Electronic Supplementary Material (ESI) for Environmental Science: Processes & Impacts.

Supplementary Information

Wood combustion particles induce adverse effects to normal and diseased airway epithelia

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Table S1

Mass loadings of chemical fractions in the smog chamber during cell exposures

Experimental conditions	Particle type	Averaged mass loading [μg m^{-3}]							
		OM	eBC	PAHs	NO_3	SO_4	NH_4	Cl	Total mass
Day 1, AL	primary	217.2	34.2	8.7	8.8	1.9	1.8	0.1	272.7
	aged	137.7	13.4	2.0	7.1	1.3	2.6	0.0	164.1
Day 2, AL	primary	54.8	46.6	1.9	5.2	1.0	0.9	0.1	110.5
	aged	72.7	19.2	0.9	4.9	1.0	2.0	0.0	100.7
Day 3, HL	primary	35.7	42.2	6.3	4.8	1.1	0.6	0.0	90.7
	aged	115.3	15.3	2.6	10.0	2.9	3.9	0.0	150.0
Day 4, HL	primary	13.4	58.2	1.9	4.7	0.6	0.6	0.0	79.4
	aged	15.4	15.4	0.3	1.4	1.1	0.8	0.0	34.4

Total mass loading represents the averaged sum of all chemical fractions. AL: average wood load; HL: high wood load; OM: organic matter; eBC: equivalent black carbon; PAHs: polycyclic aromatic hydrocarbons; NO_3: nitrate; SO_4: sulfate; NH_4: ammonium; Cl: chloride.

Table S2

Particle dose in the human tracheobronchial (TB) tract at different ambient PM concentrations.

		Particle count median diameter	
		Primary, 180 nm [a]	**Aged, 250 nm [a]**
Particle density [g cm^{-1}]		1.00	1.00
Tidal volume, V_T [m^3]		0.00065	0.00065
Breathing frequency, *f* [min^{-1}] [b, c]		12	12
Inhaled air volume/h, adult [m^3] [b, c]		0.45	0.45
Inhaled air volume/24h, adult [m^3]		10.8	10.8
Surface area TB tract [cm^2] [d]		2471	2471
Deposition efficiency [b, c]		0.078	0.066
Ambient mass concentration [μg m^{-3}]	20		
Mass/surface area TB/24h [ng cm^{-2}]		6.8	5.8
Ambient mass concentration [μg m^{-3}]	100		
Mass/surface area TB/24h [ng cm^{-2}]		34	29
Ambient mss concentration [μg m^{-3}]	400		
Mass/surface area TB/24h [ng cm^{-2}]		137	115
Ambient mass concentration [μg m^{-3}]	1000		
Mass/surface area TB/24h [ng cm^{-2}]		341	288

a: Monodisperse aerosol with geometric standard deviation of 1.00 assumed; b: [1]; c: [2]; d: [3]

Table S3

Numeric values of biological responses of cell cultures at 24 h after aerosol exposure.

Exposure	Cell model											
	Normal			Asthmatic			CF			BEAS-2B		
Day/aerosol	Cytotoxicity (%)	IL-6 (pg/mL)	IL-8 (pg/mL)	Cytotoxicity (%)	IL-6 (pg/mL)	IL-8 (pg/mL)	Cytotoxicity (%)	IL-6 (pg/mL)	IL-8 (pg/mL)	Cytotoxicity (%)	IL-6 (pg/mL)	IL-8 (pg/mL)
D1/AL-prim	8.0	214	12539	---	258	23747	25.2	567	9858	19.2	1015	1894
	10.2	91.3	7691	---	477	23322	20.6	176	15169	15.1	1467	1055
	12.1	182	9437	---	483	16493	25.6	499	10912	21.5	1561	1104
D1/AL-aged	9.9	229	11825	---	116	21895	25.0	228	11834	16.9	1993	1891
	8.6	122	8859	9.5	58.2	28280	13.1	94.7	14039	15.4	1483	1170
	15.0	170	13833	2.4	103	13529	12.2	128	17954	20.1	1769	1806
D2/AL-prim	14.3	86.8	8794	---	---	---	20.1	230	10423	15.7	1752	1738
	19.3	161	7601	---	---	---	16.5	185	9391	16.5	1552	2732
	---	---	---	---	---	---	---	---	---	---	---	---
D2/AL-aged	12.4	225	7704	3.5	73.8	19674	10.7	101	9297	---	---	---
	11.5	234	8519	7.1	129	29311	10.9	68.9	5729	---	---	---
	---	---	---	---	---	---	---	---	---	---	---	---
D3/p-free	7.4	89.1	8279	4.6	58.6	14725	10.1	293	8060	7.9	127	166
	6.7	111	9662	---	167	13258	6.1	160	9067	21.4	663	946
	6.7	169	9879	---	---	---	9.0	242	7222	17.5	700	984
D3/HL-prim	10.7	102	13772	1.5	0.02	0.02	20.3	273	12579	34.0	2071	1757
	11.9	262	16200	5.6	274	10535	18.5	118	7240	30.6	1755	1491
	13.9	137	6884	---	---	---	25.8	258	10866	33.3	1679	1754
D3/HL-aged	15.4	170	11338	4.5	155	14121	26.7	221	8646	41.0	2215	949
	14.6	115	5069	5.8	168	14527	20.7	156	9662	31.1	1100	716
	14.1	158	5150	---	---	---	15.7	256	10204	29.8	1019	669
D4/HL-prim	16.9	117	9376	5.1	232	21455	24.4	92.4	7087	13.3	407	301
	16.0	128	5530	5.0	56.6	19817	25.3	84.3	5805	13.5	64.3	120
	---	---	---	---	---	---	---	---	---	---	---	---
D4/HL-aged	11.4	48.2	6240	3.4	31.8	16570	14.9	187	8426	22.8	2028	2564
	11.3	42.7	5533	2.6	21.3	11047	18.6	99.4	7236	24.9	2091	2143
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Cytotoxicity, measured as percentage of total lactate dehydrogenase (LDH) apically released. Interleukin (IL)-6 and IL-8 release, assessed in the basal culture medium. Fully differentiated cultures of human bronchial epithelia derived from cells of normal, asthmatic and cystic fibrosis [CF]) donors plus the BEAS-2B cell line were simultaneously exposed to the aerosol. Data are presented as individual values of distinct cell cultures. For graphical illustration see Fig. 4 in the main manuscript. D1-4: Day of experiment; AL: average wood load, HL: high wood load; prim: primary particles; aged: aged particles; p-free: particle-free air control.

Table S4

Evaluation of different regression models of toxicity of chemical particle fractions to bronchial epithelia.

Biomarker & cell model	Adjusted R^2 (explained variance) for different regression models							
	OM	eBC	PAHs	SO_4	ROS	OM, eBC[a]	OM, eBC PAHs, SO_4[a]	Full model[a]
Normal	0.151	0.011	0.12	0.000	0.06	0.141*	0.330	0.506
Asthmatic	0.011	0.016	0.000	0.000	0.000	0.000	0.000	0.000
CF	0.084	0.000	0.069	0.063	0.296	0.210	0.235	0.201
BEAS-2B	0.000	0.000	0.114	0.187	0.000	0.000***	0.604**	0.854
IL-6								
Normal	0.123	0.068	0.001	0.029	0.000	0.071	0.169	0.267
Asthmatic	0.000	0.101	0.123	0.163	0.238	0.373	0.431	0.531
CF	0.000	0.094	0.122	0.000	0.071	0.249	0.322	0.478
BEAS-2B	0.015	0.000	0.000	0.194	0.133	0.000**	0.165**	0.116
IL-8								
Normal	0.000	0.000	0.244	0.004	0.000	0.000	0.249	0.197
Asthmatic	0.124	0.042	0.128	0.000	0.000	0.061	0.429	0.466
CF	0.278	0.254	0.047	0.000	0.000	0.259	0.319	0.325
BEAS-2B	0.000	0.000	0.000	0.013	0.021	0.000***	0.000***	0.000

Values are adjusted R^2 for each cell model and biological end point for single and combinations of explaining variables. a: Residuals were tested for a significant influence of day of experiment. Depending on these tests, the entries are marked with * ($p \leq 0.05$), ** ($p \leq 0.01$), *** ($p \leq 0.001$). CF: cystic fibrosis; IL: interleukin; OM: organic matter; eBC: equivalent black carbon; PAHs: polycyclic aromatic hydrocarbons; SO_4: sulfate; ROS: reactive oxygen species.

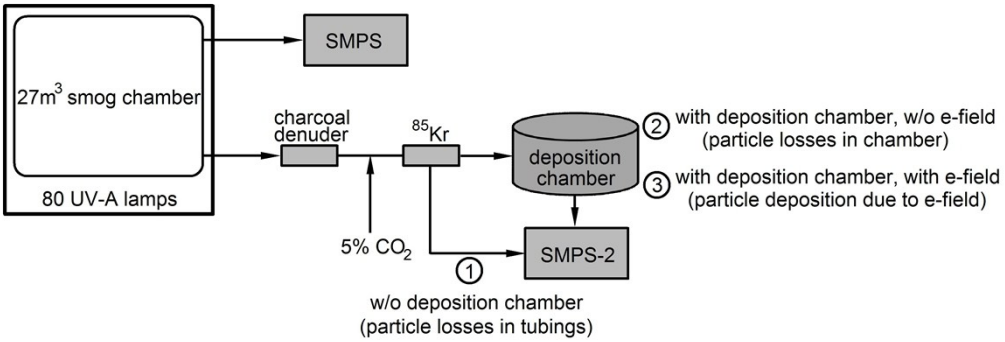

Fig. S1 Experimental set-up to estimate particle deposition on cell cultures. Particle mass concentration of wood combustion emissions in the smog chamber was continuously measured with a scanning mobility particle sizer (SMPS). An additional SMPS (SMPS-2) measuring at different positions was used to determine aerosol deposition in the deposition chamber and on cells. ① When SMPS-2 bypassed the deposition chamber, a comparison of the two SMPS reveals particle losses in aerosol delivery tubes. ② SMPS-2 measuring after the deposition chamber without electrical field yields particle losses in the deposition chamber. ③ The comparison of SMPS-2 measurements after the deposition chamber with and without electrical field results in particle deposition due to the electrical field.

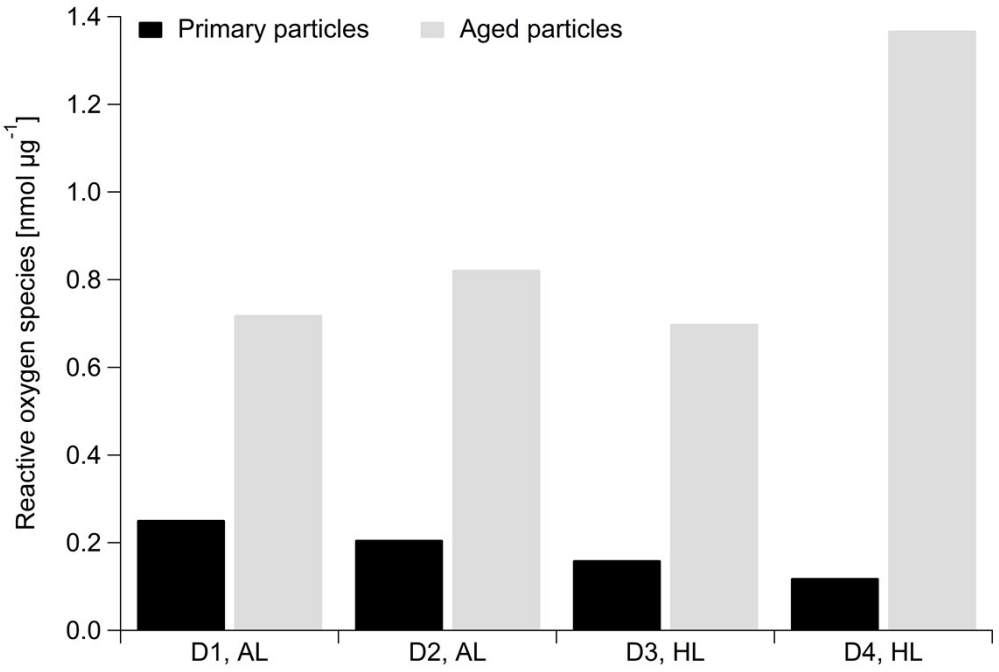

Fig. S2 Reactive oxygen species (ROS) normalized to total aerosol mass during each cell exposure. ROS content of primary and aged particles was determined from particles sampled on Teflon filters simultaneously to cell exposures. D: day; AL: average wood load; HL: high wood load.

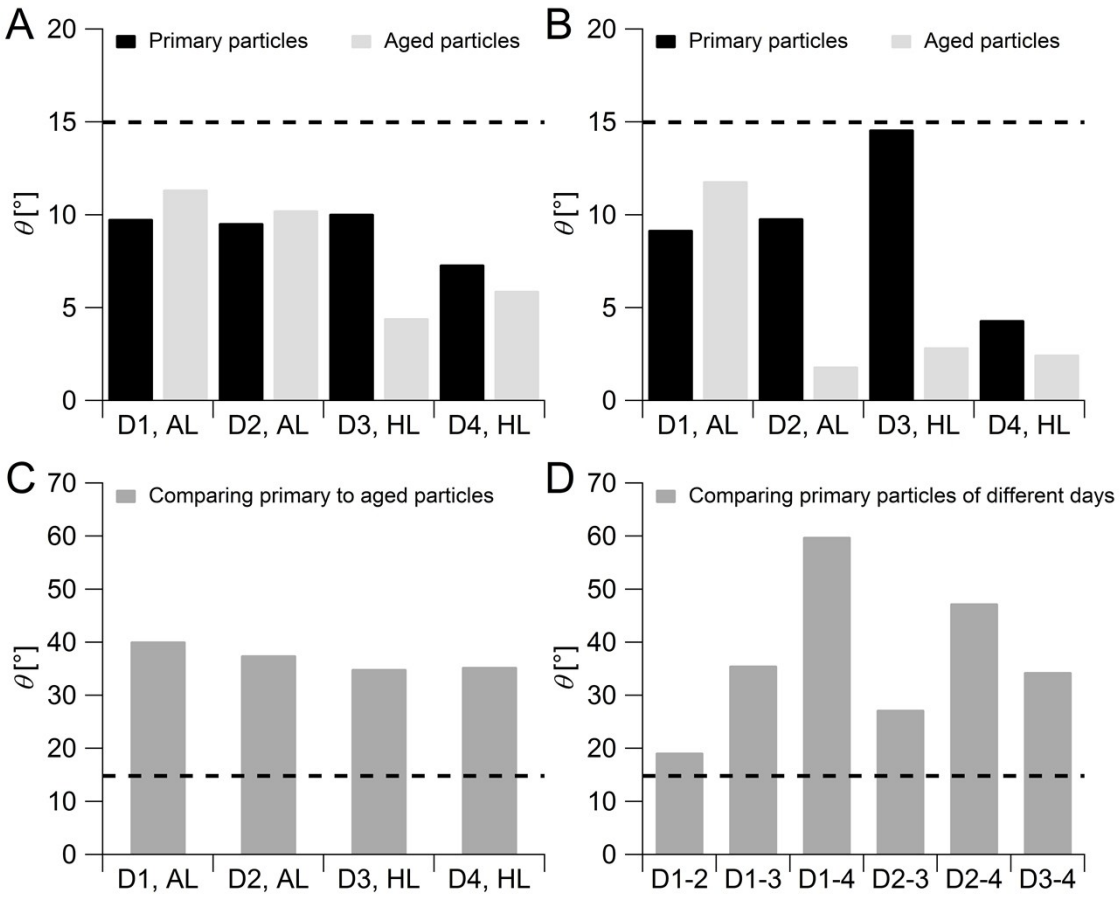

Fig. S3 Mass spectra of particles in the smog chamber and after the Versatile Aerosol Concentration Enrichment System (VACES). As a measure of agreement between two aerosol mass spectra we used θ, which is the angle between two corresponding vectors. The dashed vertical lines indicate θ = 15°. A) Effect of concentration enrichment on particle chemical composition. Mass spectra of particles measured after enrichment with the VACES were compared to those of particles from the smog chamber. B) Effect of chamber evolution on particle chemical composition. Mass spectra of particles from the smog chamber (without VACES) were compared to those of particles before and after each cell exposure. C) Effect of aging on particle chemical composition shown by comparing mass spectra of primary and aged particles. D) Comparison of mass spectra of primary particles from individual combustion processes demonstrates a considerable difference in particle chemical composition, e.g. D1-2 shows the angle Θ between primary emitted particles comparing days (D) 1 and 2, D1-3 compares days 1 and 3, etc.. AL: average wood load; HL: high wood load.

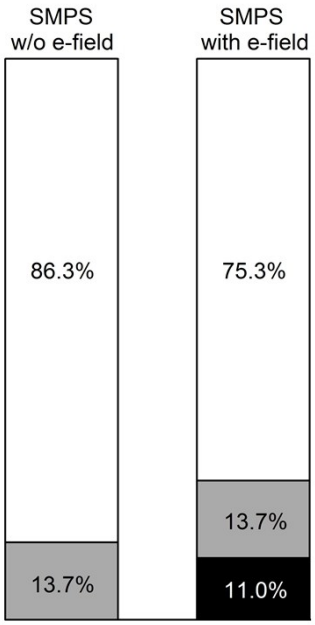

Fig. S4 Particle deposition efficiency in the aerosol deposition chamber without (w/o) and with electrical field. Scanning mobility particle sizer (SMPS) measurements are described in Figure S1. White: particles passing through the aerosol deposition chamber. Grey: particle losses in the chamber without e-field. 10% of particles lost in the chamber are assumed to be equally deposited on meshes of the delivery tubes and on cells [4]; Black: particle fraction deposited on meshes (losses) and cells by the electric field. Summing up the black (11.0%) and 10% of the grey areas (1.4%) and subsequent dividing by two (equal deposition on meshes and cells), results in a deposition efficiency of 6.2%.

References

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