

Supporting Material for

Investigation of seasonal changes in flame retardant concentrations in car interiors

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Supporting materials containing 4 texts, 8 figures and 8 tables

Contents

List of Texts

Text S1: Extraction and clean-up	S8
Text S2: Instrumental methods.....	S10
Text S3: QA/QC.....	S11
Text S4: Modelling of vehicle conditions	S13
References	S17

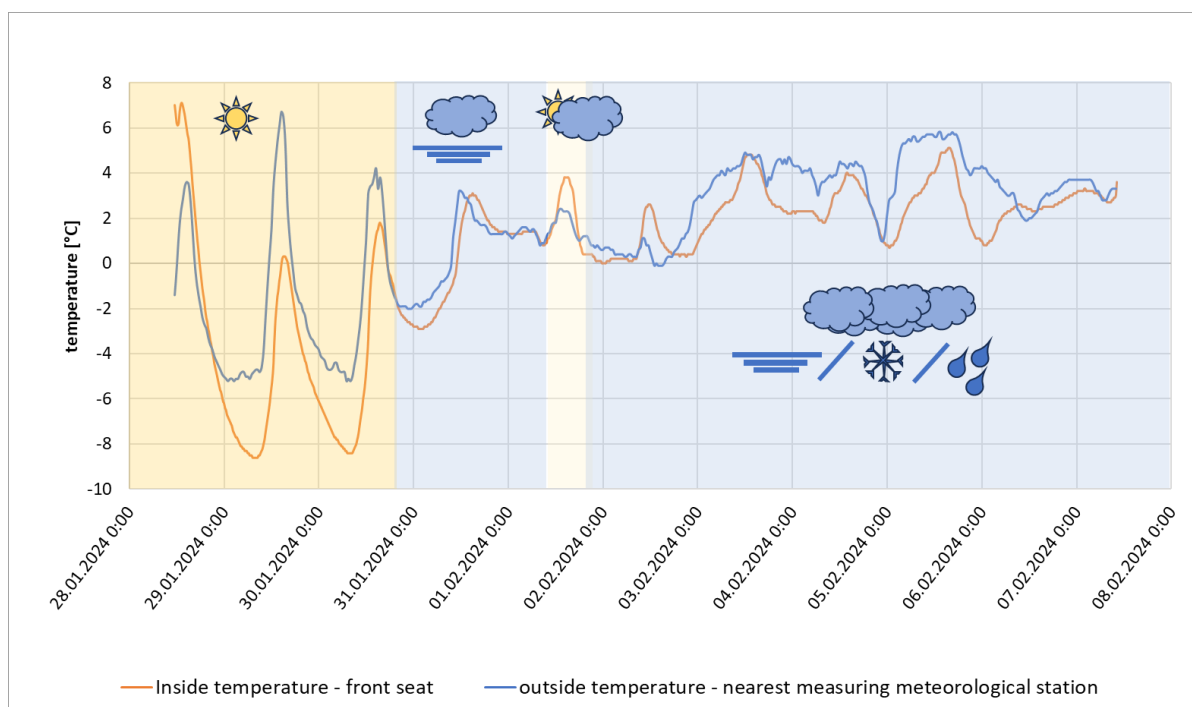
List of Figures

Figure S1 Temperatures measured inside car during sampling period, and outside temperature for: a) winter 10 day exposure in shade in January/February 2024 in Špindlerův Mlýn, Czechia, b) summer 5 day exposure on sun in September 2024 in Brno, Czechia	S3
Figure S2 Polyurethane (PUF) disk used as passive sampler	S4
Figure S3 Dust sampling head and quartz microfiber filter containing vehicle dust sample	S4
Figure S4 Forensic filter sampling head used for dust sampling	S4
Figure S5 Car materials sampled: a) seat foam from back site of front seat, b) hard plastic from right rear door panel, c) hard plastic from middle panel, hard and soft plastic layer from dashboard	S7
Figure S6 Collected car material for analysis, from left, four samples of seat foam, middle panel, door panel, soft plastic layer of dashboard, hard plastic part of dashboard and fibres	S8
Figure S7 Estimated percentage of FRs found in gas phase in air according to seasonal temperature variations within the vehicle.....	S13
Figure S8 Comparison between estimated equilibrium dust concentrations based on air concentrations in the vehicle, and measured dust concentrations.....	S14

List of Tables

Table S1 Summary of the sampling strategy	S5
Table S2 List of target compounds, abbreviations and identifiers.....	S6
Table S3 Analytical standards and suppliers	S10
Table S4 Masses determined in blanks, instrumental detection limits (iLODs), instrumental quantification limits (iLOQs) and method detection limits (MDLs).....	S11
Table S5 Temperature dependent logK _{OA}	S14
Table S6 Car air concentrations in both seasons.....	S16
Table S7 Car dust concentrations in both seasons.....	S16
Table S8 Car interior material concentrations	S16

a)



b)

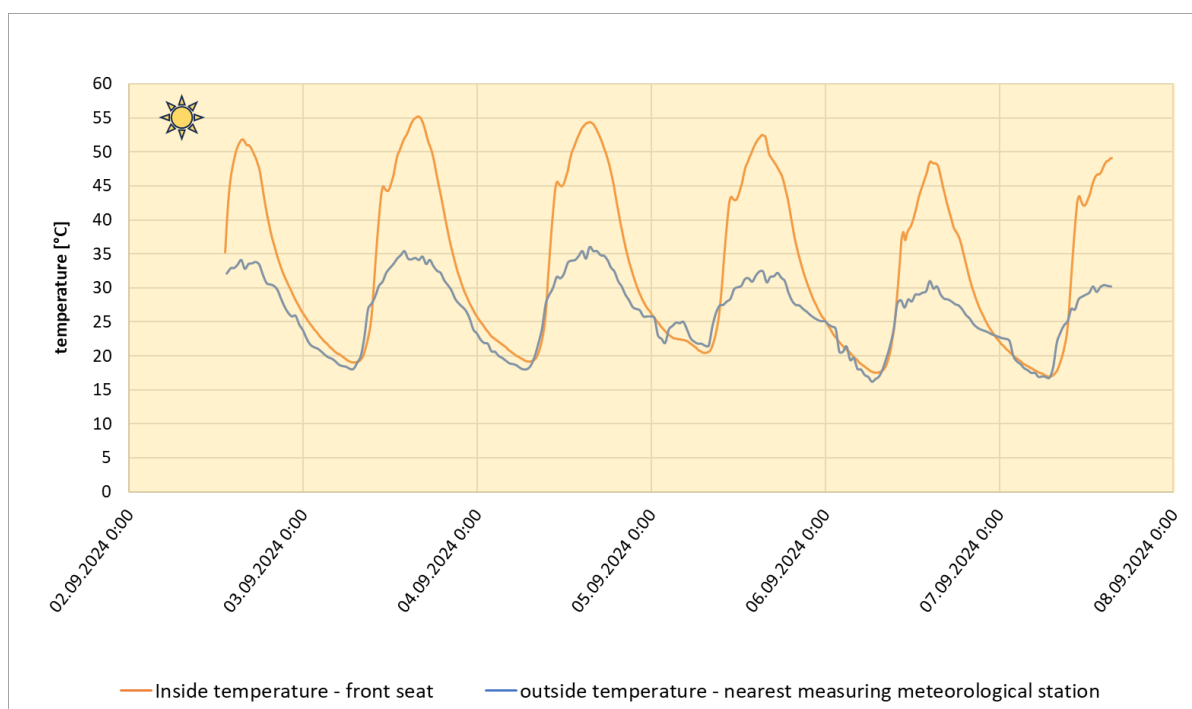


Figure S1 Temperatures measured inside car during sampling period (measured near the gear shift lever, always in the shade) at 15 min intervals, and outside temperature (data from the nearest weather station) in 30 min intervals with weather for: a) winter 10 day exposure in shade in January/February 2024 in Špindlerův Mlýn, Czechia, b) summer 5 day exposure on sun in September 2024 in Brno, Czechia



Figure S2 Polyurethane (PUF) disk used as passive sampler



Figure S3 Dust sampling head and quartz microfiber filter containing vehicle dust sample

a)



b)



Figure S4 a) and b) Special forensic filter sampling head used for dust sampling

Table S1 Summary of the sampling strategy

Sample type	Sample	Sample description/location	Outdoor air temperature, location	Average inside temperature	Deployment dates (sampling duration)
Air	Winter	PUF disk - driver's mirror - wire suspension (in shade)	<10°C, Špindlerův Mlýn, CZ	0.54°C	28.1.-7.2.2024 (10 days)
	Summer	PUF disk - driver's mirror - wire suspension (exposed to sun)	16-34.6°C, Brno, CZ	33.2°C	2.9.-7.9.2024, (5 days)
Dust	Winter – sample 1	Pre-experiment dust: Collected at start of winter experiment (dashboard + front seats) [#]	<10°C, Špindlerův Mlýn, CZ	NA	28.1.2024
	Winter – sample 2	Experiment phase dust: Collected after exposure to winter shade (dashboard + front seats) [#]	<10°C, Špindlerův Mlýn, CZ	0.54°C	7.2.2024
	Summer – sample 1	Pre-experiment dust: Collected at start of summer experiment (dashboard + seats)	16-34.6°C, Brno, CZ	NA	2.9.2024
	Summer – sample 2	Experiment phase dust: Collected after exposure to summer sun (dashboard + seats)	16-34.6°C, Brno, CZ	33.2°C	7.9.2024
Car interior materials	Soft plastic	Dashboard right - side, first layer of dashboard, soft plastic cover	Brno, CZ		1.10.2024
	Hard plastic	Dashboard right, side, hard plastic part of dashboard (plastic part nearest to window, with air holes) + (Door, back, right) + (Central plastic part in backside (between front seats)	Brno, CZ		1.10.2024
	Textile fibres	from connected sewn parts, fibres sticking out of the fabric due to wear	Brno, CZ		1.10.2024
	Foam	Left front seat (back seat part)	Brno, CZ		1.10.2024
		Right front seat (back seat part)	Brno, CZ		1.10.2024
		Left back seat (under seat part)	Brno, CZ		1.10.2024
		Right back seat (under seat part)	Brno, CZ		1.10.2024

[#] In the winter, dust was vacuumed only from the front half of the car due to a vacuum cleaner malfunction

Table S2 List of target compounds, abbreviations and identifiers

	Abbreviation	Compound name	CAS RN	InChIKey	Molecular formula	Avg mass
OPES	TCEP	Tris(2-chloroethyl) phosphate	115-96-8	HQUQLFOMPYWACS-UHFFFAOYSA-N	C6H12Cl3O4P	285.48
	TCIPP	Tris(2-chloroisopropyl)phosphate	13674-84-5	KVMPUXDNESXNOH-UHFFFAOYSA-N	C9H18Cl3O4P	327.56
	TDCIPP	Tris(1,3-dichloro-2-propyl) phosphate	13674-87-8	ASLWPAWFJZFCKF-UHFFFAOYSA-N	C9H15Cl6O4P	430.89
	TBOEP	Tris(2-butoxyethyl) phosphate	78-51-3	WTLBZVNBKMDVP-UHFFFAOYSA-N	C18H39O7P	398.477
	TPHP	Triphenyl phosphate	115-86-6	XZZNDPSIHUTMOC-UHFFFAOYSA-N	C18H15O4P	326.288
	CDP	Cresyl diphenyl phosphate	26444-49-5	NA	C19H17O4P	340.086
	oTMPP	Tri-o-cresyl phosphate	78-30-8	YSMRWXYRXBRSND-UHFFFAOYSA-N	C21H21O4P	368.369
	TDBPP	Tris(2,3-dibromopropyl) phosphate	126-72-7	PQYJRMFWJJONBO-UHFFFAOYSA-N	C9H15Br6O4P	697.613
	TnPP	Tripropyl phosphate	513-08-6	RXPQRKFMDQNODS-UHFFFAOYSA-N	C9H21O4P	224.237
	ip-TPP	Triphenyl phosphates isopropylated	68937-41-7	NA	C27H33O4P	452.212
	TEHP	Tris(2-ethylhexyl) phosphate	78-42-2	GTVWRXDRKAHEAD-UHFFFAOYSA-N	C24H51O4P	434.642
	TEP	Triethyl phosphate	78-40-0	DQWPFSLDHJDLRL-UHFFFAOYSA-N	C6H15O4P	182.156
	TBP	Tributyl phosphate	126-73-8	STCOOQWBFONSKY-UHFFFAOYSA-N	C12H27O4P	266.318
PBDE	EHDPP	2-Ethylhexyl diphenyl phosphate	1241-94-7	CGSLYBDCEGBZCG-UHFFFAOYSA-N	C20H27O4P	362.406
	BDE 209	2,2',3,3',4,4',5,5',6,6'-Decabromodiphenyl ether	1163-19-5	WHHGLZMJPIXIBX-UHFFFAOYSA-N	C12Br10O	959.17

a)



b)



c)



d)



Figure S5 Car materials sampled: a) seat foam from back site of front seat, b) hard plastic from right rear door panel, c) hard plastic from middle panel, and d) hard and soft plastic layer from dashboard

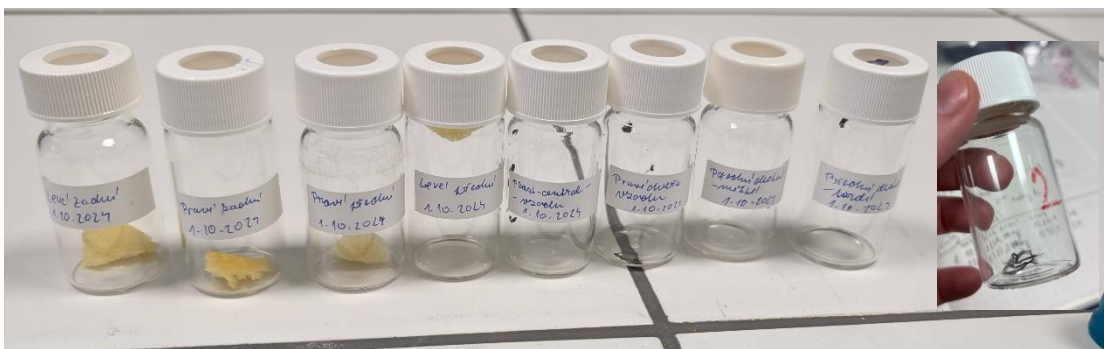


Figure S6 Collected car material for analysis, from left, four samples of seat foam, middle panel, door panel, soft plastic layer of dashboard, hard plastic part of dashboard and fibres. Plastic pieces in vials 5-8 are on the upper inside walls of the glass.

Text S1: Extraction and clean-up

Dust and car material samples

Before extraction, the filters containing the dust samples were ground using a Ball Mill (Retsch MM 301) and kept at -4°C between processing steps.

OPEs

The extraction of 1 mg of dust and car material samples (whole mass used) for the determination of OPEs was carried out using methanol. Weighed dust and products were transferred into 20 ml glass vials. 50 μl of recovery standard (50 μl deuterated OPEs) was added to all samples. Standards and suppliers are given in Table S3. Next, 3 ml of methanol (methanol absolute, LC-MS grade, Biosolve) was added to all samples, then ultrasonic extraction was performed for 20 min, and after 20 min of sedimentation, the extract was transferred to new vials. The methanol extraction step was performed 3 times.

The extraction was followed by concentration using a flow of nitrogen and heating to 35°C per 1 ml of extract. The samples were then purified using a nylon filter (Chromafil Xtra PA-45/13; Macherey-Nagel), transferred to minivials, and then again reduced to 0.5 ml using nitrogen flow and a temperature of 35°C . Methanol was added to the samples to achieve a sample volume of 0.5 ml of methanol. Then, 0.5 ml of Milli-Q water was added. The samples were stored at 4°C until analysis.

BDE 209

Extraction of up to 100 mg dust and car material samples for the determination of BDE 209 was performed using 1:1 n-hexane-acetone (hex:acn) (n-hexane: Baker analyzed for pesticide residue analysis; J.T.Baker; acetone, Baker analyzed for pesticide residue analysis; J.T.Baker). Samples were transferred to 20 ml glass vials. 50 μl of recovery standard ($^{13}\text{C}_{12}$ -BDE 209) was added to all samples. Next, 5 ml of hex:acn was added to all samples, and then ultrasonic extraction was performed for 10 min, and after 10 min of sedimentation, the extract was transferred to new vials. The hex:acn extraction step was performed 3 times, but in the second and third replicates, only 3 ml of hex:acn was used for each replicate. The extraction was followed by a gentle concentration using a flow of nitrogen and heating to 32°C per 5 ml of extract.

PBDE samples were concentrated to 0.5-1 ml before purification via column chromatography.

The purification took place in a column using 1 g of non-activated silica gel, 5 g of sulfuric silica gel (50 g of activated silica gel mixed with 22 ml of 96% H₂SO₄ -- Puriss. p.a., for determination of Hg, ACS Reagent, Reag. ISO, Reag. Ph. Eur., Reag. USP, 95.0-97.0%, Honeywell Fluka), 1 g of activated silica gel and pre-cleaned cotton wool. The extract was added directly to non-activated silica gel and then eluted with 30 ml of a 1:1 hexane: dichloromethane mixture.

50 µl of nonane (Picograde, Promochem) was added as a final solvent. Samples were concentrated using nitrogen flow and heating at 32°C to 0.5 ml of extract. Then, the samples were transferred to conical minivials, and after rinsing the previous vials, the samples were concentrated again, using nitrogen flow and heating at 32°C, down to a volume of 50 µl.

Internal standards were then added to the volume-reduced samples. 10 µl of mixture 13C12-BDE 77 and 13C12-BDE 138 was added to quantify recoveries of PBDEs. The samples were stored at 4 °C until analysis.

Passive air samplers

After collection, polyurethane foam (PUF) disks were cut in half with pre-cleaned scissors and were weighed. One half of a disk was used for OPEs extraction, and one for BDE 209.

OPEs

50 µl of recovery standard (50 µl deuterated OPEs; Table S3) was added to the half PUF disk placed in a glass patron. The extraction solvent was methanol. A 70 min Soxhlet extraction was performed, followed by 20 min rinsing and drying to the final volume 10 ml. The extract was transferred to 20 ml vials and stored at 4°C before clean-up. Before clean-up, samples were reduced in volume to 1 ml of extract using a flow of nitrogen and heating to 35 °C.

The purification and subsequent processing for the analysis of OPEs were the same as for the dust and material samples described above.

BDE 209

50 µl of recovery standard (13C12-BDE 209) was added to the half PUF disk placed in a glass patron. The extraction solvent was dichloromethane. A 40-minute Soxhlet extraction was performed, followed by 20 min rinsing and drying to the final volume of 10 ml. The extract was transferred to 20 ml vials and stored at 4°C before clean-up. Before clean-up, samples were reduced to 0.5-1 ml in volume using a flow of nitrogen and heating to 32 °C.

The purification and subsequent processing for the analysis of BDE 209 were the same as for the dust and material samples described above.

Table S3 Analytical standards and suppliers

	Standard	Concentration [ng/μl]	Spike volume [μl]	Supplier
OPEs	D27-TnBP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D33-TIPPP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D15-TEP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D18-TCPP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D12-TCEP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D15-TDCPP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D15-TPHP	0.4	50	Cambridge Isotope Laboratories, Inc.
	D51-TEHP	0.4	50	Cambridge Isotope Laboratories, Inc.
PBDE	13C12 PBDE 209	0.1	50	Wellington Laboratories, Canada
	13C12-BDE 77	0.1	10	Wellington Laboratories, Canada
	13C12-BDE 138	0.1	10	Wellington Laboratories, Canada

Text S2: Instrumental methods

OPEs

Selected OPEs were analyzed using an Agilent 1290 Infinity HPLC (Agilent Technologies, Inc., Santa Clara, CA, USA) equipped with an Aquity BEH C18 size 2.1 mm × 100 mm, 1.7 μm column (Waters Corporation, Milford, MA, USA). The column was held at 30 °C in a column oven. The injection volume was 3 μl. Separation was achieved using 0.1% water solution of formic acid (mobile phase A) and 0.1% formic acid in methanol (mobile phase B) at a flow rate of 0.2 mL/min. Analyte detection was performed on an Agilent 6495 MS (Agilent Technologies, Inc., Santa Clara, CA, USA) operating in positive electrospray ionization mode (ESI+) with the following parameters: 2700 V, a heated source at 400 °C and nitrogen as sheath gas. ¹³C or deuterium labelled TPHP, TnBP, TDCIPP and TnPP isotope dilution method was used for the quantification of the analytes. The linear range (MRM mode) was 0.09 – 90 μg/L, with limits of quantification from 0.01 to 0.79 μg/L for respective OPEs.

BDE 209

BDE 209 was analyzed using an Agilent 7890A GC (Agilent Technologies, Inc., Santa Clara, CA, USA) equipped with RTX-1614 size 15 m × 0.25 mm × 0.10 μm column (Restek, Inc., France), coupled to Waters AutoSpec Premier MS (Waters Corporation, Milford, MA, USA). The GC temperature program started at 80°C (1 min hold), increased at a rate of 20°C/min to 250°C (0 min hold), followed by an increase of 1.5°C/min to 260°C (2 min hold) and finally by 25°C/min to 320°C (4.5 min hold). The GC/MS interface and ion source temperatures were 280

and 250°C, respectively. The injected sample volume was 2 µl at 280°C in pulsed splitless mode. Helium was used as a carrier gas at 1 mL/min and 1.4 mL/min after 15 min. The MS was operated in EI+ and SIM mode at the resolution of >10000. For BDE-209, the resolution was set to >5,000.

Text S3: QA/QC

Method detection limits (MDLs) were determined based on the blank filter concentrations and were calculated as the mean of the blanks + 3* standard deviation of the blanks. If all three blanks were below the instrument detection levels, the instrument detection limit was used as the MDL. For statistical analyses, 0.5*MDL was substituted for samples that were <MDL. Samples >MDL were corrected by subtracting the mean of the blanks. Blank masses are given in Table S4.

To calculate the concentration in the air, we used a sampling rate of 1.8 m³/day,¹ with winter exposure lasting 10 days (240 h) and summer exposure lasting 5.09 days (122.17 h). The base sampling rate was adjusted based on the average sampling period temperature using the GAPS template.²

*Table S4 Masses determined in blanks, instrumental detection limits (iLODs), instrumental quantification limits (iLOQs) and method detection limits (MDLs) calculated as the average of the blanks+3*SD.*

Sample types	Compound group	Compound	Average blank mass [ng/sample]	iLOD [ng/sample]	iLOQ [ng/sample]	MDL [ng/sample]
Air (PUF blanks)	PBDEs	BDE 209	36.2	-	45.9 [#]	36.2
	OPEs	TCEP	8.1	0.465	1.55	21.3
		TCIPP	403	0.869	2.9	559
		TDCIPP	<iLOQ	0.0858	0.286	-
		TBOEP	0.245	0.0257	0.0858	0.26
		TPhP	2.28	0.0158	0.0527	5.39
		oTMPP	0.0511	0.021	0.0699	0.173
		CDP	0.964	0.00919	0.0306	1.55
		TEP	10.4	0.187	0.622	12.7
		TBP	39.8	0.0178	0.0595	40.8
Dust samples (Filter blanks)	PBDEs	BDE 209	60.1	-	81.2 [#]	60.1
	OPEs	TCEP	2.76	0.465	1.55	6.67
		TCIPP	14.9	0.869	2.9	54.1
		TDCIPP	<iLOD	0.0858	0.286	0.0429
		TBOEP	0.2	0.0257	0.0858	0.413
		TPhP	0.12	0.0158	0.0527	0.31
		oTMPP	0.0618	0.021	0.0699	0.104
		CDP	<iLOD	0.00919	0.0306	0.00459
		TEP	128	0.187	0.622	244
		TBP	94	0.0178	0.0595	446
Product samples (solvent blanks)	PBDEs	BDE 209	-	-	77.2 [#]	-
	OPEs	TCEP	0.92	0.465	1.55	1.41
		TCIPP	<iLOQ	0.869	2.9	2.97
		TDCIPP	<iLOD	0.0858	0.286	0.0429
		TBOEP	0.074	0.0257	0.0858	0.209
		TPhP	0.0614	0.0158	0.0527	0.119
		oTMPP	0.0618	0.021	0.0699	0.0862

		CDP	0.0326	0.00919	0.0306	0.151
		TEP	1.9	0.187	0.622	2.29
		TBP	1.55	0.0178	0.0595	1.78

#average

Text S4: Modelling of vehicle conditions

We estimated K_p , the mass of SVOC on airborne particles per particle mass, based on equation S1.³ We used the fraction of organic matter associated with airborne particles, f_{om_part} , of 0.4,⁴ and a density of airborne particles, ρ_{part} , of 1000000 g/m³,⁵ and octanol-air partitioning coefficients, K_{OA} , adjusted based on the average temperature of each seasonal experiment, i.e. 0.54°C for winter and 33.22°C for summer, (Table S5):

$$K_p = \frac{f_{om_part} \times K_{OA}}{\rho_{part}} \quad (\text{Eq. S1})$$

Based on the bulk air concentrations measured by the PUF, we used equation S2 to estimate the gas-phase fraction of bulk air,⁶ assuming a value 20 µg/m³, for TSP and the calculated values of K_p :

$$C_g = \frac{C_g + C_p}{1 + (TSP \times K_p)} \quad (\text{Eq. S2})$$

$X_{dust,pred}$, the predicted dust concentration based on gas-phase air concentrations, was then calculated using equation S3:⁶ where the organic matter fraction of dust, f_{om_dust} , was set to 0.2 and dust density, ρ_{dust} , was set to 2000000 g/m³ ⁵:

$$X_{dust,pred} = \frac{f_{om_dust} \times K_{OA} \times C_g}{\rho_{dust}} \quad (\text{Eq. S3})$$

The estimated gas-phase fraction of FRs in vehicle air in winter and summer is displayed in Figure S7. The results show the strong estimated effect of temperature shifts within the car on distributions of chemicals in air, and notably, the differences are even larger when peak in-vehicle summer temperatures are considered.

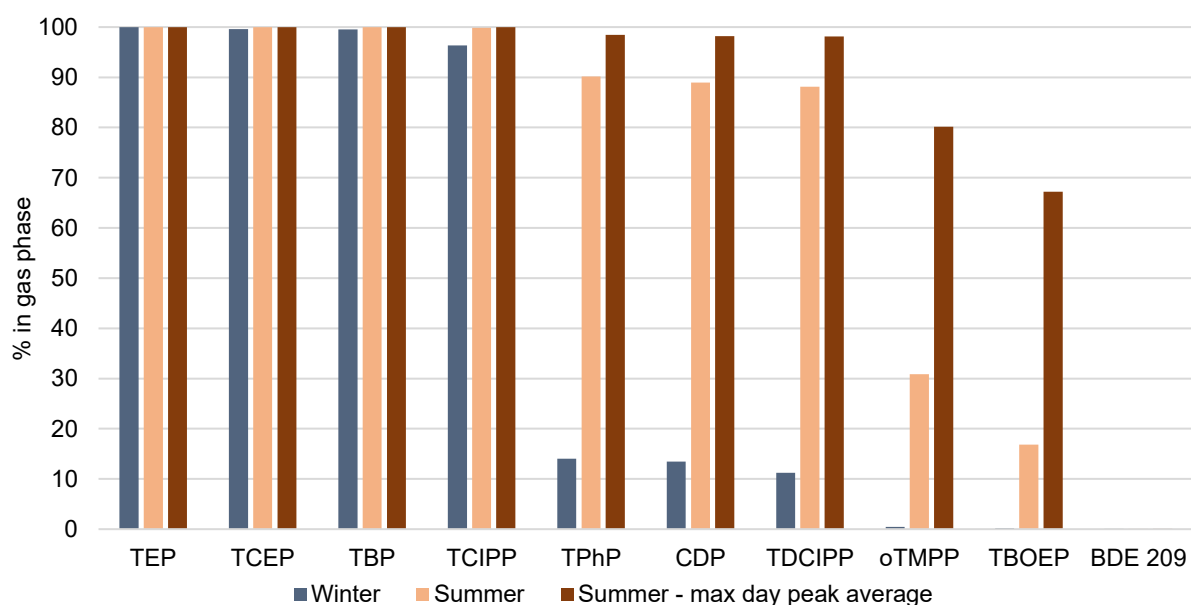


Figure S7 Estimated percentage of FRs found in gas phase in air according to seasonal temperature variations within the vehicle.

Table S5 Temperature dependent $\log K_{OA}$, determined from coefficients in Harner² Abdollahi et al.,⁷ Wang et al.,⁸ and Li et al.⁹

Compound	Winter $\log K_{OA}$, 0.54 °C	Summer $\log K_{OA}$, 33.22 °C
TEP	6.36	5.54
TCEP	8.68	7.47
TBP	8.79	7.56
TCIPP	9.67	8.30
TPhP	11.88	10.13
CDP ⁸	11.90	10.19
TDCIPP	11.99	10.23
oTMPP ⁸	13.43	11.45
TBOEP	13.87	11.79
BDE 209 ⁹	16.52	14.89

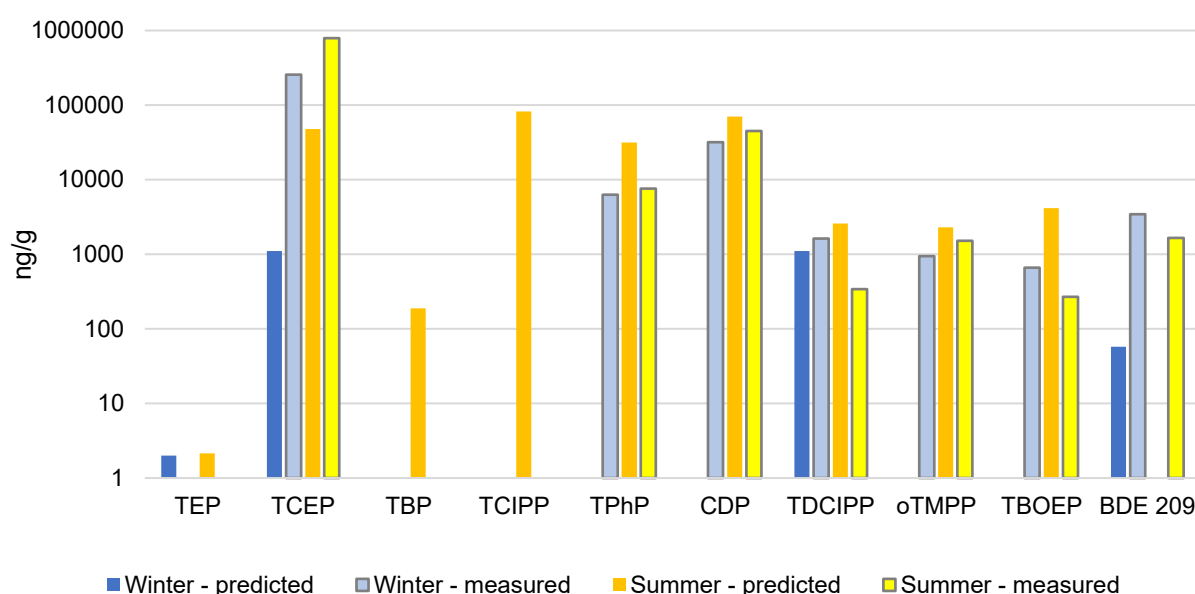


Figure S8 Comparison between estimated equilibrium dust concentrations based on air concentrations in the vehicle, and measured dust concentrations. The discrepancies for a number of compounds suggests that air and dust are not at equilibrium during the two sampling periods.

Sensitivity analysis

For prediction of dust concentration, we tested the impact of 3 variables on the prediction:

1. TSP: 20 $\mu\text{g}/\text{m}^3$,⁶ was a default value not specific to cars. We tested values between 10 and 100 $\mu\text{g}/\text{m}^3$, and the predicted dust concentrations varied by <100%: at 10 $\mu\text{g}/\text{m}^3$ <12% change for TEP, TCEP, TBP and TCIPP in both seasons and <12% change for TPhP, CDP, and TDCIPP in summer, at 100 $\mu\text{g}/\text{m}^3$ <32% change for TPhP, CDP, and TDCIPP in summer and 57-100% change within the 10-100 $\mu\text{g}/\text{m}^3$ range of TSP for all other compounds.
2. Octanol-air partitioning coefficients have a strong temperature dependence, and especially for substances with $\log K_{OA}$ is between log 9.5-12, even a minimal change

will significantly affect the proportion in the gas phase. We tested the sensitivity of the prediction to temperature by comparing the logK_{OA} values predicted at average temperatures with predictions using the peak daytime temperature inside the vehicle. Distributions of TDCIPP, TBOEP, TPhP, oTMPP and CDP are most sensitive to temperature within the range experience in the vehicle. For example, equilibrium modelling of TCIPP distributions at 33.22 °C predicts a dust concentration of 82,000 ng/g, which is substantially higher than the measured dust concentration of 26,000 ng/g. However, estimating equilibrium partitioning at the peak vehicle temperature of 51.93 °C predicts dust concentrations of 18,000 ng/g, much closer to measured concentrations. Using the daytime peak summer temperature resulted in agreement with the measured dust concentrations for TCIPP, TPhP, TDCIPP and TBOEP, however agreement decreased for CDP, oTMPP, TBP, TEP and TCEP. BDE 209 was not affected.

3. PUF passive air samplers, especially without housing, should equally collect gaseous and particle-bound compounds from air, however, we tested the sensitivity of this assumption by repeating the estimates assuming that all concentrations in air were only gas phase. In winter, the predictions for TCEP and TEP were unchanged, however predicted dust concentrations of TDCIPP increased 8x and 260000x for BDE 209. In summer, assuming fully gas-phase compounds in air resulted in no change in predicted dust concentration for TCEP, TCIPP, TDCIPP, TEP and TBP, and a 3-fold increase for oTMPP, and 6-fold increase for TBOEP. However, given the known association of such higher molecular weight compounds with particles in air, this is not expected to be a realistic scenario for BDE-209.

Table S6 Car air concentrations [ng/m³] in both seasons (<iLOQ – below instrumental limit of quantification, <iLOD – below instrumental limit of detection, <MDL – below method detection limit)

Season	TEP	TCEP	TBP	TCIPP	TPhP	CDP	TDCIPP	oTMPP	TBOEP	BDE 209
Winter	8.84	23.4	<MDL	<MDL	<MDL	<iLOD	0.0993	<iLOQ	<MDL	0.00459
Summer	62.4	16300	52.1	4170	25.7	50.7	1.75	0.265	0.4	<iLOQ

Table S7 Car dust concentrations [ng/g] in both seasons; Sample 1 is collected before the experiment period, Sample 2 is collected after the experiment period (<MDL – below method detection limit)

Season	TEP	TCEP	TBP	TCIPP	TPhP	CDP	TDCIPP	oTMPP	TBOEP	BDE 209
Winter - sample 1	<MDL	430000	<MDL	<MDL	8130	47800	783	1480	1380	1840
Summer - sample 1	<MDL	648000	<MDL	<MDL	8780	51500	301	2370	659	2000
Winter - sample 2	<MDL	255000	<MDL	<MDL	6320	31800	1630	944	1090	3440
Summer - sample 2	<MDL	793000	<MDL	<MDL	7610	44900	340	1510	269	1650

Table S8 Car interior material concentrations [ng/g] (<iLOQ – below instrumental limit of quantification, <iLOD – below instrumental limit of detection, <MDL – below method detection limit)

Type of product	Product location/specification	TEP	TCEP	TBP	TCIPP	TPhP	CDP	TDCIPP	oTMPP	TBOEP	BDE 209
PUF	Left front seat (back seat part)	112	386000	132	6390	5150	13.4	<iLOQ	<iLOD	<iLOQ	25.6
	Right front seat (back seat part)	104	640000	108	6010	4380	<MDL	287	<MDL	<MDL	259
	Left back seat (under seat part)	141	296000	368	76500	4370	66.4	<iLOQ	<iLOQ	<MDL	7.46
	Right back seat (under seat part)	384	271000	470	46000	4750	<MDL	<iLOQ	<iLOD	<MDL	7.44
Plastic	soft plastic (soft cover of dashboard)	<MDL	35900	<MDL	42800	<iLOQ	<iLOD	<iLOD	<MDL	<MDL	<iLOQ
	hard plastic (door panel, middle panel, dashboard)	<MDL	15500	<MDL	12200	124	<MDL	<iLOD	<iLOQ	<iLOQ	105
Fabrics	fibers	<MDL	111000	<MDL	2730	121	124	<iLOD	<iLOQ	<MDL	1300

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