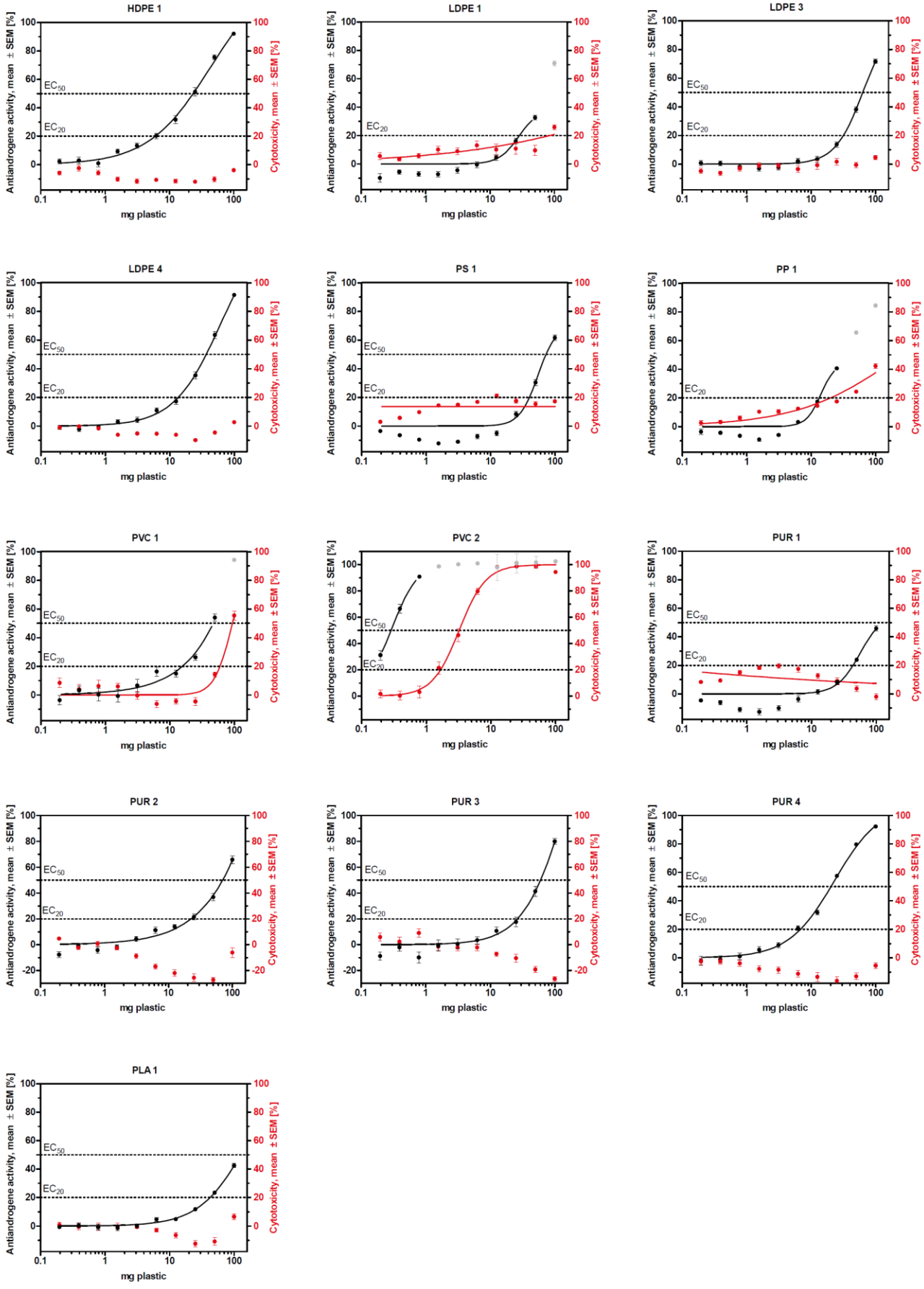



**Figure S6.** Relative estrogenic activities in the Yeast Estrogen Screen (*n* = 16–24). (*A*) Activity data of non-cytotoxic (black) and cytotoxic concentrations (c, grey) of PVC 2, the only migrate with estrogenic activity (mean effects > LOD, Table S6). (*B*) Relative receptor activation of aqueous migrates from 3.00 mg plastic well$^{-1}$.

A


HDPE 1
LDPE 1
LDPE 3
LDPE 4
PS 1
PP 1
PVC 1
PVC 2
PUR 1
PUR 2
PUR 3
PUR 4
PLA 1
Antiandrogene activity, mean ± SEM [%]
Cytotoxicity, mean ± SEM [%]
mg plastic
EC50
EC20

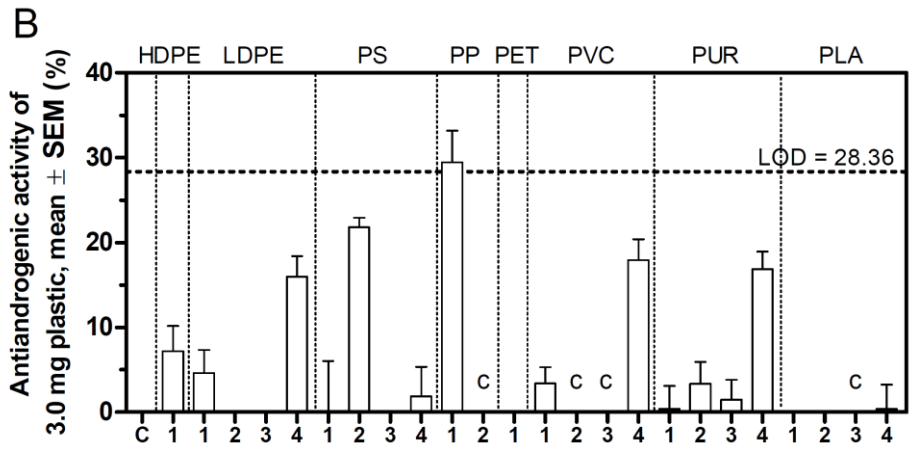



**Figure S7.** Relative antiandrogenic activities in the Yeast Antiandrogen Screen ($n$ = 16–32). (*A*) Activity data of migrates which were antiandrogenic (mean effects > LOD) at 100 mg plastic well$^{-1}$ presented with their cytotoxic (c) concentrations (grey) and non-cytotoxicity concentrations (black). (*B*) Aqueous migrates from 3.00 mg plastic well$^{-1}$.

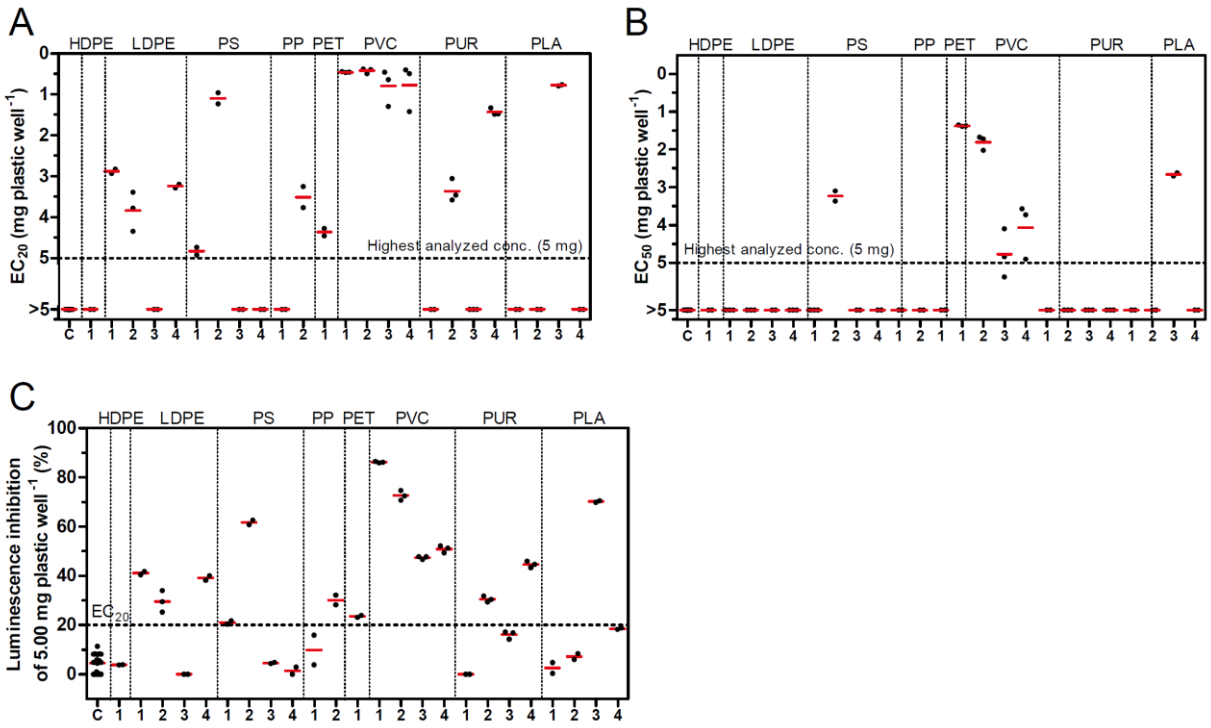
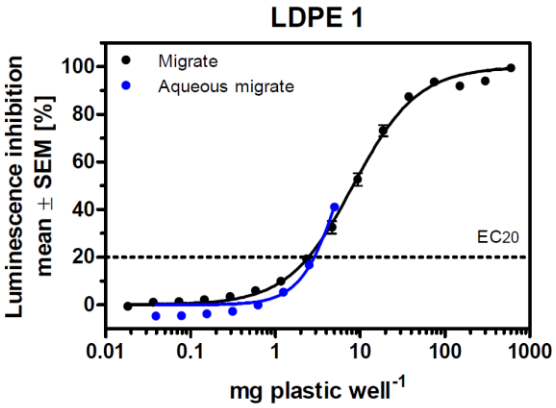
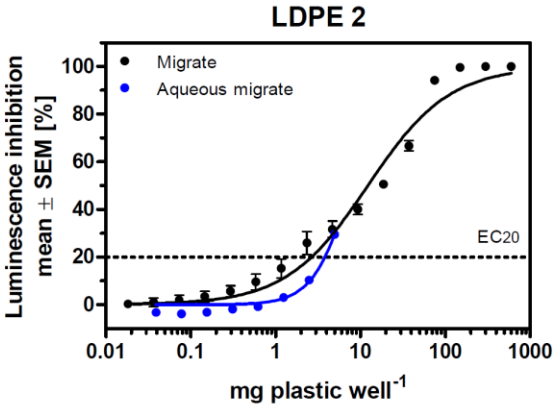
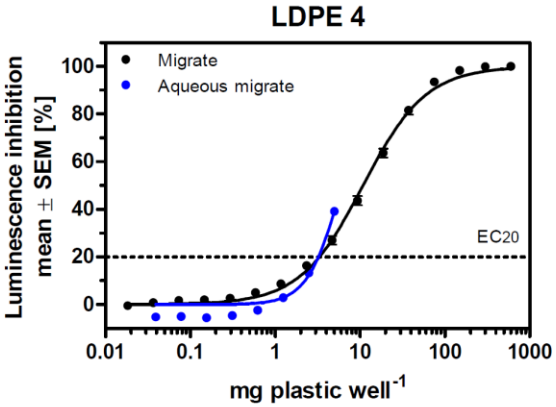
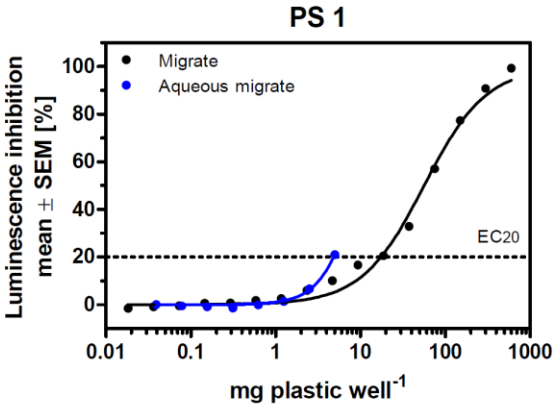


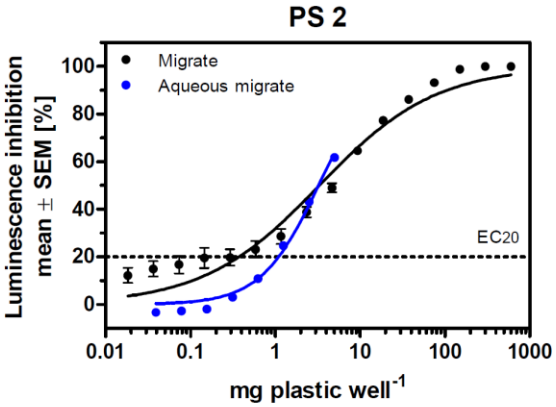

**Figure S8.** Baseline toxicity of aqueous plastic migrates in the Microtox assay presented as means (lines) $EC_{20}$ (*A*) and $EC_{50}$ (*B*) baseline toxicity as well as luminescence inhibition (*C*) from two to three independent experiments (dots). >5 indicates that migrates of 5.00 mg plastic well$^{-1}$ did not inhibit the bioluminescence by > 20% or 50 %, respectively.

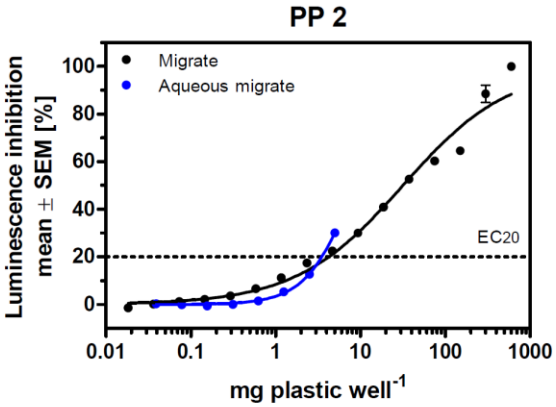

LDPE 1
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

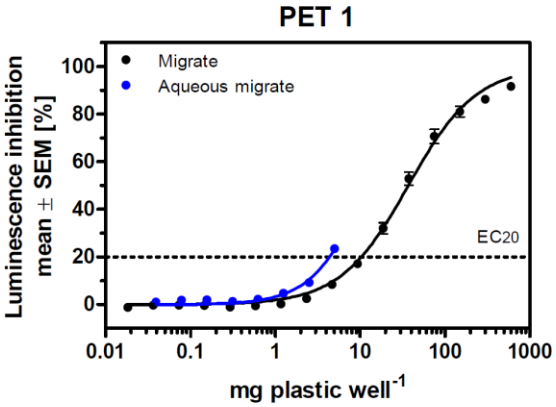

LDPE 2
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

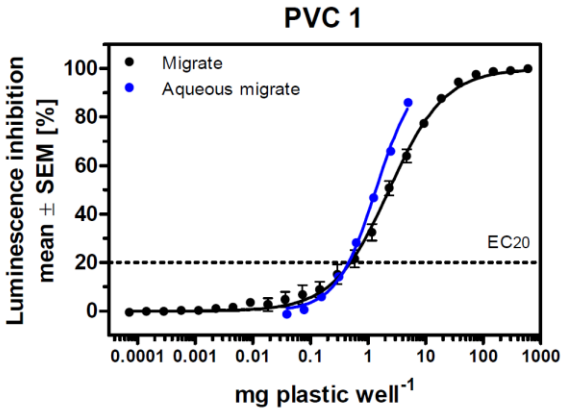

LDPE 4
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

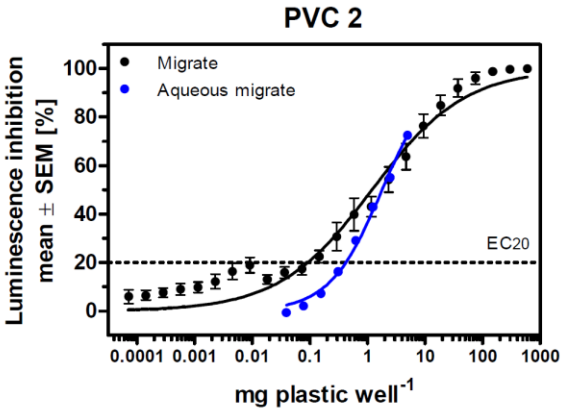

PS 1
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

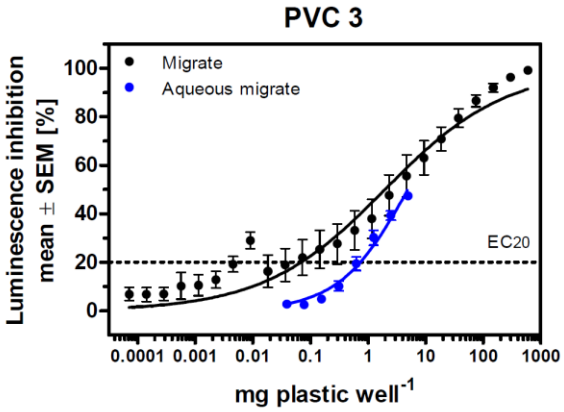

PS 2
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

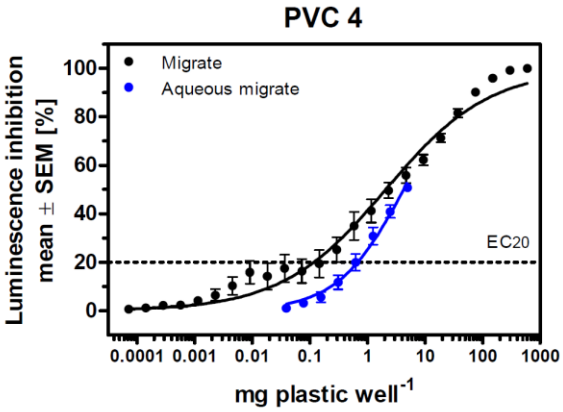

PP 2
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

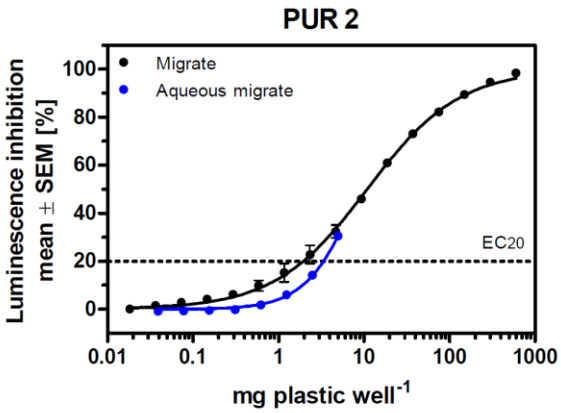

PET 1
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

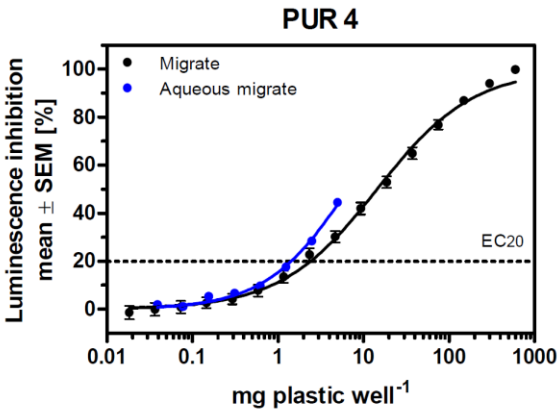

PVC 1
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1


PVC 2
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1


PVC 3
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1


PVC 4
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1


PUR 2
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1


PUR 4
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]
mg plastic well-1

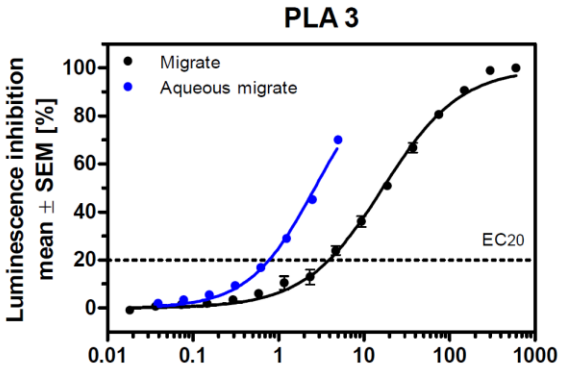

PLA 3
Migrate
Aqueous migrate
EC20
Luminescence inhibition mean ± SEM [%]

**Figure S9.** Concentration-response relationship for baseline toxicity of migrates (with SPE) and aqueous migrates (without SPE). Only samples for which both, migrates and aqueous migrates, induced luminescence inhibition ≥ 20 % at measured concentrations are shown. Data is presented as mean ± standard error of the mean (SEM). $EC_{20}$, effect concentration inducing 20 % baseline toxicity.

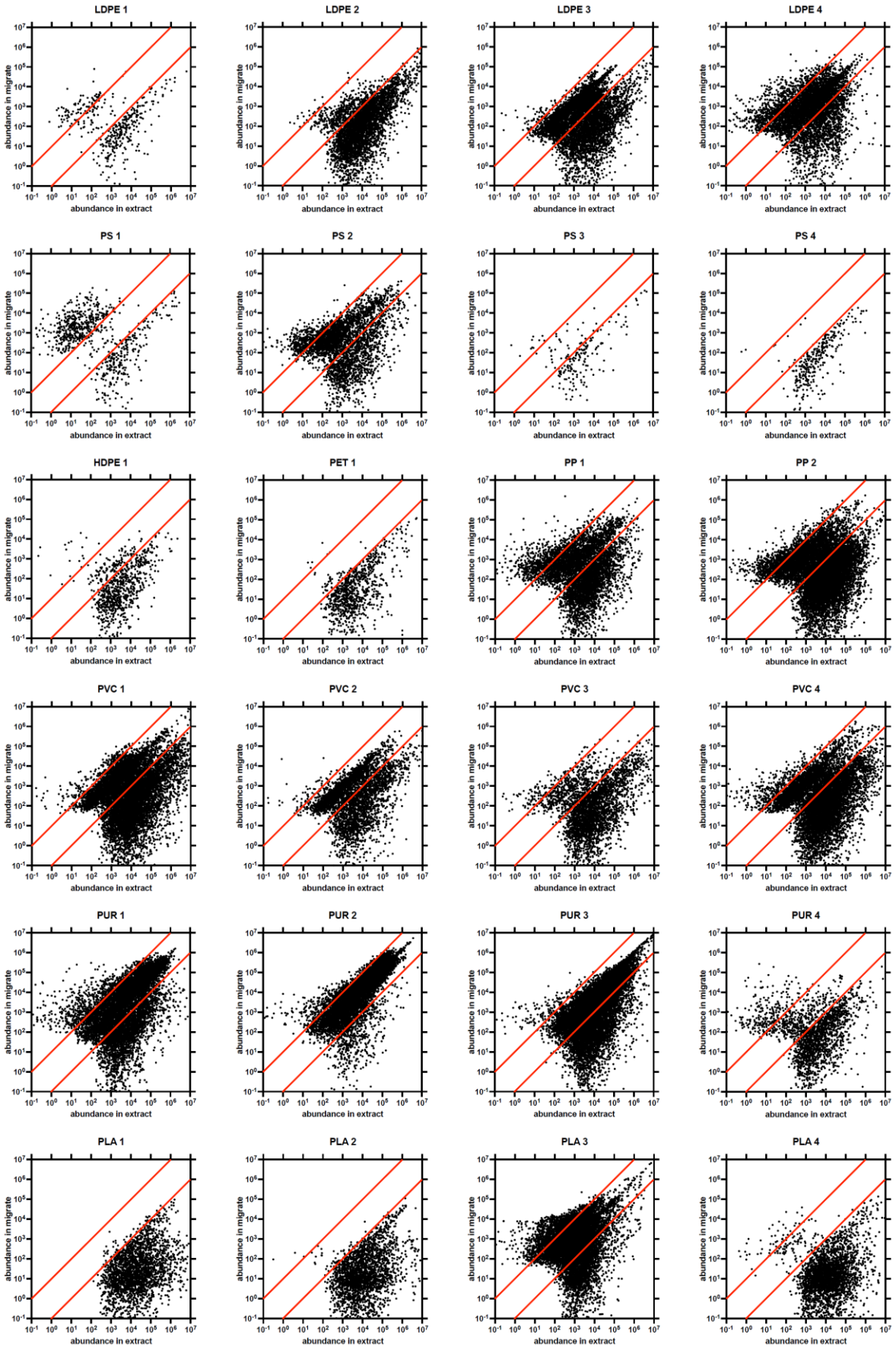



**Figure S10.** Comparison of the abundance of chemical features detected in the extracts and migrates of each plastic product. Red lines highlight the band of an abundance ratio of 0.1–10.

## Supplementary references

(1) European Commission (EC). Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come into contact with food, 2014.

(2) Groh, K., Schymanski, E. S48 | CCPDBLISTA | Database of Chemicals likely (List A) associated with Plastic Packaging (CPPdb) (Version NORMAN-SLE-S48.0.1.0) [Data set]. Zenodo, 2019.

(3) Groh, K., Schymanski, E. S49 | CCPDBLISTB | Database of Chemicals possibly (List B) associated with Plastic Packaging (CPPdb) (Version NORMAN-SLE-S49.0.1.0) [Data set]. Zenodo, 2019.

(4) Alygizakis, N., Slobodnik, J. S32 | REACH2017 | >68,600 REACH Chemicals (Version NORMAN-SLE-S32.0.1.4) [Data set]. Zenodo, 2018.