

OUTDOOR AIR POLLUTION VOLUME 109

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SUPPLEMENTAL TABLES SECTION 4

Supplemental Table S1 Summary of studies that used plant assays to assess the ability of outdoor air to induce genetic mutations

Geographical location	Sites examined	Test system/exposure	End-point examined	Results obtained	Reference
São Paulo metropolitan area, Brazil	3 sites in the vicinity of a solid waste incinerator in São Paulo, and a rural reference site	<i>Tradescantia</i> clone 4430, in situ exposures in pots for 5 months	<i>Tradescantia</i> stamen-hair mutation assay	Significant increase in mutation frequency relative to reference site. Negative correlation between mutation frequency and distance from incinerator source	<u>Ferreira et al.</u> (2000)
São Paulo metropolitan area, Brazil	Ibirapuera Park in the southern area of the city of São Paulo	<i>Tradescantia</i> clones 4430 and KU-20, in situ exposures in pots, biweekly sample collection for 1 yr	<i>Tradescantia</i> stamen-hair mutation assay	No significant increase in mutation frequency in 4430. Mutation frequency in KU-20 significantly increased in summer months and associated with elevated NO ₂ and NO	<u>Ferreira et al.</u> (2007)
São Paulo metropolitan area, Brazil	Two urban sites in the city of São Paulo, and a rural reference site	<i>Tradescantia</i> 4430, in situ exposures in pots, monthly sample collection after initial 2-month exposure	<i>Tradescantia</i> stamen-hair mutation assay	Significant increase in mutation frequency at the high-traffic site only. Positive correlation between mutation frequency and suspended PM levels	<u>Ferreira et al.</u> (2003)
São Paulo metropolitan area, Brazil	High-traffic, urban site in São Paulo, and a rural reference site	Tradescantia clone KU-20, chamber exposures of inflorescences to outdoor air or filtered (0.8 μm porosity) outdoor air, daily sampling for 35 d	<i>Tradescantia</i> stamen-hair mutation assay	São Paulo outdoor air induced a significant increase in mutation frequency relative to reference site. Particle removal contributed to a reduction in mutation frequency, but frequency still elevated relative to reference site. Mutation frequency at São Paulo significantly correlated with PM ₁₀	<u>Guimarães et al.</u> (2004)

Geographical location	Sites examined	Test system/exposure	End-point examined	Results obtained	Reference
Variety of locations in the USA	19 locations that receive emissions from a wide range of urban/industrial sources, and 2 reference (control) locations	<i>Tradescantia</i> 4430, 10 d in situ exposures of cuttings in a climate-controlled mobile monitoring vehicle	<i>Tradescantia</i> stamen-hair mutation assay	Significant increases in mutation frequency at a range of urban and industrial sites. Highest mutation frequencies at site affected by facilities processing petroleum products	<u>Schairer et al.</u> (1982)
Toulouse metropolitan area, France	8 urban/ industrial locations, and a remote reference site	Nicotiana tabacum var. Xanthi, in situ exposures in pots for 1 month	Reverse mutation at 2 independent loci involved in chlorophyll parenchyma differentiation	Significant effect of treatment (i.e. site) on reversion rate. Highest reversion rate associated with industrial site. Lower reversion rate in 1997 significantly lower than in 1981, and decline corresponds with substantial decreases (> 2-fold) in atmospheric SO ₂ and NO ₂	<u>Vergé et al.</u> (2004)
Midwestern USA	15 sites in Ohio, Kentucky, Indiana, Illinois, and Colorado that cover a range of PM ₁₀ levels	Leaves and seeds of <i>Taraxacum officinale</i> Weber, <i>sensu lato</i> collected from field sites (in situ)	Mutation rates at minisatellite loci (parent-offspring transmission)	No relationship between minisatellite mutation rate and PM ₁₀ levels	<u>Rogstad et al.</u> (2003)
Sofia region, Bulgaria	A residential site (low PM_{10}) and an urban site (high PM_{10}) in Sofia	<i>Chlamydomonas</i> <i>reinhardtii</i> (WT 137C) exposed to samples diluted in DMSO (details not provided)	Visible size, morphology, and pigmentation mutants	Weak mutagenic effect (i.e. induced mutation frequency and mutagenic index) for summer samples from both urban sites. No effect for autumn samples	<u>Dimitrova et al.</u> (2007, 2009)
Moravia region, Czech Republic	3 samples from 2 industrial locations at Brno and Valašské Meziříčí. DCM extract of material collected on PUF plugs with high-volume sampler	Gametic mutations in <i>Arabidopsis thaliana</i> , seed exposure for 24 h	Frequency of siliques bearing mutant embryos	Significant increase relative to control for 2 of 3 samples. Highest mutation frequency associated with site near coal-tar conversion facility	<u>Chroust et al.</u> (1997)

d, day or days; DCM, dichloromethane; DMSO, dimethyl sulfoxide; h, hour or hours; NO, nitrogen oxide; NO₂, nitrogen dioxide; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; PUF, polyurethane foam; SO₂, sulfur dioxide; yr, year or years.

Supplemental Table S2 Summary of studies that used *Saccharomyces cerevisiae* in vitro assays to assess the ability of outdoor air to induce genetic mutations

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Po Valley, Italy (1990)	Airborne PM from residential/commercial locations in Parma, collected on GFFs with low- volume sampler. Tl Soxhlet extraction	Diploid D7 strain of <i>S. cerevisiae</i> , 2 h exposure to extract in DMSO, with and without endogenous metabolic activation (i.e. elevated P450 in log-phase cells)	Mitotic gene conversion of <i>trp5</i> locus, reversion of <i>ilv1–92</i> mutants, mitochondrial DNA mutations ("petite" colonies)	6 of 9 24 h samples collected over a 9-month period induced significant dose-related increases in <i>trp5</i> conversions without endogenous activation. 5 of 9 samples tested with activation elicited significant responses. Only one sample with endogenous activation induced a significant dose-related increase in <i>ilv1</i> reversion. Increase in petite colonies for spring and summer samples	<u>Poli et al.</u> (1992)
Po Valley, Italy (1990–1994)	Airborne PM from residential/commercial locations in Parma, collected on GFFs with low- volume sampler. Tl or Ac Soxhlet extraction	Diploid D7 strain of <i>S. cerevisiae</i> , 2 h exposure to extract in DMSO, with and without endogenous metabolic activation (i.e. elevated P450 in log-phase cells)	Mitotic gene conversion of <i>trp5</i> locus, reversion of <i>ilv1–92</i> mutants, mitochondrial DNA mutations ("petite" colonies)	48 monthly samples analysed. Several samples induced increases in point mutations and gene conversions both with and without endogenous activation. High variability across year and season. No evidence of temporal decline in outdoor air mutagenicity between 1990 and 1994. Several samples induced > 10-fold increase over control. Stronger response for Tl extracts. Several Tl extracts induced increases in mitochondrial DNA mutations	<u>Rossi et al.</u> (1995)
Po Valley, Italy (1991–1998)	Airborne PM from residential/commercial locations in Parma, collected on GFFs with low- volume sampler. Tl Soxhlet extraction	Diploid D7 strain of <i>S. cerevisiae</i> , 2 h exposure to extract in DMSO, with and without endogenous metabolic activation (i.e. elevated P450 in log-phase cells)	Mitotic gene conversion of <i>trp5</i> locus, reversion of <i>ilv1–92</i> mutants, mitochondrial DNA mutations ("petite" colonies)	96 pooled monthly samples analysed. With endogenous activation, several samples induced significant dose-related (equiv m ³ /mL) increases in point mutations and gene conversions. No evidence of temporal decline in outdoor air mutagenicity between 1991 and 1998	<u>Poli et al.</u> (1999)
Po Valley, Italy (1996, 1997)	Airborne PM from residential/commercial locations in Parma, collected on GFFs with low- volume sampler. Tl or Ac Soxhlet extraction	Diploid D7 strain of <i>S. cerevisiae</i> , 2 h exposure to extract in DMSO, with and without endogenous metabolic activation (i.e. elevated P450 in log-phase cells)	Mitotic gene conversion of <i>trp5</i> locus, reversion of <i>ilv1–92</i> mutants	Significant induction of gene conversion and point mutations, and potency values (rev/equiv m ³) revealed significantly greater activity associated with Tl extract. Size fractionation showed that $PM_{2.5}$ is frequently more mutagenic (per µg of PM) than PM_{10} or TSP	<u>Buschini et al.</u> (2001)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Pisa, Italy (1993–1994)	Airborne PM from 2 locations, collected on cellulose nitrate filters with low-volume sampler. Sequential extraction with DCM and MeOH	Diploid D7 strain of <i>S. cerevisiae</i> , 4 h exposure to extract in DMSO, with and without PB/5,6-BF- induced murine hepatic S9	Mitotic gene conversion of <i>trp5</i> locus, reversion of <i>ilv1–92</i> mutants	Significant induction of gene conversion and point mutations without S9 (events/m ³) for high-traffic site only	<u>Bronzetti et al.</u> (1997)
Warsaw, Poland (1997–1998)	Airborne PM from an urban location, 12 composite monthly samples, collected on GFFs. DCM Soxhlet extraction	Haploid <i>S. cerevisiae</i> XV185– ¹⁴ C, 24 h exposure to extract in DMSO	Reversion of <i>trp5–</i> 48, <i>arg4–17</i> , <i>lys1–1</i> , <i>ade2–1</i> , <i>his1–7</i> , and <i>hom3–10</i> mutants	No significant increase in reversion relative to control (i.e. ratio of mutation frequency of exposed to solvent control < 2.0)	<u>Jadczyk &</u> <u>Kucharczyk</u> (2000)

Ac, acetone; 5,6-BF, 5,6-benzoflavone; DCM, dichloromethane; DMSO, dimethyl sulfoxide; equiv, equivalent; GFFs, glass-fibre filters; h, hour or hours; MeOH, methanol; PB, phenobarbital; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; Tl, toluene; TSP, total suspended particles.

Supplemental Table S3 Seasonal trends in the geometric mean *Salmonella* TA98 mutagenic potency of extracts of outdoor air particulate matter

Season	Number of samples	Mutagenic potency (revenants/m ³)			
		Without S9			With S9
		Geometric mean*	Standard error of the mean	Geometric mean*	Standard error of the mean
Winter	1163	13.5ª	1.0	13.4ª	1.1
Autumn	288	17.3ª	1.1	13.3ª	1.1
Spring	210	9.9 ^b	1.1	9.4 ^b	1.1
Summer	282	5.4°	1.1	5.2°	1.1

* Mean values accompanied by a different letter are significantly different at P < 0.01.

Supplemental Table S4 Geometric mean *Salmonella* TA98 mutagenic potency of organic extracts of outdoor air particulate matter collected from urban, industrial, residential, and rural locations

Site type	Number of samples	Mutagenic potency (revenants/m ³)			
		Without S9			With S9
	-	Geometric mean*	Standard error of the mean	Geometric mean*	Standard error of the mean
Urban	615	17.3ª	1.1	12.3ª	1.0
Industrial	323	13.5ª	1.0	10.0ª	1.1
Residential	259	7.8 ^b	1.1	6.4 ^b	1.1
Rural	441	5.0°	1.3	6.0 ^b	1.1

* Mean values accompanied by a different letter are significantly different at P < 0.01.

Supplemental Table S5 Summary of studies that used bacterial mutagenicity assays (e.g. Ames assay) to assess the ability of outdoor air to induce genetic mutations – North America

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). DCM sonication extraction	TA98, TA100, standard plate incorporation and pre-incubation assays, with and without S9 from Wistar rat liver, Aroclor 1254 induction, or PB/5,6-BF induction	S9 source had no effect on TA100 mutagenicity (per µg of EOM). Stronger response with TA98, and TA98 mutagenicity increased with Aroclor-induced S9. Similar responses for plate incorporation and pre-incubation versions	<u>Tokiwa et al. (1992)</u>
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). Sequential Soxhlet extraction with DCM and MeOH, fractionation on silica, acid-base-neutral fractionation	TA98, TA100, standard plate incorporation assay, with and without rat liver S9	Higher potency (per µg of EOM or per µg of PM) without S9, and DCM extracts more potent compared with MeOH extracts. Highest activity (92% of direct acting) in polar (MeOH) eluate. Acid-base-neutral fractionation revealed strong activity in acid fraction	<u>Nishioka et al.</u> (1985)
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). DCM sonication extraction, separate, sequential extraction with Hx, Hx:DEE (9:1), Hx:DEE (1:1), MeOH	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Stronger response on TA98 (per µL of extract), and clear increase in potency without S9. Clear increase in potency with increasing solvent polarity. PAHs, nitro-PAHs, and heterocyclic compounds detected in DCM extract. Polar extract contained some known mutagens, but compounds that contribute to activity without S9 largely unknown	<u>Savard et al. (1992)</u>
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). Two sequential DCM extractions followed by Ac Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Addition of S9 increased activity of DCM extract (per mg of PM) on TA98 and TA100. Substantial fraction of total mutagenic activity associated with Ac extract; 11–13% with S9, 20–21% without S9	<u>Nielsen (1992)</u>
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). DCM sonication extraction	TA98, TA100, microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Highest potency (per μ g of EOM) on TA98 without S9. No significant effect of S9 for TA100. Microsuspension technique contributed to 3–9-fold increases in potency, relative to plate incorporation	<u>Agurell & Stensman</u> (1992)

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). DCM Soxhlet extraction	TA98, TA98NR, TA98/1,8-DNP ₆ , microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Highest activity (per μ g of extract or per μ g of PM) on TA98 without S9. Significant reduction in potency on TA98/1,8-DNP ₆ , relative to TA98, without S9. Modest reduction on TA98NR	<u>Bagley et al. (1992)</u>
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). Sequential DCM, Ac Soxhlet extraction	TA98, TA98NR, TA98/1,8-DNP ₆ , standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Potency (per μ g of EOM) on TA98 higher without S9, and Ac extracts more potent compared with DCM extracts. Significant reduction in potency without S9 on TA98NR and TA98/1,8-DNP ₆ , relative to TA98	<u>Gundel et al. (1993)</u>
Washington, DC, and Philadelphia, Pennsylvania, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1648 and SRM 1649). Ac Soxhlet extraction	Plate incorporation assay with Ames II base-pair substitution strains TA7001, TA7002, TA70003, TA7004, TA7005, TA7006, without S9 activation	Mutagenic potency (per mg of PM) enhanced on TA7005 (CG to AT transversions), TA7004 (CG to TA transitions), TA7002 (TA to AT transversions), and TA70006 (CG to GC transversions)	<u>Erdinger et al.</u> (2004)
Elizabeth, New Jersey, USA (winter 1983)	Airborne PM from 10 suburban locations, collected on GFFs with high-volume sampler. Sequential CX, DCM, Ac Soxhlet extraction, fractionation by polarity	TA98, TA98NR, TA98/1,8-DNP ₆ , standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Potency (per μ g of EOM) on TA98 higher without S9, and increase in potency on polar fraction. Significant reduction in potency without S9 on TA98NR and TA98/1,8-DNP ₆ , relative to TA98	<u>Gundel et al. (1993)</u>
New York City, USA (1977–1979)	Airborne PM collected on GFFs from cyclone sampler. CX, DCM, or Ac Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic activity of the various extracts all elevated without S9; moderately polar fraction showed highest activity. Polar fraction generally showed higher potency (per m ³) in winter; non-polar or moderately polar fractions showed higher potency in autumn and winter. Estimated that half of activity per m ³ in winter from fuel oil combustion for residential heating. Maximum total potency on TA98 without S9 ~12 rev/m ³	<u>Daisey et al. (1980)</u>
Chicago, Illinois, USA (1975)	Airborne PM collected with high-volume sampler. Soxhlet extraction with BZ:Hx:IProp (7:1:2), TLC fractionation	TA1538, standard plate incorporation assay, with Aroclor 1254-induced rat liver S9	Higher activity (per mg of PM) with S9; no obvious seasonal trend. Higher mutagenicity associated with wind direction	<u>Commoner et al.</u> (1978)

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Wayne County, Michigan, USA (1978)	Airborne PM collected on GFFs with high-volume sampler. BZ Soxhlet extraction	TA98, TA98NR, standard plate incorporation assay, without rat liver S9	Significant mutagenic activity (per µg of EOM), frequently elevated in residential areas downwind of urban/industrial areas. Potency (per µg of EOM) significantly reduced on NR- deficient strain. Some elevation in potency (per m ³) in the autumn. Maximum potency without S9 ~177 rev/m ³	Wang et al. (1980)
Detroit, Michigan, USA (1982)	Airborne PM from urban/ industrial area, collected on quartz filters with high-volume sampler. DCM Soxhlet extraction, fractionation by TLC	TA98, TA98NR, TA98/1,8-DNP ₆ , standard plate incorporation assay, without rat liver S9	Mutagenic activity (per μ g of EOM) predominantly in polar fractions. Significant positive association between mutagenicity and atmospheric SO ₂ , and variables that reflect vehicular emissions. Potency (per m ³) generally lower in summer. Maximum potency without S9 ~26 rev/m ³	<u>Wolff et al. (1986)</u>
West Virginia, USA (1984)	Outdoor PM collected on GFFs with high-volume sampler. Ac extraction	TA98 and TA98NR, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Similar potency (per mg of PM) with and without S9	<u>Krishna et al. (1986)</u>
Southern California, USA (1993)	Airborne PM from central Los Angeles, Azusa, Rubidoux, Long Beach, and control site (San Nicolas Island). Collected on quartz-fibre filters with virtual impactor. DCM Soxhlet extraction	<i>Salmonella</i> TM677 forward mutation assay (<i>Xprt</i>), with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) significantly decreased with S9. Source apportionment revealed that diesel PM and natural gas combustion made the largest contribution to potency without S9; natural gas combustion made the largest contribution to potency with S9	<u>Hannigan et al.</u> (2005)
Southern California, USA (1982)	Airborne PM from Azusa and Long Beach, collected on quartz-fibre filters from cyclone separator. DCM Soxhlet extraction	<i>Salmonella</i> TM677 forward mutation assay (<i>Xprt</i>), with and without rat liver S9	Higher mutagenicity without S9; higher potency (per μ g of EOM) for Long Beach. Mutagenic activity much higher than expected based on examined sources	<u>Hannigan et al.</u> (1994)
Southern California, USA (1993)	Airborne PM from 4 urban/industrial locations and 1 reference location, collected with virtual impactor or cyclone separator. DCM Soxhlet extraction	<i>Salmonella</i> TM677 forward mutation assay (<i>Xprt</i>), with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) of fine PM from urban sites showed pronounced seasonality; high in winter and low in summer; generally higher without S9. No seasonal differences in potency (per μ g of EOM). Urban sites also more potent (per μ g of EOM); 10-fold above reference location. Location downwind of urban area more potent with S9	<u>Hannigan et al.</u> (1996)

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Southern California, USA (1976)	Airborne PM from 8 urban locations and 1 rural location, collected on GFFs with high-volume sampler. DCM:MeOH:BZ (1:1:1) sonication extraction	TA98, TA100, TA1535, TA1537, TA1538, standard plate incorporation assay, with and without rat liver S9	Highest mutagenicity on TA98 without S9. Mutagenic potency (per µg of EOM) similar across urban sites, with highest activity at Los Angeles. No response for rural site	<u>Pitts et al. (1977)</u>
Claremont, California, USA (1987)	Airborne PM collected on Teflon-impregnated GFFs with high-volume sampler. DCM Soxhlet extraction, detailed fractionation	TA98, microsuspension assay, without S9 activation	High mutagenic activity in fractions containing nitro-PAHs. PAH reaction products such as nitro- PAH lactones suspected as major contributors to mutagenicity. Maximum potency without S9 ~160 rev/m ³	<u>Arey et al. (1992)</u>
Claremont and Torrance, California, USA (1986)	Airborne PM collected on Teflon-impregnated GFFs with high-volume sampler. DCM Soxhlet extraction	TA98, standard plate incorporation assay, without S9 activation	Mutagenic potency (per m ³) frequently elevated in winter compared with autumn. Maximum potency ~120 rev/m ³ . Results indicate that atmospheric formation of nitroarenes is an important determinant of PM mutagenicity	<u>Arey et al. (1988)</u>
El Monte, California, USA (1980)	Airborne PM (< 20 μm) collected on GFFs, Teflon, quartz, or Teflon- impregnated filters. Tl:DCM:MeOH sonication extraction	TA98, standard plate incorporation assay, with and without rat liver S9	Mutagenic potency (per µg of EOM or m ³) similar with and without S9; no difference for different filter media. Maximum potency without S9 ~35 rev/m ³ ; with S9 ~33 rev/m ³	<u>Fitz et al. (1984)</u>
Redlands, California, USA (1984)	SVOCs collected on PUF plugs. DCM Soxhlet extraction, HPLC fractionation	TA98, microsuspension assay, without rat liver S9	Higher mutagenic activity (per m ³) at night. Nitro- and methylnitro-naphthalenes accounted for 18–32% of SVOCs mutagenic activity. Maximum potency without S9 ~31 rev/m ³	<u>Gupta et al. (1996)</u>
West Virginia, USA (1982)	Airborne PM collected on GFFs with high-volume sampler. Shaker extraction with Ac, BZ, CX, MeOH, DCM, or BZ:CX:MeOH (1:1:1)	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Evaluation of various extraction solvents; highest mutagenic activity associated with DCM extract. Highest activity on TA100 with S9. Additional analyses showed that sonication is effective for extraction and possibly superior to Soxhlet	<u>Krishna et al. (1983)</u>
West Virginia, USA (1982)	Airborne PM collected on GFFs with high-volume sampler. Ac extraction by sonication, shaker, or Soxblet	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Sonication is effective for extraction, and some indication that it is superior compared with Soxhlet and shaker	<u>Krishna et al. (1983)</u>

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
West Virginia, USA (1982)	Airborne PM collected on GFFs with high-volume sampler. Ac extraction	TA98, standard plate incorporation assay, without metabolic activation	Significant positive response on both TA100 and TA98. Potency (per mg of PM) higher on TA98	<u>Krishna et al. (1984)</u>
West Virginia, USA (1980)	Airborne PM from 5 sites near Morgantown, collected with cascade impactor. Sequential shaker extraction with DCM and EA:MeOH (1:1), acid–base–neutral fractionation	TA98, TA100, SV50 (Ara test), standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) higher without S9 in summer, with S9 in autumn and winter. High activity in acidic and PAH fractions. Drop in potency (per m ³) after rain or snow. Maximum potency on TA98 with S9 ~46 rev/m ³ ; without S9 ~85 rev/m ³	<u>Whong et al. (1981)</u>
West Virginia, USA (1982)	Airborne PM from Morgantown, collected on GFFs with high-volume sampler. DCM shaker extraction	TA98, standard plate incorporation assay, without metabolic activation	52% of mutagenic activity associated with PM $< 2~\mu m.$ Mutagenic potency of PM $< 1.1~\mu m$ ${\sim}25~rev/m^3$	<u>Sorenson et al.</u> (1982)
Lexington, Kentucky, USA (1980)	Airborne PM before and during forest fires, collected on GFFs with high-volume sampler. BZ and Ac sonication extraction	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) on TA98 elevated with S9 for "smoky" conditions; higher without S9 for "non-smoky" conditions. Potency (per µg of EOM) similar for different sample types; "smoky" far higher per mg of PM or per m ³ . Potency on TA98 for "smoky" conditions with S9 ~45 rev/m ³ ; without S9 ~27 rev/m ³ . For "non- smoky" conditions, potency on TA98 with S9 ~1 rev/m ³ ; without S9 ~2 rev/m ³	<u>Viau et al. (1982)</u>
Houston, Texas, USA (1977–1978)	Airborne PM collected on GFFs with high-volume sampler. Combined Ac, CX, MeOH, Chl extraction	TA1535, TA1537, TA98, TA100, plate incorporation assay, with and without rat liver S9	Mutagenic activity highest on TA98 with S9. Positive correlation between mutagenic activity (per m ³) and lung cancer incidence. Maximum mutagenic potency on TA98 ~6 rev/m ³	<u>Walker et al. (1982)</u>
Houston, Texas, USA	TSP collected on GFFs with high-volume sampler. Soxhlet extraction with CX or BZ:MeOH:DCM (1:1:1)	TA1535, TA1537, TA1538, TA98, TA100, spot test, TA98 and TA100 plate incorporation assay, with and without rat liver S9	Elevated mutagenic activity (per mg of TSP) without S9; solvent mixture more effective for extracting mutagens	<u>Preidecker (1980a)</u>

Test article Geographical Salmonella strains^a, assay version Results Reference location Houston, Texas, Airborne TSP or size-TA98, TA100, TA1538, TA1978 Mutagenic potency (per mg of TSP) highest Preidecker (1980b) (TA1538 *uvrB*+), standard plate at sites downtown or south/south-west of USA fractionated PM collected incorporation assay, with and without downtown. Potency highest without S9 for both with high-volume sampler Aroclor 1254-induced rat liver S9 or cascade impactor. TA98 and TA100. Cascade impactor samples PM collected on GFFs. showed dramatically increased potency (per BZ:MeOH:DCM (1:1:1) m³) for finest material (< 1 μ m). No significant Soxhlet extraction response on TA1978, relative to strong response on TA1538. Maximum mutagenic potency on TA98 without S9 ~3.5 rev/m3 Mutagenic potency (per m³ or per µg of EOM) Contra Costa Airborne PM from 4 TA98, standard plate incorporation Flessel et al. (1985) higher in summer, and highest potency (per m³) County, urban/residential sites, assay, with and without Aroclor California, USA collected on GFFs with 1254-induced liver S9 at sites near heavy traffic and oil-burning electric (1981 - 1982)high-volume sampler. BZ power facility. All samples more potent (per m³) with S9. Significant correlations between potency extraction (per m³) and lead in fine PM ($< 2.5 \mu m$) as well as atmospheric NO₂. Maximum mutagenic potency on TA98 without S9 ~12 rev/m3; with S9 ~30 rev/m3 Contra Costa Airborne PM from 15 All samples mutagenic in at least one strain; Flessel et al. (1980) TA1537, TA1537, TA1538, TA98, County, urban/residential sites. TA100, standard plate incorporation highest activity on TA98. Mutagenic potency assay, with and without rat liver S9 (per m³) higher with S9 and generally higher in California, USA collected on GFFs with (1978 - 1979)high-volume sampler. BZ winter. Elevated potency at more-urban settings with higher PAH levels. Maximum mutagenic extraction potency on TA98 without S9 ~6 rev/m3; with S9 ~25 rev/m³ Diurnal analyses showed rapid changes (over TA98, TA98NR, standard plate Pitts et al. (1985) Los Angeles, PM₁₀ from 2 sites near San Diego Freeway, collected several hours) in mutagenic potency (per m³). California, USA incorporation assay, with and without Aroclor 1254-induced rat liver S9 (1983)on Teflon-coated filters Similar potencies with and without S9, and with high-volume sampler. clear reduction on TA98NR. Potency of samples DCM Soxhlet extraction downwind of freeway showed elevated potency and correspondence with atmospheric lead, CO, and NO₂. Maximum potency on TA98 with S9 ~140 rev/m3; without S9 ~140 rev/m3

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
California, USA (1982)	Coarse (< 15 µm) and fine (< 2.5 µm) PM from 3 locations, collected on GFFs with high-volume sampler. MeOH:DCM:TI (1:1:1) sonication extraction	TA98, TA98NR, standard plate incorporation and microsuspension assays, with and without rat liver S9	10-fold increase in mutagenic potency on microsuspension assay, relative to plate incorporation. Mutagenic potency without S9 (per m ³) lower on TA98NR relative to TA98. Elevated potency without S9 and activity associated with PM _{2.5} only. Maximum potency on TA98 (microsuspension) with S9 ~1100 rev/m ³ ; without S9 ~450 rev/m ³ . Potency correlated with lead levels	<u>Kado et al. (1986)</u>
Buffalo, New York (1962) and Berkeley, California, USA	Airborne PM, collected on GFFs. Ac Soxhlet extraction	TA1535, TA1537, TA98, and TA100, standard plate incorporation assay, with and without rat liver S9 (induced and uninduced)	Archived Buffalo PM samples showed enhanced potency (per μ g of EOM) with S9 and maximum response with Aroclor-induced S9. Berkeley sample potency (per μ g of EOM) higher without S9. Berkeley samples more potent than Buffalo samples without S9, and less potent than or equivalent to Buffalo samples with S9	<u>Talcott & Wei</u> (<u>1977)</u>
Boise, Idaho, USA (1986–1987)	PM _{2.5} from 7 sites during winter, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, TA100, plate incorporation and microsuspension assays, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher without S9 for microsuspension only. Highest mean potency > 100 rev/m ³ . Microsuspension potency ~3-fold higher than plate incorporation potency. Potency at primary sites significantly higher than at reference (background) site. Background activity accounted for 7–19% of activity at primary sites. Maximum potency on TA98 (microsuspension) with S9 ~147 rev/m ³ ; without S9 ~312 rev/m ³	<u>Claxton et al. (2001)</u>
Boise, Idaho, USA (1986–1987)	Composite PM _{2.5} sample, collected on GFFs with high-volume sampler. DCM sonication extraction, fractionation on anion exchange resin	TA98, TA1538, TA100, plate incorporation and microsuspension assays, with and without Aroclor 1254-induced rat liver S9	Composite sample prepared to maximize contributions from mobile-source emissions. Highest activity (TA98 with S9 per μ g of EOM) in base-neutral fraction. Majority of TA98 reversion mutations similar to that observed for B[<i>a</i>]P. GC to TA transversion mutations in TA100 predominated for both PM extract and B[<i>a</i>]P	<u>DeMarini et al.</u> (1994)
Newark, New Jersey, USA (1988)	Airborne PM from residential/industrial location, collected on GFFs with high-volume sampler. Sequential Soxhlet extraction with DCM and Ac detailed fractionation	TA98, TA98NR, TA98/1,8-DNP ₆ , TA100, plate incorporation and microsuspension assays, with and without Aroclor 1254-induced rat liver S9	Analyses of fractions from seasonal composites indicated similar profiles of mutagenic activity across seasons; shift to molar polar compounds in summer. Polar neutral aromatics important contributor to activity of all samples; highly polar acidic fractions showed strong activity on TA100	<u>Greenberg et al.</u> (1993)

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Rutland, Vermont, USA (1987–1988)	TSP and PM ₁₀ from a site near a municipal incinerator, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Similar mutagenic potency (per m ³) with and without S9. Maximum potency on TA98 without S9 ~35 rev/m ³ ; with S9 ~25 rev/m ³ . No detection of mutagens that could be linked to incinerator activities	<u>Watts et al. (1989)</u>
Allegheny County, Pennsylvania, USA	Airborne PM from 4 locations, collected with cascade impactor. Ac Soxhlet extraction	TA98, TA100, TA1538, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per μ g of EOM) higher with S9 and elevated on TA98 compared with other strains. Highest mutagenicity associated with finest size fractions (0.5–1.5 μ m)	<u>Sideropoulos & </u> Specht (1994)
Ann Arbor, Michigan, USA (1991)	Airborne PM, collected on GFFs with cascade impactor. Sequential Soxhlet extraction with DCM	TA98, TA100, TA102, plate incorporation and plate incorporation assay, without S9 activation	Strong responses on TA98 and TA100; lower for TA102. Maximum potency on TA98 without S9 ~4 rev/m ³ ; on TA100 without S9 ~9 rev/m ³	<u>Hoyer et al. (1992)</u>
New Mexico and North Carolina, USA (1984–1985)	PM _{2.5} samples from Albuquerque (New Mexico) and Raleigh (North Carolina), collected with cascade impactor. DCM sonication extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency on TA98 with ~20–30 rev/ m ³ . Contributions from wood smoke emissions to activity per unit of EOM greater than mobile- source emissions. Mobile sources made a much larger contribution to activity per m ³	<u>Stevens et al. (1990)</u>
Durham, North Carolina, USA (1978–1979)	Airborne PM, collected with cascade impactor. Ac Soxhlet extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) almost always higher without S9; highest activity for PM < 2 μ m. Maximum potency without S9 ~10 rev/m ³ ; with S9 ~6 rev/m ³	<u>Talcott & Harger</u> (1980, 1981)
Durham, North Carolina, USA (1989–1990)	PM ₁₀ , collected on several types of media with high- volume sampler. DCM sonication extraction	TA98, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) of most samples elevated with S9; Teflon-impregnated GFFs yielded highest activity. Maximum potency with S9 ~50 rev/m ³	<u>Watts et al. (1992)</u>
Camden, New Jersey, USA (1983)	Airborne PM collected on GFFs with cascade impactor. Sequential Soxhlet extraction with CX, DCM, and Ac	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Potency (per μ g of EOM) of Ac extracts higher without S9. Potency of CX extracts slightly higher with S9. Ac extracts far more potent on TA98 than on TA100	<u>Miguel et al. (1990)</u>

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Hamilton, Canada (1990–1991)	PM ₁₀ from 2 monitoring stations, collected on GFFs with high-volume sampler. Sequential Soxhlet extraction with DCM and MeOH, detailed fractionation	YG1021, standard plate incorporation assay, without S9 activation	Mutagenic potency (per m ³) of non-polar PAC fractions without S9 ranged from 1 rev/m ³ to 134 rev/m ³ . Highest mutagenic activity associated with winds from east (e.g. urban Toronto), low wind velocities, high SO ₂ , and high NO ₂	<u>Morris et al. (1995)</u>
Hamilton and Burlington, Canada (1990–1991)	Airborne PM collected on Teflon-coated GFFs with high-volume sampler. Sequential DCM, Ac extraction, extensive fractionation	YG1021, YG1024, YG1026, YG1029, TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) of non-polar fraction without S9 dramatically increased on both YG1021 and YG1024 compared with TA98. Potency of non-polar fraction increased with S9 on all strains except YG1024. Relatively small fraction of activity contained in polar fraction (i.e. 12–33%). Maximum potency on TA98 without S9 ~4 rev/m ³ ; with S9 ~12 rev/m ³ . Several potent PAHs and nitro-PAHs identified in non- polar fraction	<u>Legzdins et al.</u> (1995)
Hamilton, Canada (1975–1976)	Seasonal composites of airborne PM. BZ Soxhlet extraction, fractionation by TLC	TA1537, TA1538, TA1535, TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per μ g of EOM) greater on TA98, compared with TA100; similar results with and without S9	<u>Salamone et al.</u> (1979)
Mexico City, Mexico (1991)	Airborne TSP and PM ₁₀ , collected on GFFs with high-volume sampler. MeOH Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Increased mutagenic activity with S9, and $\rm PM_{10}$ extracts more mutagenic relative to TSP	<u>Delgado-Rodríguez</u> et al. (1999)
Mexico City, Mexico (1998)	PM ₁₀ from an urban site, collected on GFFs with high-volume sampler. DCM sonication extraction, fractionation on silica	TA98, YG1021, plate incorporation assay, with and without PB/5,6-BF- induced rat liver S9	Highest mutagenic activity in moderately polar fraction (PAHs and nitro-PAHs) and polar fraction. Positive correlation between activity without S9 and nitro-PAH concentrations. Positive correlation between activity with S9 and PAH concentrations. Maximum potency (per µg of EOM) on YG1021 associated with moderately polar fraction	<u>Villalobos-Pietrini</u> <u>et al. (2007)</u>
Mexico City, Mexico (1992)	PM ₁₀ from an urban site, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) without S9 similar, or elevated, relative to with S9. Potency without S9 correlated with PM levels. Potency with S9 correlated with atmospheric NO _x and CO. Maximum potency on TA98 without S9 ~18 rev/m ³ ; with S9 ~20 rev/m ³	<u>Villalobos-Pietrini</u> et al. (1999)

Geographical location	Test article	Salmonella strains ^a , assay version	Results	Reference
Mexico City, Mexico (1989– 1990)	PM ₁₀ and TSP from 5 locations, collected on GFFs with high-volume sampler. MeOH Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m^3) generally higher for PM_{10} compared with TSP; response significantly increased with S9	<u>Villalobos-Pietrini</u> <u>et al. (1995)</u>
Mexico City, Mexico (1998)	PM ₁₀ from south-western Mexico City, collected on GFFs with high-volume sampler. Sonication extraction with DCM	TA98, YG1021, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) higher with S9 in summer; opposite for colder months. Highest potency during a dry period with many fires. Significant increases on YG1021 relative to TA98 only for colder months with ground-based inversions. Some positive associations between mutagenic activity and atmospheric levels of CO, NO ₂ , and SO ₂ ; negative association with rainfall. Maximum potency on TA98 without S9 ~25 rev/m ³ ; with S9 ~52 rev/m ³	<u>Villalobos-Pietrini</u> <u>et al. (2006)</u>
Mexico City, Mexico (1990)	Airborne PM from 2 urban locations, collected on GFFs with high-volume sampler. DCM extraction of shredded filter	TA1538, TA98, YG1024, YG1021, TA98NR, plate incorporation assay, without rat liver S9	Mutagenic potency (per m ³) increased on both YG1021 and YG1024, and reduced on TA98NR, relative to TA98. Maximum potency on TA98 without S9 ~21 rev/m ³	<u>Espinosa-Aguirre</u> <u>et al. (1993)</u>
Mexico City, Mexico (1993)	PM ₁₀ and TSP from a residential area, collected on GFFs. Sequential Soxhlet extraction with CX, DCM, and Ac	TA98, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) highest for Ac extract and frequently higher with S9. TSP extracts frequently more potent than PM_{10} extracts. Maximum potency on TA98 without S9 ~4.7 rev/m ³ ; with S9 ~4.2 rev/m ³	<u>Amador-Muñoz</u> <u>et al. (2001)</u>

^a YG1021: TA98 with plasmid pYG216, NR-overproducing strain; YG1024: TA98 with plasmid pYG219, OAT-overproducing strain; YG1041: TA98 with plasmid pYG233, NR- and OAToverproducing strain; YG1026: TA100 with plasmid pYG216, NR-overproducing strain; YG1029: TA100 with plasmid pYG219, OAT-overproducing strain; YG1042: TA100 with plasmid pYG233, NR- and OAT-overproducing strain.

Ac, acetone; 5,6-BF, 5,6-benzoflavone; B[a]P, benzo[a]pyrene; BZ, benzene; Chl, chlorophyll; CO, carbon monoxide; CX, cyclohexane; DCM, dichloromethane; DEE, diethyl ether; EA, ethyl acetate; EOM, extractable organic matter; GFFs, glass-fibre filters; HPLC, high-performance liquid chromatography; Hx, hexane; MeOH, methanol; NO₂, nitrogen dioxide; NO₃, nitrogen oxides; NR, nitroreductase; OAT, *O*-acetyltransferase; PACs, polycyclic aromatic compounds; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PM, particulate matter; PM₁₀, particulate matter with particles of aerodynamic diameter < 10 μ m; PM_{2.5}, particulate matter with particles of aerodynamic diameter < 2.5 μ m; PUF, polyurethane foam; rev, revertants; SO₂, sulfur dioxide; SRM, standard reference mixture; SVOCs, semivolatile organic compounds; Tl, toluene; TLC, thin-layer chromatography; TSP, total suspended particles.

Supplemental Table S6 Summary of studies that used bacterial mutagenicity assays (e.g. Ames assay) to assess the ability of outdoor air to induce genetic mutations – South America

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Rio de Janeiro, Brazil (1984)	Airborne PM collected on GFFs with cascade impactor. Sequential Soxhlet extraction with CX, DCM, and Ac	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	For Rio de Janeiro, potency higher (per m^3 or per μ g of EOM) without S9 on TA98; higher with S9 on TA100. Some indication of increased potency during the day. DCM extract potency generally exceeds Ac and CX extract potency. CX potency (per μ g of EOM) similar across sites. Maximum potency on TA98 without S9 ~11.8 rev/m ³ ; with S9 ~8.5 rev/m ³	<u>Miguel et al. (1990)</u>
Rio Grande do Sul state, Brazil (2004–2005)	Airborne PM from 2 sites, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, TA100, YG1021, YG1024, microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Elevated mutagenic potency (per m ³) at urban/ industrial site, relative to non-industrial reference. PAH concentrations at urban/industrial site ~25- fold higher than at non-industrial site. Potency increased with S9. Potency for urban/industrial sample (per μ g of PM) elevated on both NR- and OAT-enhanced strains. Maximum potency on TA98 without S9 ~19 rev/m ³ ; with S9 ~32 rev/m ³	<u>Pereira et al. (2013)</u>
Rio Grande do Sul state, Brazil (2004–2005)	TSP from an urban/ industrial site and a rural site, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, TA100, YG1021, YG1024, microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per μ g of EOM) increased on YG1021 and YG1024, relative to TA98; generally highest on YG1024. Potency (per m ³) substantially higher for urban/industrial site. Maximum potency on TA98 without S9 ~3 rev/m ³ ; with S9 ~4 rev/m ³	<u>Pereira et al. (2010)</u>
Rio Grande do Sul state, Brazil (2009–2010)	TSP and PM _{2.5} from 2 urban/industrial sites, collected on GFFs and Teflon membrane filters with high-volume sampler. Sequential sonication extraction with DCM and water	TA98, YG1021, YG1024, microsuspension assay, with and without rat liver S9	Potency (per µg of PM) of all DCM extracts higher for $PM_{2.5}$, relative to TSP. Water extracts all negative for mutagenic activity. Enhanced activity on metabolically enhanced strains, particularly NR-enhanced strain YG1021. High temporal variability in effect of S9 on TA98 mutagenic potency (per m ³). Maximum potency on TA98 without S9 ~2.5 rev/m ³ ; with S9 ~2.3 rev/m ³	<u>Lemos et al. (2012)</u>

Supplemental Table S6 (continued) Test article Geographical Salmonella strains^a/assay version Results Reference location TA98, YG1021, YG1024, São Paulo, Brazil Airborne PM₁₀ from a Mutagenic potency (per m³) of crude extract De Martinis et al. residential/commercial site increased with S9. Moderately polar fractions of (1999, 2002)(1994)microsuspension assay, with and in São Paulo city, collected without Aroclor 1254-induced rat liver DCM extract increased with S9. Polar fractions on Teflon-coated GFFs higher without S9. Moderately polar and polar **S9** with high-volume sampler. fractions of Ac extract more mutagenic without Sequential sonication S9. Maximum potency on TA98 without S9 extraction with DCM ~150 rev/m3; with S9 ~165 rev/m3. The most and Ac, fractionation on mutagenic DCM extract fractions contained ketones, aldehydes, and guinolines. PAH bonded-phase column concentration ratios indicate mobile-source contributions São Paulo, Brazil Airborne PM from 2 sites TA98, pre-incubation assay, with and Mutagenic potency (per µg of EOM) dramatically Umbuzeiro et al. in São Paulo city, collected without Aroclor 1254-induced rat liver reduced with S9 (2010)on GFFs with high-volume S9 sampler. DCM sonication extraction São Paulo, Brazil Airborne PM from 2 sites Elevated potency (per µg of PM) on TA98 without Umbuzeiro et al. TA98, YG1041, microsuspension in São Paulo city, collected assay, with and without Aroclor S9 for nitro-PAH and oxy-aromatic fractions. (2004)(2008)1254-induced rat liver S9 on quartz filters with Dramatic increases on YG1041, relative to TA98. high-volume sampler. Elevated potency for higher-traffic site DCM Soxhlet extraction, fractionation on silica São Paulo state, Airborne PM from 3 TA98, TA100, TA98NR, plate Mutagenic potency (per m³) highest on TA98, Sato et al. (1995) urban/industrial locations, incorporation assay, with and without except for winter. Highest activity for heavy-Brazil (1990–1991) collected on GFFs or Aroclor 1254-induced rat liver S9 traffic site. Pooled seasonal samples showed lower quartz filters with highpotency in winter; generally higher on TA98 volume sampler. DCM without S9. Maximum potency on TA98 ~246 rev/ Soxhlet extraction m³ without S9; ~120 rev/m³ with S9. Reductions in potency on TA98NR and TA98/1,8-DNP, relative to TA98 São Paulo state, PM₁₀ from Araraquara, YG1024, pre-incubation assay, with Mutagenic potency (per m³) higher without de Andrade et al. Brazil collected on Teflonand without Aroclor 1254-induced rat S9 and elevated (5-10-fold) during sugar-cane (2011)(2002 - 2004)coated GFFs or quartz liver S9 harvest period (i.e. field burning) compared with filters with high-volume non-harvest period sampler. DCM:MeOH (4:1)

sonication extraction

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Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
São Paulo state, Brazil (2003)	PM ₁₀ from Araraquara, Piracicaba, and São Paulo city, collected on GFFs with high-volume sampler. DCM Soxhlet extraction	TA98, TA100, YG1041, YG1042, microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) on TA98 higher without S9; highest in areas affected by cane- burning activities. Dramatic increase in potency on YG1041 without S9, relative to TA98. Maximum potency on TA98 ~320 rev/m ³ without S9; ~57 rev/m ³ with S9 (cane-burning area); ~450 rev/m ³ without S9; ~140 rev/m ³ with S9 (São Paulo City). Strong contributions from nitro- PAHs and oxy-aromatics	Umbuzeiro et al. (2008)
Santiago, Chile (1996)	PM_{10} and TSP from an urban, heavy-traffic site, collected on Teflon-coated filters and GFFs with high- volume sampler. DCM sonication extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency varied during the year, reached up to 700 rev/m ³ during summer (without S9). Potency often elevated without S9. Maximum potency on TA98 with S9 ~620 rev/m ³ . Potency of TSP elevated relative to PM_{10} . PAHs elevated during summer months	<u>Adonis & Gil</u> (<u>2000)</u>
Santiago, Chile (1990–1991)	Airborne PM from an urban, heavy-traffic site, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, TA98NR, TA98/1,8-DNP $_{6}$, plate incorporation assay, with and without rat liver S9	Mutagenic potency higher with S9, and potency on TA98 reached maximum of 309 rev/m ³ . Maximum potency on TA98 without S9 ~106 rev/m ³ . Potency without S9 dramatically reduced on TA98NR and TA98/1,8-DNP ₆ compared with parent strain	<u>Adonis & Gil (1993)</u>
Santiago, Chile (1991)	Airborne PM from an urban, site, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Higher potency (per m ³) with S9. Maximum potency without S9 177 rev/m ³ ; with S9 166 rev/m ³	<u>Adonis et al. (1997)</u>
Santiago, Chile (1991)	Airborne PM from downtown area, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Higher potency (per m ³) with S9 in winter; opposite in summer. Corresponds with elevated levels of PAHs in cold season. Maximum potency with S9 ~300 rev/m ³ ; without S9 ~100 rev/m ³	<u>Gil & Adonis (1996)</u>
Santiago, Chile (1994)	Airborne PM from downtown area and rural control, collected on Teflon filters with low-flow sampler. DCM sonication extraction	TA98, microsuspension assay, with and without rat liver S9	Mutagenic potency (per m ³) at urban site higher without S9; at rural site higher with S9. Maximum potency with S9 ~2100 rev/m ³ ; without S9 ~2800 rev/m ³	<u>Gil et al. (1997)</u>

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Santiago, Chile (1996, 1997)	PM ₁₀ from several urban, residential, and industrial sites, collected on quartz filters with high-volume sampler. DCM sonication extraction	TA98, TA100, pre-incubation assay, with and without rat liver S9	Mutagenic potency (per m ³) frequently higher without S9; highest values for Santiago on TA100 without S9 (~140 rev/m ³). Potency on TA98 without S9 ~67 rev/m ³ for Santiago; with S9 ~60 rev/m ³ . Strong correlation between activity on TA98 with S9 and PAH concentration	<u>Koyano et al. (2002)</u>
Pamplona, Colombia (2010)	PM _{2.5} from an urban location, collected on GFFs. DCM Soxhlet extraction	TA98, TA100, plate incorporation assay, with rat liver S9	Significant mutagenic activity (per μg of EOM) with responses up to 10-fold above control. Responses higher on TA98	<u>Melendez-Gelvez et</u> <u>al. (2012)</u>
La Plata, Argentina (1994)	TSP from an urban/ industrial location, collected on GFFs with high-volume sampler. Hx Soxhlet extraction	TA98, TA100, plate incorporation assay, with Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) higher on TA98; elevated in autumn and winter, compared with summer. Increased potency corresponded to increase in PAHs	<u>Müller et al. (2001)</u>
La Plata, Argentina (1992)	TSP and PM ₁₀ from an industrial location, collected on GFFs with high-volume sampler. Hx Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) highest on TA100 with S9. Potency of TSP higher than that of PM_{10}	<u>Alzuet et al. (1996)</u>
La Plata, Argentina (2000)	PM ₁₀ from 3 industrial locations, collected with cascade impactor. ASE with Hx	TA98, plate incorporation assay, with rat liver S9	Mutagenic potency (per mg of PM) associated with fine PM (< 1.5 μ m) and ultrafine PM (< 0.49 μ m). Potency (per m ³) of ultrafine fraction 10-fold higher than that of larger PM (0.49–3.0 μ m). Potency highest at industrial site in winter. Mutagenic potency with S9 correlated with levels of PAHs. Maximum potency S9 ~1.2 rev/m ³	<u>Massolo et al.</u> (2002)

^a YG1021: TA98 with plasmid pYG216, NR-overproducing strain; YG1024: TA98 with plasmid pYG219, OAT-overproducing strain; YG1041: TA98 with plasmid pYG233, NR- and OAToverproducing strain; YG1026: TA100 with plasmid pYG216, NR-overproducing strain; YG1029: TA100 with plasmid pYG219, OAT-overproducing strain; YG1042: TA100 with plasmid pYG233, NR- and OAT-overproducing strain.

Ac, acetone; ASE, accelerated solvent extraction; CX, cyclohexane; DCM, dichloromethane; EOM, extractable organic matter; GFFs, glass-fibre filters; Hx, hexane; MeOH, methanol; NR, nitroreductase; OAT, O-acetyltransferase; PAHs, polycyclic aromatic hydrocarbons; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; rev, revertants; TSP, total suspended particles.

Supplemental Table S7 Summary of studies that used bacterial mutagenicity assays (e.g. Ames assay) to assess the ability of outdoor air to induce genetic mutations – Europe

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Rijnmond area, Netherlands	Airborne PM collected on GFFs with high-volume sampler. Soxhlet extraction with MeOH, CX, BZ, or Ac, liquid–liquid fractionation	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Increased potency (per m ³ equiv) with S9. Polar solvent confirmed as satisfactory for PM mutagen extraction. Higher potency for samples collected downwind of urban/industrial areas (~40–60 rev/m ³), relative to locations influenced by sea air. Maximum potency on TA98 without S9 ~50 rev/m ³ ; with S9 ~60 rev/m ³	<u>de Raat (1983)</u>
Rijnmond area, Netherlands (1981)	Airborne PM from 5 locations, collected on GFFs with high- volume sampler. MeOH:Ac (1:1) Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Strong influence of location and wind direction on potency (per m ³). Potency lower when wind from south-west (from sea). Highest potency for urban/industrial locations. Most samples more potent with S9. Follow-up work showed correlations between mutagenic activity and PAH concentration	<u>de Raat et al.</u> (1985, 1987)
Rijnmond area, Netherlands (1982)	Airborne PM from 3 locations, collected on GFFs with high- volume sampler. MeOH Soxhlet extraction	TA98, TA98NR, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) generally elevated without S9; somewhat higher at location downwind of major roadway. Significant decline in activity on TA98NR relative to TA98. Diurnal trend, with decreasing potency with S9 between morning and evening. Elevated activity corresponded with winds from south-west (Rotterdam). Maximum potency on TA98 without S9 ~58 rev/m ³ ; with S9 ~63 rev/m ³	<u>de Raat & de</u> <u>Meijere (1988)</u>
Rijnmond area, Netherlands	Airborne PM and SVOCs from 2 locations, collected on GFFs and PUFs. MeOH or Ac Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) of PM extracts far exceeded that of PUF extracts, which yielded false positives. Potency of PM from more-urban sites higher with S9; opposite for less-urban location. Maximum potency on TA98 without S9 ~35 rev/m ³ ; with S9 ~38 rev/m ³	<u>de Raat et al.</u> (1987)
Maastricht, Netherlands	TSP, PM ₁₀ , and PM _{2.5} from 6 locations with pronounced differences in traffic, collected on GFFs with high-volume sampler. DCM Soxhlet extraction	TA98, pre-incubation assay, with and without rat liver S9	Mutagenic potency (per mg of PM) generally elevated without S9; $PM_{2.5}$ higher than PM_{10} or TSP. Some correspondence with traffic density, except city periphery, more-rural location	<u>de Kok et al.</u> (2005)
Several locations, Netherlands (1985–1986)	Airborne PM from 4 urban locations and 1 rural location, collected on GFFs with high- volume sampler. MeOH Soxhlet extraction, extensive fractionation	TA98, TA98NR, TA98/1,8-DNP ₆ , TA100, TA97, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) of crude extract higher with S9. Strong contributions of mono-nitroarene fractions to activity without S9 (~24%). PAH fraction accounted for ~5% of activity with S9. Substantial reduction in activity on TA98NR, relative to TA98. Maximum potency on TA98 with S9 ~43 rev/m ³ ; without S9 ~26 rev/m ³	<u>de Raat et al.</u> (1994)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Wageningen and Terschelling, Netherlands (1979–1980)	Airborne PM from 2 rural sites, collected on GFFs with high- volume sampler. MeOH Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced liver S9	Monitoring for a year. Similar potency (per m ³) with and without S9. Marked day-to-day variability, but generally higher values in autumn, relative to summer. Potency heavily influenced by wind direction; winds from east and south-east (i.e. Germany and Belgium) associated with higher potency. Maximum potency on TA98 without S9 ~20 rev/m ³	<u>Alink et al.</u> (1983)
Wageningen, Netherlands (1982–1983)	PM collected by local meteorological station, on GFFs. MeOH Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Potency (per m ³) of outdoor samples from a rural town elevated with S9. Potency on TA98 without S9 \sim 5 rev/m ³ ; with S9 \sim 7 rev/m ³	<u>van Houdt et</u> <u>al. (1984)</u>
Wageningen, Netherlands (1988–1990)	PM collected by local meteorological station, on GFFs. MeOH Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher with S9 when wind from south; higher without S9 when wind from north-west and air quality improved. Maximum potency on TA98 with S9 ~10 rev/m ³ ; without S9 ~8 rev/m ³	<u>Heussen (1991)</u>
Wageningen and Terschelling, Netherlands (1979–1985)	Airborne PM collected from a rural town and an island, collected on GFFs with low- or high-volume sampler. MeOH Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) elevated with S9 and generally higher in autumn and winter months. Potency of samples from background site (Terschelling) frequently exceeded that for rural site (Wageningen). Maximum potency on TA98 with S9 ~35 rev/m ³ ; without S9 ~18 rev/m ³ . Negative correlation between potency and temperature; positive correlation with NO ₂ , SO ₂ , and CO. Strong influence of wind direction and air mass trajectory	<u>van Houdt et</u> <u>al. (1987)</u>
Wageningen, Netherlands	PM collected by local meteorological station. MeOH Soxhlet extraction	TA98, standard plate incorporation assay, with and without induced and uninduced rat liver S9, induced and uninduced mouse liver S9, mouse and rat lung S9	Potency (per m ³) elevated with S9 and far higher in winter than in summer. Maximum potency with S9 ~12 rev/m ³ ; negligible potency without S9. Highest activity with induced rat liver S9. Lung S9 results similar to those for uninduced liver	<u>van Houdt et</u> <u>al. (1988)</u>
Liège, Belgium (1980)	Airborne PM, collected on GFFs with high-volume sampler. Extraction with BZ, fractionation on silica	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic activity (per μ g of EOM) on TA98 higher without S9; slight elevation with S9 on TA100. Highest activity in aromatic fraction; elevated with S9. Substantial activity in polar fraction without S9	<u>de Wiest et al.</u> (<u>1982)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Flanders, Belgium (2000)	PM_{10} from urban, rural, and industrial sites, collected on GFFs with low-volume sampler. ASE with THF/Hx (20:80), or aqueous PM_{10} suspension	TA98, standard plate incorporation assay, with and without rat liver S9	Highest potency for PM_{10} extract from urban site with S9. Without S9, only the highest concentration (m ³ equiv) elicited a positive response for the urban and industrial locations. With S9, both the PM_{10} suspensions and the PM_{10} extracts elicited positive responses. Urban PAH concentration (ng/m ³) higher than for rural and industrial sites	<u>Brits et al.</u> (2004)
Flanders, Belgium (2000–2001)	PM ₁₀ and SVOCs from urban, rural, and industrial sites; PM collected on quartz filters and SVOCs on PUFs with high- volume sampler. Ac Soxhlet extraction	TA98, standard plate incorporation assay, with and without rat liver S9	Winter PM extracts from all sites more potent (per m ³ equiv and per µg of PM) with S9. PM extracts for summer samples generally less potent (per m ³ equiv and per µg of PM) than winter samples. PUF extracts generally more potent (per m ³ equiv) with S9, and frequently more potent in summer. Combined (PUF + PM) activity significantly higher in winter than in summer. Total potency with S9 (per m ³) empirically related to PAH concentration. Maximum potency on TA98 ~60 rev/m ³ without S9; ~97 rev/m ³ with S9	<u>Du Four et al.</u> (2004)
Flanders, Belgium (2002)	PM_{10} and SVOCs from 15 urban, rural, and industrial sites; PM collected on quartz filters and SVOCs on PUFs with high- volume sampler. Ac Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Metabolic activation contributed to an increase in potency (per m ³) for SVOCs; similar responses with and without S9 for PM samples. Potency somewhat elevated at urban sites and industrial area with known point sources, but highest potency at rural site. Maximum potency on TA98 ~47 rev/m ³ without S9 and ~43 rev/m ³ with S9 for PM extracts; ~11 rev/m ³ without S9 and ~30 rev/m ³ with S9 for SVOCs	<u>Du Four et al.</u> (2005)
Several locations in Finland (1985)	Airborne PM and SVOCs collected in Helsinki, Lahti, and Ätäri with high-volume sampler; PM collected on filter, SVOCs on XAD-2 resin. Ac extraction, fractionation on silica	TA98, TA100, TA98NR, standard plate incorporation assay, with and without PB/5,6-BF- induced rat liver S9	For rural reference site (Ätäri), minimal or no mutagenic activity. Potency (per m ³) higher with S9. For Helsinki, SVOCs and PM extracts similar in summer; SVOCs higher in winter. Potency on TA98NR substantially reduced relative to TA98. Potency on TA98 ~2–10 rev/m ³ with S9; ~1–8 rev/m ³ without S9. Most polar fraction generally showed highest potency	<u>Tuominen et</u> <u>al. (1988)</u>
Coastal area in central Finland (1985)	Airborne PM and SVOCs collected in Kokkola with high-volume sampler; PM collected on filter, SVOCs on XAD-2 resin. Ac extraction, fractionation on silica	TA98 and TA100, standard plate incorporation assay, with and without PB/5,6- BF-induced rat liver S9	Mutagenic activity in both PM and vapour-phase extracts. Similar potency (per m ³ equiv) on TA98 with and without S9. Potency at urban site higher without S9. Significant response for polar (e.g. nitro-PAH) fraction from urban sample only. Potency on TA98 ~3 rev/m ³ with S9; ~2 rev/m ³ without S9	<u>Pyysalo et al.</u> (<u>1987)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Oslo, Norway (1981)	Airborne PM and SVOCs from 2 urban sites, collected on GFFs and XAD-2 with high-volume sampler. Ac Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced liver S9	All XAD extracts more potent (per m ³) without S9. Similar potency (per m ³) of PM extracts with and without S9. Percentage of total activity on XAD increased with increasing temperature. No relationship between mutagenic activity and PAH concentration. Maximum potency on TA98 ~136 rev/m ³ with S9; ~57 rev/m ³ without S9	<u>Alfheim et al.</u> (1985)
Oslo, Norway (1978)	Airborne PM from 2 urban sites, collected on GFFs and XAD-2 with high-volume sampler. CX Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced liver S9	Mutagenic potency (per m ³) frequently elevated with S9; generally higher in winter than in spring. Potency (per m ³) similar for both sites; potency on TA98 per mg of PM 2-fold higher at industrial site compared with heavy-traffic site. Strong influence of meteorological conditions, and diurnal changes in mutagenic potency followed changes in SO ₂ . Maximum potency on TA98 without S9 ~12 rev/m ³ ; with S9 ~11 rev/m ³	<u>Møller &</u> <u>Alfheim (1980)</u>
Oslo, Norway (1979)	Airborne PM from 2 urban sites, collected with cascade impactor. Ac Soxhlet extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced liver S9	Mutagenic potency (per m ³) on TA98 of street-level samples higher with S9; rooftop samples sometimes higher without S9. Night-time samples generally far less potent than daytime samples. Maximum potency on TA98 without S9 ~45 rev/m ³ ; with S9 ~180 rev/m ³ . Mutagenic activity of street samples related to traffic intensity and CO, NO, and PAH levels. Activity with S9 associated with acidic and neutral-aromatic fractions	<u>Møller et al.</u> (1982)
Scandinavia (1979–1981)	Airborne PM from 5 locations in Sweden and 8 locations in Norway, collected on GFFs with high-volume sampler or cascade impactor. Ac Soxhlet extraction, acid-base-neutral fractionation	TA98, TA98NR, TA98/1.8-DNP ₆ , YG1021, YG1024, standard plate incorporation assay, with and without Aroclor 1254-induced liver S9	Elevated mutagenic potency (per m ³) without S9 for Stockholm; elevated potency with S9 for Oslo. Substantial increases in winter, and higher potency for urban areas relative to background. Highest mutagenic potency for PM < 0.5 μ m. Potency reductions on NR- and OAT-impaired strains. Reduced potency for samples collected at high elevations. Fractionation showed high activity without S9 for fractions containing polar compounds (e.g. ketones, quinones, acids, and N-heterocyclics). Maximum potency ~68 rev/m ³ without S9 (Stockholm); ~180 rev/m ³ with S9 (Oslo)	<u>Alfheim et al.</u> (1983)
Sundsvall, Sweden (1981)	Airborne PM from 4 sites near aluminium smelting plant, collected on GFFs with high- volume sampler. Ac Soxhlet extraction	TA98, TA100, TA98NR, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) comparable to other Scandinavian urban areas, and marginal influence of aluminium smelter. Similar responses with and without S9. Similar responses for winter and summer. Decreased potency without S9 on TA98NR relative to TA98. Maximum potency ~20 rev/m ³ without S9; ~22 rev/m ³ with S9	<u>Alfheim et al.</u> (1984)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Stockholm, Sweden	Airborne PM from city centre, collected on GFFs with high- volume sampler. DCM Soxhlet extraction, fractionation on silica	TA98, TA98NR, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Analysis of subfractions of moderately polar fraction indicated activity without S9 for polar fractions containing aromatic acids and ketones. Substantial reduction of subfraction mutagenicity on TA98NR, relative to TA98. Maximum potency without S9 ~30 rev/m ³ ; with S9 ~94 rev/m ³	<u>Strandell et al.</u> (1994)
Gothenburg, Sweden (1981–1984)	Airborne PM from 13 locations including tunnel, collected on GFFs, electrostatic precipitator and consecutive high-volume sampler. Ac Soxhlet extraction	TA96, TA98, TA100, TA102, TA104, TA98NR, TA98/1.8-DNP ₆ , standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) on TA98 of most samples higher without S9; tunnel samples higher with S9. Generally higher potency (per mg of PM) in winter, except for tunnel samples. Urban samples generally more potent (per m ³) than coastal samples. Maximum potency on TA98 without S9 ~130 rev/m ³ ; with S9 ~110 rev/m ³ . Potency (per mg of PM) substantially reduced on NR- and OAT-deficient strains	Löfroth et al. (1985), Alfheim & Lindskog (1984)
Copenhagen, Denmark (1988)	Airborne PM from 10 suburban locations, collected on GFFs with high-volume sampler. Separate DCM and Ac sonication extraction	TA98 and TA98NR, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Similar potency on TA98 (per m ³) with and without S9 for both solvents. Some samples showed increased activity for Ac extract. Reduced potency on TA98NR relative to TA98. Average potency of DCM extract significantly higher for TA98NR. Maximum potency on TA98 without S9 ~19 rev/m ³ ; with S9 ~20 rev/m ³	<u>Nielsen (1992)</u>
Copenhagen, Denmark (1981)	Airborne PM from 2 urban sites, collected on GFFs with high- volume sampler. Sequential Soxhlet extraction with CX and MeOH, detailed fractionation	TA1537, TA1538, TA100, TA1535, plate incorporation assay, with and without PB/5,6-BF- induced rat liver S9	Mutagenic potency of PAH fraction on TA1538 without S9 (per m ³) higher at street level than at rooftop (22 m). Height effect less pronounced for polar fraction. Maximum potency on TA1538 ~22 rev/m ³ without S9. Potency of PAH fraction from street level correlated with lead levels	<u>Madsen et al.</u> (1982)
Copenhagen, Denmark	Airborne PM from urban site and reference, collected with electrostatic sampler. ASE with Hx:Ac (1:1), fractionation on silica	TA98, YG5161, YG1041, microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) higher without S9, with little difference between urban site and reference site. Higher potency on YG1041, particularly for polar fraction without S9. No increase in potency on YG5161, relative to TA98	<u>Sharma et al.</u> (<u>2007)</u>
Copenhagen, Denmark (1996, 1998)	Airborne PM from urban and semi-rural sites, collected on GFFs with high-volume sampler. Sequential sonication extraction with DCM and Ac	TA98, TA98NR plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) higher with S9; for rural site, higher without S9. Degree of contribution of PAHs influenced by air mass movement. Air masses from central Europe enriched in mutagenic activity, PAHs, and nitro-PAHs. Maximum potency on TA98 with S9 ~335 rev/m ³ ; without S9 ~150 rev/m ³	<u>Feilberg et al.</u> (2002)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Bohemia region, Czech Republic (2000–2001)	PM ₁₀ from 3 sites in northern Bohemia, collected on GFFs with high-volume sampler. DEE:Hx (1:9) Soxhlet extraction	TA98, YG1041, standard plate incorporation assay, with and without Delor 103 (PCB)-induced rat liver S9	Significant elevation in potency (per m ³) for winter for both strains with and without S9. Increase in potency on TA98 with S9. YG1041 more potent without S9. No significant difference in potency (per m ³) across sites. Significant correlation between potency on TA98 with S9 (per μ g of EOM) and potency on YG1041 with and without S9 (per μ g of EOM) and levels of PAHs. Maximum potency on TA98 without S9 ~12 rev/m ³ ; with S9 ~25 rev/m ³	Binková et al. (2003)
Bohemia region, Czech Republic (1996–1997)	PM ₁₀ from polluted location in northern Bohemia and rural control, collected on GFFs with high-volume sampler. DCM Soxhlet extraction, acid-base- neutral fractionation	TA98, YG1041, standard plate incorporation assay, with and without Delor 103 (PCB)-induced rat liver S9	Mutagenic potency (per m ³) marginally higher at polluted site; elevated in winter, and higher in winter with S9. Potency of winter samples from less-polluted site higher without S9. Marked increase in potency on YG1041, relative to TA98. Maximum potency on TA98 without S9 ~24 rev/m ³ ; with S9 ~30 rev/m ³ . Significant contribution (25–50%) of acidic compounds to activity without S9	<u>Cerná et al.</u> (2000)
Bohemia region, Czech Republic (1996–1997)	PM ₁₀ from 2 heavy-traffic locations and 2 rural locations, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, YG1041, microsuspension and plate incorporation assays, with and without Delor 103 (PCB)-induced rat liver S9	Mutagenic potency (per m ³ or per µg of EOM) significantly higher in winter. Significant increase in potency on YG1041 relative to TA98. Microsuspension results did not show the same increases in potency in winter. Higher potency at industrial and high-traffic locations. Correlation between mutagenic activity and carcinogenic PAHs. Maximum potency (plate incorporation) on TA98 without S9 ~107 rev/m ³ ; with S9 ~85 rev/m ³	<u>Cerná et al.</u> (<u>1999)</u>
Brno, Czech Republic (2001)	PM ₁₀ , TSP, and SVOCs from 2 urban sites, collected on Teflon- coated filters, GFFs, and PUFs with high-volume sampler. DCM Soxhlet extraction, fractionation on silica	TA98, YG1041, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher with S9; 15–40% of total activity associated with SVOCs. Maximum potency of PM_{10} on TA98 ~10 rev/m ³ with S9; ~7 rev/m ³ without S9. Substantial increase in potency without S9 on YG1041, compared with TA98	<u>Ciganek et al.</u> (2004)
Czech Republic (1999–2003)	Airborne PM from 4 towns with different levels of industrial and commercial activities, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, YG1041, standard plate incorporation assay, with and without rat liver S9	Mean mutagenic potency (per m ³) higher with S9; elevated at heavy-traffic urban location (Prague). Significant elevation on NR- and OAT-enhanced strain without S9, relative to TA98 (100- fold). Maximum potency on TA98 ~41 rev/m ³ with S9; ~24 rev/m ³ without S9. Correlation between mutagenic activity and PAH levels	<u>Pastorková et</u> <u>al. (2004)</u>
Wrocław, Poland (2007)	PM_{10} and $PM_{2.5}$ from an urban site, collected on sintered glass filters. DCM Soxhlet extraction, fractionation on silica	TA98, TA100, YG1041, YG1042, plate incorporation assay, with and without rat liver S9	Enhanced potency (per m ³) for PM_{10} , relative to $PM_{2.5}$, $PM_{2.5}$ potency reduced with S9. PM_{10} potency similar with and without S9. Potency with and with S9 increased on YG1041 and YG1042, relative to parent strains. Maximum potency on TA98 ~4 rev/m ³ with S9; ~50 rev/m ³ without S9	<u>Piekarska</u> (2009)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Wrocław, Poland (2007)	PM ₁₀ from an urban site, collected on sintered glass filters. DCM Soxhlet extraction, fractionation on silica	TA98, YG1041, plate incorporation assay, with and without rat liver S9	Potency (per m ³) higher without S9 and higher in winter than in summer. Increased potency on YG1041, particularly for winter samples. High mutagenic activity in winter fractions containing nitro-PAHs and summer fractions containing nitro-PAHs and dinitro-PAHs. Maximum potency on TA98 ~20 rev/m ³ with S9; ~30 rev/m ³ without S9	<u>Piekarska et al.</u> (2009)
Wrocław, Poland (1997–1998)	PM_{10} from an urban site, collected on GFFs. DCM Soxhlet extraction, fractionation on silica	TA98, YG1021, YG1024, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) elevated with S9 and elevated in winter compared with summer. Elevated activity on YG1021 and YG1024, relative to TA98, with highest activity in aromatic and polar fractions	Jadczyk & Kucharczyk (2005), Zwodziak et al. (2001)
Wrocław, Poland	Airborne PM from an urban site, collected on GFFs. DCM Soxhlet extraction	TA98, TA100, YG1041, YG1042, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) on TA98 elevated without S9 for winter. Similar responses with and without S9 for summer. Highest responses on YG1041 with S9. Maximum potency on TA98 without S9 ~24 rev/m ³ ; with S9 ~12 rev/m ³	<u>Piekarska & Karpińska-</u> <u>Smulikowska</u> (2007)
Wrocław, Poland	Airborne PM from an urban site, collected on GFFs. DCM Soxhlet extraction	TA98, YG1041, plate incorporation assay, with and without Aroclor 1254- or PB-induced rat liver S9	Mutagenic potency (per m ³) higher for winter. Higher activity without S9 for winter only. Similar potency for Aroclor- and PB-induced S9. Dramatic increase in potency on YG1041, relative to TA98. Maximum potency on TA98 without S9 ~11 rev/m ³ ; with S9 ~15 rev/m ³	Piekarska & Karpińska- Smulikowska (2006)
Wrocław, Poland (2007)	PM ₁₀ from an urban site, collected on sintered glass filters. DCM Soxhlet extraction	TA98, TA100, YG1041, YG1042, plate incorporation assay, with and without rat liver S9	Higher mutagenic potency (per m ³) without S9 and elevated in autumn (~5-fold), relative to summer. Maximum potency on TA98 without S9 ~17 rev/m ³ ; with S9 ~11 rev/m ³ . Highest responses on YG1041	<u>Piekarska</u> (2010)
Wrocław, Poland	Airborne PM from 2 urban sites, collected on glass filters. DCM Soxhlet extraction	TA98, YG1041, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) on TA98 higher without S9, elevated in winter, and higher at heavy-traffic site. Potency elevated on YG1041, relative to TA98. Maximum potency on TA98 without S9 ~150 rev/m ³ ; with S9 ~300 rev/m ³	<u>Piekarska et al.</u> (2011)
Wrocław, Poland (1997–1998)	Airborne PM from an urban location, 12 composite monthly samples, collected on GFFs. DCM Soxhlet extraction	TA98, standard plate incorporation assay, with and without rat liver S9	All samples elicited significant positive responses. Frequently higher responses with S9 activation, highest responses in winter months. Far smaller winter sample (m ³ equiv) required to elicit a positive response. Maximum potency on TA98 without S9 ~6 rev/m ³ ; with S9 ~10 rev/m ³	<u>Jadczyk &</u> <u>Kucharczyk</u> (2000)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Silesia, Poland (1984–1985)	Airborne PM from 23 locations, collected on GFFs with high- volume sampler. BZ Soxhlet extraction, fractionation on silica	TA100, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per mg of EOM) enhanced with S9 for moderately polar fractions (e.g. PAHs and heterocyclics), particularly for winter months. Potency (per m ³) enhanced with S9 for winter months, but potency of more polar fractions equal with and without S9, or higher without S9. Maximum potency with and without S9 ~200 rev/m ³ . Increased potency with decreasing temperature	<u>Motykiewicz</u> <u>et al. (1989)</u>
Silesia, Poland (1987)	Airborne PM from 20 locations, collected on GFFs with high- volume sampler. CX Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) on TA98 elevated with S9, particularly in winter. Potency on TA100 generally higher in winter, especially without S9. Maximum potency on TA98 with S9 ~90 rev/m ³ ; without S9 ~32 rev/m ³ . Potency inversely correlated with temperature	<u>Motykiewicz</u> <u>et al. (1985,</u> <u>1990</u>)
Ajka and Pápa, Hungary (1982–1984)	Airborne PM from highly polluted Ajka and non- industrial Pápa, collected on GFFs with high-volume sampler. BZ:Ac (19:1) Soxhlet extraction or DCM sonication extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) highest on TA98 with S9, and potency 3–9-fold higher in winter compared with summer. Winter values at non-industrial site higher than at industrial site, but potency (per m ³) higher at industrial site. Maximum potency on TA98 with S9 ~32 rev/m ³	<u>Pintér et al.</u> (<u>1990), Török</u> et al. (<u>1989)</u>
Leipzig, Germany (2000)	PM_{10} from 2 urban locations, collected with cascade impactor. ASE with Hx	TA98, plate incorporation assay, with rat liver S9	Mutagenic potency (per mg of PM) associated with fine PM (< 1.5 μ m) and ultrafine PM (< 0.49 μ m). Potency (per m ³) of ultrafine fraction 10-fold higher than that of larger PM (0.49–3.0 μ m). Potency generally higher in winter. Mutagenic potency with S9 correlated with levels of PAHs. Maximum potency with S9 ~0.1 rev/m ³	<u>Massolo et al.</u> (2002)
Berlin, Germany (1980–1981)	Airborne PM from 3 locations, collected with cascade impactor. Soxhlet or sonication extraction with CX	TA98, plate incorporation assay, with rat liver S9	Mutagenic potency (per m ³) at the suburban site showed strong winter peak and decline in spring. Industrial and urban sites showed a decline in summer. Maximum potency with S9 ~14 rev/m ³ . PAH concentration ratios suggest domestic heating emissions in winter. Correlation between mutagenic potency and SO ₂ levels	<u>Israël &</u> <u>Busing (1983)</u>
Berlin, Germany	TSP collected during a 4 day smog event. CX extraction	TA98, plate incorporation assay, with rat liver S9	Mutagenic potency (per m ³) during smog event > 4-fold higher than other outdoor measurements. Maximum potency ~265 rev/m ³	<u>Israel et al.</u> <u>(1984)</u>
Berlin, Germany (1983–1984)	Airborne PM from 2 urban sites, collected with cascade impactor. CX:DEE (2:8) sonication extraction, acid-base-neutral fractionation	TA98, TA100, TA98NR, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) higher with S9; higher on TA98 than on TA100. Maximum potency with S9 ~80 rev/m ³ ; without S9 ~41 rev/m ³ . Marked decline in potency during summer. Higher potency at residential site. Highest activity in neutral polar organic fraction. Marked decrease on NR-deficient strain, particularly for polar fraction	<u>Moriske et al.</u> (1985)

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Berlin, Germany (1983–1985)	Airborne PM from 2 urban sites and a highway tunnel, collected with cascade impactor. CX:DEE (2:8) sonication extraction, acid-base-neutral and TLC fractionation	TA98, TA98NR, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	For urban sites and highway tunnel, mutagenic potency (per µg of EOM) higher with S9 and elevated in neutral and acid fractions. Highest activity in polar organic fraction. Activity of fractions from tunnel samples showed 10–25% reduction on TA98NR compared with TA98	<u>Moriske &</u> Rüden (1988)
Berlin, Germany (1981–1982)	Airborne PM collected with cascade impactor. DEE:BZ (9:1) sonication extraction	TA98, TA100, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) of 0.4–0.7 μ m size fraction elevated with S9; higher in autumn and winter compared with spring and summer. Maximum potency on TA98 with S9 ~22 rev/m ³ ; without S9 ~11 rev/m ³	<u>Wullenweber</u> et al. (1982, <u>1984</u>)
Berlin, Germany (1981)	Airborne PM collected with cascade impactor. DEE sonication extraction, polar organic and PAH fractions	TA98, TA100, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per μ g of EOM) much higher for polar aromatics compared with PAH fraction. Polar aromatics slightly elevated without S9; PAH fraction higher with S9. Higher mutagenic activity for smaller particles (0.4–1.3 μ m), compared with particles of 1.3–10.2 μ m. Maximum potency on TA98 with S9 ~8 rev/m ³ ; without S9 ~6 rev/m ³	<u>Moriske et al.</u> (1982)
Berlin, Germany (1981–1982)	Airborne PM collected with cascade impactor. DEE:BZ (9:1) sonication extraction, acid- base-neutral fractionation	TA98, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) of ultrafine PM (< 0.4μ m) marginally higher without S9; significantly elevated in autumn and winter compared with summer. Some indication of reduced mutagenicity on weekends. Substantial portion of mutagenic activity in acidic (18–31%) and neutral (41–66%) fractions. Maximum potency on TA98 with S9 ~15 rev/m ³ ; without S9 ~17 rev/m ³	<u>Gottlieb et al.</u> (1983)
Herne, Germany (1977)	Airborne PM from urban site in Ruhr area, collected on polystyrene or cellulose nitrate filters. DCM or DEE extraction	TA98, plate incorporation assay, with and without PCB-induced mouse liver S9	Mutagenic potency (per μ g of PM) higher with S9. Highest activity in PAH and polar fractions; ~30–50% of the activity does not require S9	<u>Hoffmann et</u> <u>al. (1980)</u>
Baden- Württemberg, Germany (1995)	Airborne PM from an urban, site, collected on GFFs with high-volume sampler. Ac Soxhlet extraction	Plate incorporation assay with Ames II base-pair substitution strains TA7001, TA7002, TA7003, TA7004, TA7005, TA7006, without S9 activation	Mutagenic potency (per m ³) enhanced on TA7005 (CG to AT transversions), TA7004 (CG to TA transitions), TA7002 (TA to AT transversions), and TA70006 (CG to GC transversions). Potency higher in winter	<u>Erdinger et al.</u> (2004)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Baden- Württemberg, Germany (1993)	Airborne PM from 8 locations including a background forest site, collected on GFFs with high-volume sampler. Ac Soxhlet extraction	TA98, TA100, TA98NR, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	No significant difference between potency (per m ³) with and without S9. Higher potency generally associated with more- congested urban sites; highest activity associated with a low- traffic site. Maximum potency on TA98 without S9 ~30–100 rev/ m ³ for urban and suburban sites; ~10 rev/m ³ for background site. Significant but seasonally variable decrease on TA98NR; positive correlation between mutagenic activity and NO, NO ₂ , and SO ₂	Erdinger et al. (2005)
Duisburg, Germany (~1977)	Airborne PM, Draeger Box Micron filter. MeOH extraction, fractionation on alumina	TA98, TA1537, TA1538, TA100, standard plate incorporation assay, with and without Clophen C-induced rat liver S9	Enhanced potency with S9, and highest potency for crude MeOH extract. Low activity in PAH-containing fraction, relative to crude extract	<u>Dehnen et al.</u> (1978)
Piedmont, Tuscany, and Sicily, Italy	PM_{10} and/or $PM_{2.5}$ from 5 locations, collected on GFFs with low-volume sampler. Ac Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher without S9. Similar potency across sites, although slightly elevated at industrial and high-traffic site. $PM_{2.5}$ potency higher than site-matched PM_{10} potency. Maximum potency on TA98 ~8 rev/m ³ with S9; ~20 rev/m ³ without S9	<u>Gilli et al.</u> (2007c)
Bormida Valley, Italy (1989)	PM ₁₀ from 6 locations, collected on GFFs with high- volume sampler. Sequential Ac sonication and Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) similar with and without S9. Significant negative relationship between mutagenic potency and air temperature, and significant positive relationship between potency and traffic density. Samples near industrial zone did not yield high potency values. Maximum potency on TA98 ~23 rev/m ³ with S9; ~24 rev/m ³ without S9	<u>Scarpato et al.</u> (1993)
La Spezia, Italy (1988)	Airborne PM from an urban location, collected on GFFs, with high-volume sampler. CX sonication extraction, PAH- containing TLC fraction	TA98, TA98NR, TA98/1,8- DNP ₆ , plate incorporation assay, with and without PB/5,6-BF-induced rat liver S9	Mutagenic potency (per m ³) higher with S9, and positively correlated with PAH concentration. Mutagenic potency and PAH concentration declined with increasing air temperature. No appreciable differences on TA98NR and TA98/1,8-DNP ₆ , relative to TA98. Maximum potency on TA98 ~17 rev/m ³ with S9; ~3 rev/m ³ without S9	<u>Barale et al.</u> (1991a)
Brescia, Italy (1991)	PM_{10} from a residential area, collected on GFFs with cascade impactor. DCM Soxhlet extraction	TA98, TA98/1,8-DNP ₆ , microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of EOM or m ³) similar with and without S9 for PM of 0.5–10 μ m. PM < 0.5 μ m more potent than larger PM and more potent without S9. Substantial potency reduction on TA98/1,8-DNP ₆ compared with TA98. Maximum potency on TA98 ~5 rev/m ³ with S9; ~4 rev/m ³ without S9	<u>Monarca et al.</u> (1997)
Unidentified town, Italy	Airborne PM from a mid- sized town, collected on GFFs with high-volume sampler. Ac Soxhlet extraction	TA98, TA98NR, plate incorporation assay, without Aroclor 1254-induced rat liver S9	Higher potency (per m ³) with S9, and elevated in autumn and winter. Maximum potency ~40 rev/m ³ with S9; ~35 rev/m ³ without S9. Substantial reductions in potency on TA98NR relative to TA98	<u>Morozzi et al.</u> <u>(1992)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Several towns, Italy (1988)	PM ₁₀ from 11 towns, collected on GFFs with high-volume sampler. Ac sonication extraction	TA98, TA100, TA98NR, TA100NR, plate incorporation assay, with and without PB/5,6-BF- induced rat liver S9	Average TA98 mutagenic potency (per m ³) mostly higher without S9. Significant correlations between TA98 activity with and without S9 and NO ₂ , CO, and NMHC. Maximum potency on TA98 ~280 rev/m ³ without S9; ~20 rev/m ³ with S9. Substantial reductions of NR-deficient strains	<u>Barale et al.</u> (1991b)
Several towns, Italy (1990)	PM ₁₀ from 17 towns, collected on GFFs with high-volume sampler. DCM sonication extraction, acid–base–neutral fractionation, TLC fractionation	TA98, TA98NR, TA98/1,8- DNP ₆ , plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) generally modestly elevated with S9; potency generally higher at colder temperatures. Potency positively correlated with lead levels. Acid and polar fraction generally showed higher activity, and potency reduced 20–38% on TA98NR and 48–77% on TA98/1,8-DNP ₆ , relative to TA98. Maximum potency on TA98 without S9 ~100 rev/m ³ ; without S9 ~150 rev/m ³	<u>Barale et al.</u> (1994)
Pisa, Italy (1986–1987)	Airborne PM from 5 locations in a non-industrial town, collected on GFFs with high-volume sampler. Ac Soxhlet extraction	TA98, TA100, pre- incubation assay, with and without PB/5,6-BF induced mouse liver S9	Mutagenic potency (per m ³) higher with S9; potency without S9 significantly elevated in autumn and winter. Potency (per µg of PM) on TA98 without S9 significantly elevated in autumn and winter. Potency (per m ³) generally lower at rural site; higher but variable at sites with moderate to heavy traffic. Maximum potency on TA98 without S9 ~140 rev/m ³ ; without S9 ~28 rev/m ³	<u>Barale et al.</u> (1989)
Pisa, Italy (1993–1994)	Airborne PM from 2 locations, collected on cellulose nitrate filter with low-volume sampler. Sequential extraction with DCM and MeOH	TA98, TA100, plate incorporation assay, without rat liver S9	Significant response only for high-traffic site, and mutagenic activity higher with S9. Similar responses in summer and autumn. Maximum potency on TA98 without S9 ~80 rev/m ³ ; with S9 ~180 rev/m ³	<u>Bronzetti et al.</u> (1997)
Pisa, Italy	Airborne PM from 3 urban locations, collected on GFFs. Sequential sonication extraction with DCM and Hx	TA98, TA100, plate incorporation assay, with and without PB/5,6-BF- induced mouse liver S9	Mutagenic potency (per m ³) generally higher at higher-traffic location. Potency generally higher with S9 in autumn and winter; without S9 in summer. Maximum potency on TA98 without S9 ~33 rev/m ³ ; with S9 ~33 rev/m ³	<u>Vellosi et al.</u> (1994)
Turin, Italy (2001–2004)	$PM_{2.5}$ from a high-traffic location, collected on Teflon filters with high-volume sampler. Ac Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mean mutagenic potency (per m ³) higher without S9. Pooled monthly samples showed elevated potency in autumn and winter compared with summer, and this corresponds to higher PM _{2.5} levels. Some positive correlations between mutagenic potency and PAH levels. Maximum potency on TA98 without S9 ~100 rev/m ³ ; with S9 ~35 rev/m ³	<u>Gilli et al.</u> (2007a)
Turin, Italy (2003–2004)	PM ₁₀ from an urban site, collected on GFFs with high- volume sampler. Ac Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher without S9 and elevated in autumn and winter, relative to summer. Maximum potency on TA98 without S9 ~38 rev/m ³ ; with S9 ~29 rev/m ³	<u>Gilli et al.</u> (2007b)

Geographical location	Test article	Salmonella strains²/assay version	Results	Reference
Turin, Italy (2007)	PM_{10} and $PM_{2.5}$ from 2 urban sites, collected on GFFs with high-volume sampler. Ac Soxhlet extraction	TA98, TA98NR, YG1021, YG1024, plate incorporation assay, without S9 activation	Mutagenic potency on TA98 (per m ³) higher in autumn and winter; elevated for high-traffic site. Increase in activity on YG1021 and decrease on TA98NR, relative to TA98. Potency of PM _{2.5} higher than that of PM ₁₀ . Maximum potency on TA98 without S9 ~113 rev/m ³	<u>Traversi et al.</u> (2011)
Padana Plain, Italy (2006)	PM _{2.5} from high-traffic locations in 3 cities, collected on Teflon filters with high-volume sampler. Ac Soxhlet extraction	TA98, TA98NR, YG1021, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher without S9. No appreciable difference between locations. Without S9, substantial reduction in activity on TA98NR, relative to TA98; substantial increase in activity on YG1021, relative to TA98. Maximum potency on TA98 without S9 ~35 rev/m ³ ; with S9 ~20 rev/m ³	<u>Traversi et al.</u> (2009)
Padana Plain, Italy (1994–1995)	PM _{2.5} from high-traffic locations in Bologna, collected with cascade impactor. Sequential Soxhlet extraction with Ac and DCM	TA98, TA100, plate incorporation assay, with and without PB/5,6-BF- induced rat liver S9	Mutagenic potency (per m ³) higher with S9; highest potency (per m ³ or per mg of PM) for smallest size fraction (< 0.4 μ m). Maximum potency on TA98 without S9 ~17 rev/m ³ ; with S9 ~36 rev/m ³	<u>Pagano et al.</u> (1996)
Padana Plain, Italy (2005–2006)	PM _{2.5} from high-traffic locations in 3 cities, collected on Teflon filters with high-volume sampler. Ac Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher without S9, and substantially elevated in autumn and winter. No appreciable difference between locations. Maximum potency on TA98 without S9 ~50 rev/m ³ ; with S9 ~38 rev/m ³	<u>Traversi et al.</u> (2008)
Po Valley, Italy (1990–1994)	Airborne PM from residential/ commercial locations in Parma, collected on GFFs with low-volume sampler. Tl or Ac Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without PB/5,6- BF-induced rat liver S9	48 pooled monthly samples analysed; almost all elicited positive response. Similar responses with and without S9 for 3 of 4 years; without S9 exceeded with S9 in final year. Responses frequently elevated in autumn and/or winter months. Elevated airborne PAHs in autumn and winter months	<u>Rossi et al.</u> (1995)
Po Valley, Italy (1990)	Airborne PM from residential/ commercial locations in Parma, collected on GFFs with low- volume sampler. Tl Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without rat liver S9	9 pooled monthly samples; significant mutagenicity for most sampling periods. Responses elevated on TA98 and generally higher for autumn and spring, relative to summer. Similar responses with and without S9, but somewhat elevated with S9 for autumn. Mutagenicity positively correlated with atmospheric CO and NO _x , and negatively correlated with temperature. Maximum potency on TA98 without S9 ~150 rev/m ³ ; with S9 ~110 rev/m ³	<u>Poli et al.</u> (<u>1992)</u>
Po Valley, Italy (1991–1998)	Airborne PM from residential/ commercial locations in Parma, collected on GFFs with low- volume sampler. Tl Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without rat liver S9	96 pooled monthly samples analysed. Continuous evidence of mutagenic activity, with evidence of temporal decline in mutagenic potency (per m ³ equiv) from 1991 to 1998. Earliest samples showed enhanced potency without S9. Highest potency in autumn and winter months. Maximum potency on TA98 without S9 ~130 rev/m ³ ; with S9 ~150 rev/m ³	<u>Poli et al.</u> (1999)

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Po Valley, Italy (1996, 1997)	Airborne PM from residential/ commercial locations in Parma, collected on GFFs with low-volume sampler. Tl or Ac Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without rat liver S9	4 pooled monthly samples analysed; late autumn and late winter samples generally more potent (per m ³ equiv). Ac and Tl extracts showed similar potency, except for TA98 with S9, where Ac extract 2-fold more potent. Similar potency with and without S9, except for TA100, which showed enhanced potency with S9. Maximum potency on TA98 without S9 ~35 rev/m ³ ; with S9 ~40 rev/m ³	Buschini et al. (2001)
Genoa, Italy (1986–1987)	Airborne PM from 10 sites with low, moderate, or high traffic, collected on GFFs with high- volume sampler. CX sonication extraction, TLC fractionation	TA1535, TA98, TA100, TA102, TA104, TA97, TA98NR, TA98/1,8- DNP ₆ , standard plate incorporation assay, with and without rat liver S9	Mutagenic potency (per μ g of EOM) of crude extracts high on TA98 with S9. High frequency of positive responses for TA100 with S9 and TA98 without S9. Marked reduction of some samples on NR- and OAT-deficient strains. Some evidence of seasonal variation, with low activity in summer. TA100 activity with S9 significantly correlated with PAH levels	<u>De Flora et al.</u> (1989)
Emilia- Romagna region, Italy (1999–2000)	TSP, PM ₁₀ , and PM _{2.5} from urban and rural areas, collected on GFFs with low-volume sampler. Tl or Ac Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Higher mutagenic potency (per m^3) during winter months. Potency frequently similar or higher with S9, except for high- traffic locations, where potency reduced with S9. Potency (per μ g of PM) increased with decreasing PM size. Maximum potency on TA98 ~500 rev/m ³ without S9; ~380 rev/m ³ with S9	<u>Cassoni et al.</u> (2004)
Emilia- Romagna region, Italy (2000–2002)	PM _{2.5} from urban and rural areas, collected on quartz filters with low-volume sampler. Ac Soxhlet extraction	TA98, TA100, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) showed clear increase in autumn and winter months; generally higher without S9. Substantial variability in mutagenic potency across sites with similar population and traffic density. Local agreement between mutagenic potency and levels of CO and NO ₂ . Maximum potency on TA98 ~100 rev/m ³ without S9; ~90 rev/m ³ with S9	<u>Cassoni et al.</u> (2004)
Padua, Italy (1990–1991)	Airborne PM from 8 high-traffic locations, collected on GFFs with high-volume sampler. Sequential Soxhlet extraction with DCM and MeOH	TA98, TA100, TA98NR, TA100NR, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³ or per mg of PM) higher with S9, and elevated in winter compared with summer. Decrease in activity on NR-deficient strains relative to parent strains. Maximum potency on TA98 ~51 rev/m ³ without S9; ~98 rev/m ³ with S9	<u>Nardini &</u> <u>Clonfero</u> (1992)
Ispra, Italy (1980–1981)	Airborne PM from rural location, collected with high- volume sampler. Sequential DCM, MeOH extraction, DMSO shaker extraction	TA98, TA1537, TA100, standard plate incorporation assay, with and without PB-5,6-BF- induced rat liver S9	Mutagenic potency (per mg of PM) elevated with S9 and markedly higher in winter, especially for TA1537 and TA100. Maximum potency on TA98 ~10 rev/m ³ without S9; ~10 rev/m ³ with S9. Mutagenicity on TA98 with S9 highest for organic base fraction, followed by polar neutrals	<u>Reali et al.</u> (1984)

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Rome, Italy (1986)	Airborne PM collected on GFFs with high-volume sampler. DCM Soxhlet extraction, acid- base-neutral fractionation	TA98, TA98NR, TA98/1,8- DNP ₆ , standard plate incorporation assay, with and without rat liver S9	Potency (per m ³ equiv) similar with and without S9, and substantially higher in winter. Relatively little reduction on TA98NR relative to TA98; substantial reductions on TA98/1,8- DNP_6 . Neutral and basic fractions elicited strongest responses, enhanced with S9, particularly for winter sample. Maximum potency on TA98 ~18 rev/m ³ without S9; ~17 rev/m ³ with S9	<u>Crebelli (1989)</u>
Rome, Italy (1988)	Airborne PM from heavy-traffic site, collected on GFFs with high-volume sampler. DCM Soxhlet extraction, acid–base– neutral fractionation	TA97a, TA98, TA100, TA102, TA100NR, TA98NR, TA98/1,8-DNP ₆ , <i>E. coli</i> WP2, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of EOM) highest on TA98 with S9, followed by TA97a. Low response on TA102. High potency on TA98 (per mg of EOM) without S9 for acidic fraction; neutral fraction similar with and without S9; basic fraction higher with S9	<u>Crebelli et al.</u> (1991)
Rome, Italy (1990–1991)	PM ₁₀ from heavy-traffic location, collected on GFFs with high-volume sampler. DCM Soxhlet extraction	TA98, microsuspension assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher without S9, highest in winter months and lowest in spring. Maximum potency without S9 ~32 rev/m ³ ; with S9 ~10 rev/m ³ . Microsuspension potency 2–5- fold higher than for plate incorporation. Potency negatively correlated with temperature	<u>Crebelli et al.</u> (1995)
Rome, Italy (1992–1993)	PM ₁₀ from heavy-traffic site, collected on GFFs with high- volume sampler. DCM Soxhlet extraction	TA98, plate incorporation assay, without S9 activation	Mutagenic potency (per m ³) modestly increased in autumn and winter collection periods. Maximum mutagenic potency ~6 rev/ m ³ . Weak association between mutagenic activity and VOC concentration	<u>Fuselli et al.</u> (1995)
Sicily, Italy (1993)	Airborne PM from urban and rural locations, collected on GFFs with high-volume sampler. CX Soxhlet extraction	TA97a, TA98, TA100, YG1021, YG1026, YG1024, YG1029, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Substantial increases in potency (per m ³ equiv) for urban area, relative to rural. Substantial increase in potency on YG1024 with S9, relative to TA98. PAH concentrations far higher in urban area (e.g. 5–25-fold). Maximum potency on TA98 with S9 ~28 rev/m ³	<u>Izzotti et al.</u> (1996)
Athens, Greece (1983)	Monthly PM samples, collected on cellulose filters with high- volume sampler. Hx sonication extraction	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	All 19 samples showed significant positive responses, with little difference between with and without S9. Little seasonal variability. Maximum potency without S9 (per m ³ equiv) ~72.8	<u>Athanasiou et</u> <u>al. (1987)</u>
Athens, Greece (1984)	TSP from 4 urban sites and 1 rural site, collected on GFFs with cascade impactor. BZ Soxhlet extraction	TA98, standard plate incorporation assay, without S9	Highest potency (per m ³) for high-traffic city centre site; lowest for light-traffic industrial area. Correlation between potency and PAH levels. Maximum potency ~4 rev/m ³	<u>Athanasiou et</u> <u>al. (1986)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Athens, Greece (1984)	TSP from 4 urban sites and 1 rural site, collected on GFFs with cascade impactor. Soxhlet extraction with BZ or CX	TA98, standard plate incorporation assay, without S9	Highest potency (per m ³) for city centre locations, and increase in winter relative to summer. Correlation between mutagenic potency without S9 and B[<i>a</i>]P concentration. Maximum potency ~35 rev/m ³ . Largest fraction of mutagenic activity associated with PM < 1.1 μ m. Reduced potency (per m ³) at increased sampling heights	<u>Viras et al.</u> (1990)
Paris, France (1979–1980)	Airborne PM from 2 urban sites, collected on GFFs or Teflon filters. Sequential extractions with Chl and Ac, with agitation, or Chl extraction with agitation and sonication	TA1535, TA1537, TA1538, TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Highest potency (per m ³) on frameshift strains without S9. Maximum potency on TA98 without S9 ~190 rev/m ³ ; with S9 ~40 rev/m ³	<u>Courtois et al.</u> (1981)
Paris, France (1983–1985)	Airborne PM from an urban site, collected on GFFs with high-volume sampler. DCM or Ac sonication extraction	TA1538, TA98, TA1537, TA97, TA1535, TA100, TA102, TA98NR, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per μ g of EOM) on TA98 and TA100 increased with S9 for winter samples; S9 effect much lower or negligible for spring samples. Decrease in activity on TA98NR, relative to TA98. Maximum potency on TA98 without S9 ~60 rev/m ³ ; with S9 ~100 rev/m ³	<u>Courtois et al.</u> (1988)
Paris, France (1980–1981)	Size-fractionated PM from central Paris, collected with cascade impactor. ACN extraction	TA98, TA98NR, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Highest potency (per mg of PM) on with S9, and increase in activity with decreasing PM size; maximum potency for PM < 1.1 μ m. 75% of activity associated with PM < 1.1 μ m, 90% with PM < 2 μ m. Strong decrease in mutagenicity of TA98NR, relative to TA98	<u>Festy et al.</u> (1984)
Paris region, France (1978–1979)	Airborne PM from 5 sites in city centre and suburbs, collected on Teflon filters. Agitation and sonication with Chl, or sequential extraction with Chl and Ac	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Highest potency (per mg of PM) without S9, and substantial elevations in winter months. Close correspondence between mutagenic potency and PAH concentration	<u>Festy (1980)</u>
Dunkirk, France	Airborne PM, collected with cascade impactor. Organic fraction thermally desorbed	TA98, YG1041, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9, suspensions of PM or thermally desorbed PM in DMSO	No significant responses on TA98. Significant positive response to a single concentration of $PM_{2.5}$ on YG1041; higher activity without S9	<u>André et al.</u> (2011)
Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
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Barcelona, Spain (1989– 1990)	Airborne PM from 2 urban locations, collected on GFFs with high-volume sampler. DCM sonication extraction, fraction by GPC	TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) 13–32% higher with S9; similar potency for PM from 2 locations. Maximum potency with S9 ~100 rev/m ³ ; without S9 ~78 rev/m ³ . Potency without S9 corresponds with levels of nitroarenes; activity with S9 corresponds with levels of aromatic ketones and quinones. Seasonal variation in ketones, quinones, and lactones indicate strong contribution of atmospheric transformation processes	<u>Bayona et al.</u> (<u>1994)</u>
Barcelona, Spain (1990)	Airborne PM from urban locations, collected on GFFs with high-volume sampler. DCM sonication extraction, fraction by GPC and subfractionation	TA98, TA98NR, TA98/1,8- DNP ₆ , standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency higher without S9. Several potent fractions reduced on TA98NR and TA98/1,8-DNP ₆ , relative to TA98. Active fractions contain PAHs, nitro-PAHs, aromatic ketones and quinones, and N-heterocyclics. Maximum potency with S9 \sim 45 rev/m ³ ; without S9 \sim 100 rev/m ³	<u>Casellas et al.</u> (1995)
Caserta, Italy	Atmospheric contamination at 5 locations in Caserta affected by different levels of vehicle traffic, in situ 3 wk deployments of semipermeable membrane devices	Reverse mutation of dark mutants of <i>Vibrio fischeri</i> , with and without rat liver S9 (Mutatox test)	Significant positive response with S9 for all sites examined; 3 of 5 positive without S9. No quantitative analyses	<u>Isidori et al.</u> (2003)

^a YG1021: TA98 with plasmid pYG216, NR-overproducing strain; YG1024: TA98 with plasmid pYG219, OAT-overproducing strain; YG1041: TA98 with plasmid pYG233, NR- and OAToverproducing strain; YG1026: TA100 with plasmid pYG216, NR-overproducing strain; YG1029: TA100 with plasmid pYG219, OAT-overproducing strain; YG1042: TA100 with plasmid pYG233, NR- and OAT-overproducing strain.

Ac, acetone; ASE, accelerated solvent extraction; 5,6-BF, 5,6-benzoflavone; B[a]P, benzo[a]pyrene; BZ, benzene; Chl, chlorophyll; CO, carbon monoxide; CX, cyclohexane; DCM, dichloromethane; DEE, diethyl ether; DMSO, dimethyl sulfoxide; EOM, extractable organic matter; equiv, equivalent; GFFs, glass-fibre filters; GPC, gel permeation chromatography; Hx, hexane; MeOH, methanol; NMHC, non-methane hydrocarbons; NO, nitrogen oxide; NO₂, nitrogen dioxide; NR, nitroreductase; OAT, *O*-acetyltransferase; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PCB, polychlorinated biphenyl; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; PUF, polyurethane foam; rev, revertants; SO₂, sulfur dioxide; SVOCs, semivolatile organic compounds; THF, tetrahydrofuran; Tl, toluene; TLC, thin-layer chromatography; TSP, total suspended particles; VOCs, volatile organic compounds; wk, week or weeks.

Supplemental Table S8 Summary of studies that used bacterial mutagenicity assays (e.g. Ames assay) to assess the ability of outdoor air to induce genetic mutations – Asia

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Mumbai, India	Airborne PM (> 0.8 μm) collected on GFFs. DMSO or water extraction using mortar and pestle	TA1537, TA1535, TA1538, TA100, TA98, standard plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Only TA100 results recorded. Highest potency (per μ L of extract) for DMSO extracts with S9. Highest response for site with high population density, traffic, and industrialization	<u>Shenoy & Chaubal</u> (1984)
Agra, India (2010)	PM_{10} and $PM_{2.5}$ from a single site, collected on GFFs. DCM sonication extraction	TA98, TA100, plate incorporation assay, without rat liver S9	Weak responses with modest increase for $\mathrm{PM}_{2.5}$ relative to PM_{10}	<u>Singla et al. (2012)</u>
Tokyo, Japan (1988, 1990)	Airborne PM from an urban site during winter, collected on GFFs with high-volume sampler. DCM sonication extraction	TA98, TA100, YG1024, YG1029, pre-incubation assay and Spiral assay, without S9 activation	Mutagenic activity higher with Spiral assay; enhanced on YG1024 and YG1029 compared with parent strains. Maximum potency on TA98 ~450 rev/m ³ for Spiral and 318 rev/m ³ for pre-incubation	<u>Houk et al. (1992)</u>
Tokyo, Japan (1980–1981)	Airborne PM, collected on GFFs with high-volume sampler. BZ:EtOH (4:1) sonication extraction, acid-base-neutral fractionation	TA98, TA100, pre- incubation assay, with and without PCB-induced rat liver S9	Mutagenic potency (per mg of EOM or m ³) on TA98 and TA100 higher with S9; highest activity for neutral fraction. Potency on TA98 without S9 ~11 rev/m ³ ; with S9 ~12 rev/m ³ . Neutral fraction induced tumours in ICR mice	<u>Sasaki et al. (1987)</u>
Tokyo, Japan (1983)	Airborne PM ₁₀ , collected on GFFs with high-volume sampler. MeOH extraction and extract fractionation	TA98 pre-incubation assay, with and without PCB- induced rat liver S9	Potency (per μ g of EOM) higher without S9 activation. Highest potency for crude MeOH extract and NM fraction	<u>Sakitani & Suzuki</u> <u>(1986)</u>
Tokyo, Japan (1978–1979)	Airborne PM, collected on GFFs with high-volume sampler. NM extraction	TA98, pre-incubation assay, with and without rat liver S9	Mutagenic potency (per m ³) higher with S9; elevated in autumn compared with spring and summer. With S9, daytime higher than night-time, but only for summer and autumn. Maximum potency without S9 ~8 rev/m ³ ; with S9 ~16 rev/m ³	<u>Sakitani &</u> Hayashi (1986)
Tokyo, Japan	Airborne PM, collected on GFFs with high-volume sampler. MeOH extraction, solvent fractionation	TA98, pre-incubation assay, with and without rat liver S9	Mutagenic potency (per µg of EOM) higher without S9; highest activity in NM fraction, compared with CX. No response for MeOH/water fraction	<u>Sakitani & Suzuki</u> <u>(1986)</u>
Tokyo, Japan (1978–1979)	Airborne PM from city centre, collected on GFFs with high- volume sampler. MeOH extraction	TA98, pre-incubation assay, with PCB-induced rat liver S9	Mutagenic potency (per m ³) frequently higher during the day, especially in autumn. Seasonal average potency higher without S9; progressive potency decline from winter to autumn, to summer and spring. Some correlation between mutagenic activity and B[<i>a</i>]P concentration. Maximum potency without S9 ~10 rev/m ³ ; with S9 ~8 rev/m ³	<u>Shimizu et al.</u> (1983)

Geographical location	Test article	Salmonella strains³/assay version	Results	Reference
Tokyo, Japan (1978–1979)	Airborne PM from city centre, collected on GFFs with high- volume sampler. NM extraction	TA98, pre-incubation assay, with PCB-induced rat liver S9	Seasonal average mutagenic potency (per m ³) higher with S9 in spring, autumn, and summer; opposite in winter. Daytime potency with S9 higher than night-time in winter, lower in summer. Progressive decline in potency with S9 from winter to autumn, to summer and spring. Some correlation between mutagenic activity and B[<i>a</i>]P concentration. Maximum potency without S9 ~18 rev/m ³ ; with S9 ~18 rev/m ³	<u>Shimizu et al.</u> (1982)
Tokyo, Japan (1980)	Airborne PM from an urban location, collected on GFFs. BZ:EtOH (3:1) sonication extraction	TA98, TA100, pre- incubation assay, with PCB- induced rat liver S9	Mutagenic potency (per m ³) higher without S9, and higher in colder months (January and October) compared with April and July. Analysis of daily values showed > 10-fold variation from lowest to highest within a given month. Significant correlations between mutagenic activity and PAH levels. Maximum potency on TA98 without S9 ~58 rev/m ³ ; with S9 ~47 rev/m ³	<u>Goto et al. (1982)</u>
Tokyo, Japan (1983–1984)	Airborne PM from urban locations, collected on GFFs. Sonication extraction	TA98, TA100, pre- incubation assay, with PCB- induced rat liver S9	Mutagenic potency (per m ³) frequently higher with S9. At one location, values for winter and autumn exceeded those for spring and summer. At second location, autumn and spring values greater than winter values. Variation attributed to seasonal wind direction. Maximum potency on TA98 without S9 ~70 rev/m ³ ; with S9 ~52 rev/m ³	<u>Ohtani et al.</u> (1985)
Tokyo area, Japan (2005)	Size-fractionated PM samples from Saitama City, collected with cascade impactor. DCM sonication extraction	TA98, YG1024, pre- incubation assay, with and without rat liver S9	Mutagenic potency without S9 (per mg of PM) increased with decreasing PM size; highest potency for PM < 0.1 μ m. Potency (per m ³) highest for size fractions of 0.1–1.0 μ m. Activity enhanced ~10-fold on YG1024 relative to TA98. Five nitro-PAHs accounted for 14–24% of activity of ultrafine PM without S9; ~90% of mutagenic activity associated with PM < 2.1 μ m. Potency (per mg of PM) increased with decreasing PM size. Maximum potency on TA98 without S9 ~13 rev/m ³	<u>Kawanaka et al.</u> (2006, 2008)

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Tokyo area, Japan (2003)	Size-fractionated PM samples from Saitama City, collected with cascade impactor. DCM sonication extraction	TA98, YG1024, pre- incubation assay, with and without rat liver S9	Mutagenic potency (per m ³) higher without S9, and maximum potency corresponded to PM sizes of 0.68–1.2 µm. Potency significantly increased on YG1024 relative to TA98. Similar peak for PAH concentrations (pg per m ³). Approximately 90% of the mutagenic activity associated with PM < 2.1 µm. Potency (per mg of PM) increased with decreasing PM size. Maximum potency on TA98 without S9 ~18 rev/m ³ ; with S9 ~14 rev/m ³	<u>Kawanaka et al.</u> (2004)
Tokyo area, Japan (2005)	Size-fractionated PM samples from Saitama City, collected with cascade impactor. DCM sonication extraction	TA98, YG1024, pre- incubation assay, with and without rat liver S9	Similar mutagenic potency (per m ³) with and without S9; similar results for roadside and suburban locations with marginal increase at roadside location. Maximum potency for particles between 0.3 μ m and 1.2 μ m. Potency (per mg of PM) increased with decreasing PM size. Although ultrafine PM made small contributions to PM mass, it accounted for 11–30% of the mutagenic activity that would be expected to be deposited in alveolar regions. Maximum potency on TA98 without S9 ~10 rev/m ³ ; with S9 ~10 rev/m ³	<u>Kawanaka et al.</u> <u>(2011)</u>
Tokyo area, Japan (2001)	Size-fractionated PM samples from Minato-ku, collected with cascade impactor. DCM sonication extraction	YG1024, microsuspension assay, with and without rat liver S9	Mutagenic potency (per m ³) higher without S9 and maximum potency corresponds to PM sizes of 0.33–1.25 µm. Potency (per mg of PM) increased with decreasing PM size; maximum associated with ultrafine PM < 0.22 µm. Potency higher without S9	<u>Endo et al. (2003)</u>
Tokyo area, Japan (1989–1990)	Size-fractionated PM samples from Setagaya and Itabashi, collected with cascade impactor. Organic extraction	TA98, microsuspension assay, with and without rat liver S9	Mutagenic potency (per m ³) elevated for smaller PM sizes (< 1 μ M). Potency positively correlated with levels of BkF. Dramatic increase in mutagenic potency (per μ g of PM) with decreasing PM size. Total potency on TA98 > 500 rev/m ³ . Potency much higher in winter	<u>Matsumoto et al.</u> (1993)
Tokyo and Okayama, Japan (1991–1992)	Airborne PM from urban locations, collected on quartz filters with high-volume sampler. MeOH sonication extraction, blue cotton purification	TA98, pre-incubation assay, with and without PCB- induced rat liver S9	Potency (per m ³) elevated with S9 for autumn and winter. Some indication of general elevation in potency for autumn and winter compared with summer. Blue cotton extracts generally more potent without S9. Maximum potency for Okayama ~16 rev/m ³ without S9; ~23 rev/ m ³ with S9; for Tokyo ~26 rev/m ³ without S9; ~30 rev/m ³ with S9. Potency of blue cotton extracts almost always lower than that of crude extract	<u>Iwado et al. (1994)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Izu and Kawasaki, Japan (1979)	Airborne PM from 2 locations, collected on GFFs with vacuum sampler. Sequential Soxhlet extraction with MeOH and CX	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Higher mutagenic activity (per mg of PM) without S9; elevated at high-traffic site. Maximum potency on TA98 with S9 ~13 rev/m ³	<u>Sutou et al. (1980)</u>
Õhmuta and Fukuoka, Japan (1974–1980)	Airborne PM samples from 6 industrial and residential locations, collected on GFFs with high-volume sampler. MeOH Soxhlet extraction	TA1535, TA1536, TA1537, TA1538, TA98, TA100, plate incorporation assay, with Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) with S9 activation higher for industrial locations, compared with residential sites. Strongest response on TA98. Maximum potency on TA98 with S9 ~445 rev/m ³	<u>Tokiwa et al.</u> (<u>1977)</u>
Õhmuta and Fukuoka, Japan (1977–1978)	Airborne PM samples from 24 industrial and residential locations, collected on GFFs with high-volume sampler or cascade impactor. MeOH or BZ Soxhlet extraction, detailed fractionation	TA98, TA100, plate incorporation assay, with Aroclor 1254-induced rat liver S9 or rat lung S9	Mutagenic potency (per m ³) at industrial site higher on TA98 compared with TA100; opposite for heavy-traffic site. Maximum potency on TA98 with S9 ~73 rev/m ³ . Fractionation showed highest S9-activated potency for neutrals and acidic compounds. Collection of a large PM sample from a heavily polluted site yielded potency on TA98 with S9 ~444 rev/m ³ . Highest activity in the 0.3–1.0 µm fraction (38% of total)	<u>Tokiwa et al.</u> (<u>1980)</u>
Ōhmuta and Fukuoka, Japan (1974–1980)	Airborne PM samples from several locations, collected on GFFs with high-volume sampler. MeOH Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) frequently elevated with S9. Samples divided into 5 "pollution level" groups based on mutagenic potency on TA98 with S9; minimum of < 2.3 rev/m ³ to maximum of > 116 rev/m ³ . Maximum potency ~200 rev/m ³ . Samples from sites with low levels of NO, NO ₂ , and SO ₂ showed lowest mutagenic activity	<u>Tokiwa et al.</u> (<u>1983)</u>
Fukuoka, Japan	Airborne PM and SVOCs, collected on Teflon-coated filters and XAD-4 resin with high- volume sampler. DCM sonication extraction, detailed fractionation	TA98, plate incorporation assay, without S9 activation	Basic and neutral fractions of SVOCs accounted for 59% and 20%, respectively, of activity without S9. Detected several previously unknown nitro-azabenzo[<i>a</i>]pyrene derivatives	<u>Sera et al. (1994)</u>
Osaka, Japan (1983–1984)	Airborne PM, collected on GFFs with high-volume sampler. BZ:EtOH (3:1) sonication extraction, acid-base-neutral fractionation, HPLC fractionation	TA98, TA100, pre- incubation assay, with and without Aroclor 1254-induced rat liver S9	Higher mutagenic potency (per µg of EOM) with S9; except for subfractions of the neutral fraction. Highest mutagenic activity in moderately polar fraction. High activity in subfractions containing ketones, azarenes, quinones, and PAHs	<u>Matsumoto &</u> Inoue (1987)
Osaka, Japan (1998)	Airborne PM from a suburban site, collected on quartz filters. BZ:EtOH (3:1) sonication extraction	YG1024, pre-incubation assay, with and without rat liver S9	Diurnal variation in potency (per m ³) that followed temporal patterns of NO, CO, and 1-nitropyrene (i.e. high in early morning and late evening). Despite strong correlations, nitroarene concentrations estimated to account for a maximum of 1% of mutagenic activity	<u>Kameda et al.</u> (2004)

Geographical location	Test article	Salmonella strains ^a /assay version	Results	Reference
Several cities, Japan (1986, 1987, 1988)	Airborne PM samples from 6 urban and 2 rural locations, collected on GFFs with high- volume sampler. Sonication or Soxhlet extraction with BZ:EtOH (3:1), TI:EtOH (3:1), or DCM	TA98, TA100, microsuspension and plate incorporation assays, with and without PCB- or PB/5,6-BF-induced rat liver S9	Mutagenic potency (per mg of EOM) higher with S9. Similar results for Aroclor- and PB/5,6-BF-induced S9. Similar results for plate incorporation and microsuspension versions of the assay	<u>Matsushita et al.</u> (1992)
Several cities, Japan (1985, 1988, 1990)	Airborne PM from 5 urban and suburban locations, collected on quartz filters with high-volume sampler. MeOH sonication extraction, blue cotton purification	TA98, TA100, TA98/1,8- DNP ₆ , plate incorporation assay, with and without PCB-induced rat liver S9	Mutagenic activity (per m ³) generally higher with S9, and response for blue cotton extracts enhanced relative to crude extract. Responses reduced on TA98/1,8-DNP ₆ , relative to parent strain	<u>Iwado et al. (1991)</u>
Kawasaki and Sagamiko, Japan (1985, 1988, 1990)	Airborne PM from several urban/industrial and non-urban locations, collected on GFFs with high-volume sampler. BZ Soxhlet extraction, transfer to FBS	TA98, TA100, pre- incubation assay, with and without rat liver S9	Mutagenic potency (per m ³) higher with S9; higher for urban sites. Benzene extract more potent than serum extract. Maximum potency on TA98 with S9 ~19 rev/m ³ ; with S9 ~7 rev/m ³	<u>Ohsawa et al.</u> (1983)
Sagamihara, Japan (1984– 1985)	Airborne PM collected on quartz filters with high-volume sampler. BZ:EtOH (3:1) sonication extraction	TA98, TA100, TA98NR, plate incorporation assay, with and without PCB- induced rat liver S9	Mutagenic potency (per m ³) higher without S9, and elevated in winter compared with summer or spring. Large daily potency variations (i.e. up to 10-fold); low values on Sundays and holidays. Strong potency (per μ g of EOM) reductions on TA98NR relative to TA98. Maximum potency on TA98 with S9 ~52 rev/m ³ ; with S9 ~43 rev/m ³	<u>Takagi et al.</u> (1992)
Kobe, Japan (1975)	Airborne PM collected on GFFs with high-volume sampler. BZ Soxhlet extraction	TA1535, TA1537, TA1538, TA1536, TA98, TA100, with and without PB-induced rat liver S9	Mutagenic potency (per µg of EOM) increased with S9; strong response on TA98. Substantial fraction of activity accounted for by acidic (16%) and neutral (30%) fractions. Strong response from neutral subfractions containing PACs such as oxygenated aromatics	<u>Teranishi et al.</u> (1978)
Kobe, Japan	Airborne PM collected on GFFs with high-volume sampler. BZ:MeOH (4:1) Soxhlet extraction; extract transferred to FBS, saline, or DMSO	TA98, plate incorporation assay, with and without PB/ DBA-induced rat liver S9	Mutagenic activity with S9 for serum solution ~60% of DMSO. Without S9, activity of DMSO and serum solutions about equal	<u>Takeda et al.</u> (1983)
Kobe, Japan (1983)	Airborne PM collected on GFFs with high-volume sampler. BZ:EtOH (3:1) sonication extraction	TA98, plate incorporation assay, with and without with and without PB- induced rat liver S9	Mutagenic potency (per mg of PM) elevated with S9	<u>Takeda &</u> <u>Teranishi (1986)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Kobe, Japan (1992)	Airborne PM from high-traffic site, collected on GFFs with high- volume sampler. BZ:EtOH (3:1) sonication extraction, blue rayon fractions	TA98, TA100, YG1021, YG1024, TA98NR, TA98/1,8-DNP ₆ , with and without PB-induced rat liver S9	Mutagenic potency (per m ³) on TA98 increased with S9; elevated in autumn. Reductions in potency on TA98NR and TA98/1,8-DNP ₆ , relative to TA98; dramatic increases in potency on YG1021 (3–4-fold) and YG1024 (5–10-fold). Positive correlation between potency and NO ₂ ; negative correlation with temperature. Maximum potency on TA98 without S9 ~26 rev/m ³ ; with S9 ~52 rev/m ³	Yamaguchi et al. (1994)
Hyōgo Prefecture, Japan (1978)	TSP collected at 2 urban/ industrial sites and 1 rural site, collected on GFFs. BZ Soxhlet extraction	TA98, plate incorporation assay, with and without PB/ DBA-induced rat liver S9	Mutagenic potency (per m ³) elevated with S9 for industrial area only; potency at industrial site higher than at residential and rural site (lowest). Maximum potency on TA98 without S9 ~14 rev/m ³ ; with S9 ~20 rev/m ³	<u>Takeda et al.</u> (1984)
Kanazawa, Japan (1993)	Airborne PM from downtown area, collected with cascade impactor. BZ:EtOH (3:1) sonication extraction	TA98, YG1024, without S9 activation	Mutagenic potency (per m ³) increased with decreasing PM size; maximum for fraction < 1.1 μ m. Noteworthy contributions from DNPs (< 2%); source of most of mutagenicity unknown. Total mutagenic potency of 5 size fractions ~15 rev/m ³	<u>Hayakawa et al.</u> (1995)
Sapporo, Japan (1974–1992)	Airborne PM from residential area, collected on GFFs with high- volume sampler. DCM sonication extraction	TA98, TA100, pre- incubation assay, with and without Aroclor 1254-induced rat liver S9	Strong seasonal trend in mutagenic potency (per m ³), with peak in winter and trough in summer. Temporal decline in potency with S9 from 1974 to 1992; no such trend without S9. B[<i>a</i>]P showed similar temporal decline. Maximum potency on TA98 without S9 ~35 rev/m ³ (1986); with S9 ~40 rev/m ³ (1974)	<u>Matsumoto et al.</u> (1998)
Yahata, Japan (Kyushu, 1980–1983)	Airborne PM from suburban area, collected on GFFs or quartz filters with high-volume sampler. Organic extraction	TA98, with and without PCB-induced rat liver S9	Mutagenic potency (per m ³) generally higher with S9, particularly in colder months (September to March). Potency values quite variable, with a tendency to higher levels in colder months (i.e. winter). Maximum potency on TA98 without S9 ~7.5 rev/m ³ ; with S9 ~12 rev/m ³ . Positive association between mutagenicity and both atmospheric PAHs and heavy metals	<u>Kodama et al.</u> (1983)
Chiba, Japan (1979–1980)	Airborne PM from residential area, collected on GFFs with high-volume sampler. BZ Soxhlet extraction	TA98, pre-incubation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per μ g of EOM) higher with S9. Higher potency (per m ³) in winter. Maximum potency on TA98 without S9 ~17 rev/m ³ . Activity without S9 correlated with atmospheric B[<i>a</i>]P and NO _x	<u>Fukino et al.</u> (<u>1982)</u>
Tokyo, Japan (1996, 1997)	PM ₁₀ collected from several urban, residential, and industrial sites, collected on quartz filters with high-volume sampler. DCM sonication extraction	TA98, TA100, pre- incubation assay, with and without rat liver S9	Mutagenic potency (per m ³) frequently higher without S9. Potency on TA98 without S9 ~67 rev/m ³ ; with S9 ~33 rev/m ³ . Strong correlation between activity on TA98 with S9 and PAH concentration	<u>Koyano et al.</u> (2002)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Taiwan, China (1987–1989)	Airborne PM from 8 urban and suburban locations, collected on GFFs with high-volume sampler. Ac shaker extraction, fraction by size exclusion	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Potency (per mg of PM) similar for urban and suburban areas, and potency of all samples increased with S9. Concentration of $B[a]P$ not correlated with mutagenic activity. DNPs suspected as significant contributors to outdoor air mutagenicity	<u>Chou & Lee (1990)</u>
Taiwan, China (1989)	Airborne PM, collected on GFFs with high-volume sampler. DCM or Ac shaker extraction, fraction by size exclusion	TA98, TA98NR, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM) higher for Ac extract; higher with S9. Marked reduction in potency on NR- deficient strain. PAHs and nitro-PAHs detected only in Ac extract	<u>Lee et al. (1991)</u>
Taiwan, China (1990–1991)	Airborne PM from 12 urban and suburban locations, collected on GFFs with high-volume sampler. Ac shaker extraction, fraction by size exclusion	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher with S9. Potency higher in autumn (dry season) and substantially reduced during heavy rainfall period. Low wind speed contributed to higher potency. Maximum potency on TA98 without S9 ~13 rev/m ³ ; with S9 ~26 rev/m ³ . Nitroarene levels corresponded with mutagenic activity, and PAH profile indicated important contributions from mobile sources	<u>Lee et al. (1994)</u>
Taiwan, China (1993–1994)	TSP and SVOCs from sites near a petrochemical complex, collected on GFFs and PUFs. Hx:Ac:DCM (50:25:25) Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) of TSP and SVOCs elevated with S9; correlated with concentration of PAHs. Maximum potency of TSP on TA98 without S9 ~60 rev/m ³ ; with S9 ~85 rev/m ³ . Maximum potency of SVOCs on TA98 without S9 ~42 rev/m ³ ; with S9 ~82 rev/m ³	<u>Tsai et al. (1995)</u>
Taiwan, China (1994)	Airborne PM from 7 sites in Taichung City, collected on GFFs with high-volume sampler. Ac shaker extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per m ³) higher with S9; variable across seasons. Maximum potency on TA98 without S9 ~41 rev/m ³ ; with S9 ~50 rev/m ³ . Close correspondence between monthly mean mutagenicity and average concentrations of BghiP, an indicator of mobile-source emissions	<u>Kuo et al. (1998)</u>
Taiwan, China (1999)	TSP from site in Taichung City, collected on GFFs. Ac sonication extraction	TA98, TA100, plate incorporation assay, with and without induced rat liver S9	Mutagenic potency (per m ³) higher with S9; higher in winter months. PAH levels also higher in winter months. Maximum potency on TA98 without S9 ~13 rev/m ³ ; with S9 ~50 rev/m ³	<u>Wu & Fang (2001)</u>
Taiwan, China (1986–1990)	TSP from 9 locations in Taipei area, collected on GFFs with high- volume sampler. DCM Soxhlet extraction, TLC fractionation	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per mg of PM or per m ³) on TA98 with S9 elicited highest response; decreased potency with increase in sampling altitude. Significant increases associated with heavy traffic; decrease in summer. Maximum potency on TA98 with S9 ~24 rev/m ³ . Tunnel sample vielded 130 rev/m ³	<u>Wei et al. (1991)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Shanghai, China (1992–1993)	Airborne PM from 13 urban sites, collected on GFFs with high- volume sampler. DCM sonication extraction, acid-base-neutral fractionation	TA98, TA100, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	No significant responses on TA100. Mutagenic potency on TA98 generally higher without S9, and substantially increased in winter. Substantial temporal and spatial variation in potency (per m ³). Maximum potency on TA98 without S9 ~20 rev/m ³ ; with S9 ~25 rev/m ³ . With S9, high activity in basic and polar fraction; without S9, high activity in numerous fractions, including acidic, basic, neutral, and aromatic	<u>Zhao et al. (2002)</u>
Shanghai, China	Airborne PM from residential and industrial locations affected by coking facility. NM extraction	TA98, TA100, plate incorporation assay, with and without S9	Mutagenic potency (per m ³) higher without S9; elevated in residential and industrial areas when coke oven in operation (10–23-fold above control site). When coke oven not operating, 3-fold decrease in potency. Maximum potency on TA98 without S9 (factory entrance) ~2600 rev/m ³ ; with S9 ~2000 rev/m ³	<u>Yu et al. (1989)</u>
Shanghai, China	TSP from 10 representative areas. NM sonication extraction	TA98, TA100, plate incorporation assay, with and without S9	Mutagenic potency (per m ³) higher without S9; similar results for TA98 and TA100. Maximum potency on TA98 without S9 \sim 260 rev/m ³	<u>Zhu et al. (1991)</u>
Shanghai, China (1988)	TSP from 5 representative areas. NM sonication extraction; extract separated into 5 fractions	TA98, TA100, plate incorporation assay, with and without S9	Mutagenic potency (per m ³) marginally higher without S9. Maximum potency on TA98 without S9 ~205 rev/m ³ . One polar fraction accounted for up to 86% of mutagenic activity. PAH-containing fraction accounted for only 2.6–11% of mutagenicity	<u>Zhu et al. (1990)</u>
Shanghai, China	TSP from 13 representative areas. DCM sonication extraction	TA98, TA100, plate incorporation assay, with and without S9	Positive response for all sites, including control (suburban park). Higher response without S9; higher activity at industrial and commercial sites with heavy vehicle traffic	<u>Zhao et al. (1996)</u>
Shanghai, China	TSP from 4 commercial and suburban areas. DCM sonication extraction; extract separated into 5 fractions	TA98, plate incorporation assay, with and without S9	Mutagenic activity generally higher with S9, and higher in winter compared with summer. Strong responses for fractions containing PAHs and polar aromatics	<u>Zhao & Zhu</u> (1997)
Beijing, China (1981)	TSP from 1 commercial site, 2 residential sites, and control site. BZ Soxhlet extraction	TA98, plate incorporation assay, without S9	Mutagenic potency (per m ³) highest at commercial site, followed by residential site. Negative at control. Maximum potency TA98 ~2.3 rev/m ³	<u>Chen et al. (1982)</u>
Beijing, China (1982)	TSP from 1 commercial site, 2 residential sites, and 1 industrial site. BZ Soxhlet extraction	TA97, TA98, plate incorporation assay, with and without S9	Positive responses on both TA98 and TA97. Mutagenic potency of commercial and residential sites higher with S9	<u>Chen et al. (1983)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Beijing, China (1984)	TSP from downtown and industrial area. BZ:EtOH (3:1) Soxhlet extraction	TA100, TA98, plate incorporation assay, with and without S9	Higher potency (per mg of EOM) in downtown area, relative to site with petrochemical industry. Lower or equivalent potency with S9	<u>Chen et al. (1987)</u>
Beijing, China (1980)	TSP from 7 urban, suburban, and industrial sites. BZ:MeOH (4:1) Soxhlet extraction, fractionation on silica	TA98, plate incorporation assay, with and without S9	Mutagenic potency (per m ³) highest at downtown (urban) site and lowest at suburban site. Higher response with S9. Fractionation showed highest S9-activated response in PAH-containing fractions and highest response without S9 in polar fractions. Maximum potency on TA98 with S9 ~66 rev/m ³	<u>Zhao et al. (1983)</u>
Beijing, China (2005)	PM ₁₀ and PM _{2.5} from urban/ commercial and industrial sites. Chl Soxhlet extraction, fractionation on silica	TA98, plate incorporation assay, with and without S9	Higher mutagenic activity with S9, and higher in industrial area. Winter samples more potent than summer samples for industrial area only	<u>Che et al. (2008)</u>
Several cities in China (1983–1984)	TSP from residential, industrial, and suburban sites in Shenyang, Guangzhou, Xi'an, Beijing, and Shanghai. DCM Soxhlet extraction	TA98, plate incorporation assay, with and without S9	Mutagenic activity higher in winter compared with summer. Potency (per mg of PM) similar across 5 cities. Highest potency (per m ³) in Shenyang in winter. Maximum potency on TA98 ~90 rev/m ³	<u>Li et al. (1985)</u>
Shenyang, China (1990)	TSP from 3 residential, industrial, and commercial sites, plus control. DCM Soxhlet extraction	TA98, TA98NR, TA98/1,8- DNP ₆ , plate incorporation assay, without S9	Significant mutagenicity at all sites, including control. Reduced responses on enzymatically deficient strains without S9 confirm contributions from nitro-PAHs. Similar potency (per mg of PM) across all sites	<u>Kong et al. (1994)</u>
Shenyang, China (1990)	TSP from 3 residential, industrial, and commercial sites, plus control. DCM Soxhlet extraction	TA98, YG1021, YG1024, plate incorporation assay, without S9	Significant mutagenicity at all sites, including control. Enhanced responses on enzymatically enhanced strains without S9 confirm contributions from nitro-PAHs	<u>Kong et al. (1995)</u>
Shenyang, China (2000– 2002)	TSP from 4 urban, industrial, and suburban sites. DCM sonication extraction	TA98, TA100, plate incorporation assay, with and without S9	Positive responses for all sites, and higher mutagenicity with S9. Similar potency (per mg of PM) across sites investigated	<u>Piao et al. (2007)</u>
Guangzhou, China (1991)	TSP. DCM Soxhlet extraction	TA98, plate incorporation assay, without S9	Mutagenic potency (per m ³) higher in February (winter) and lowest in June (summer). Maximum potency on TA98 without S9 ~23 rev/m ³	Qian & Zhang (1997), Qian et al. (1997)
Guangzhou, China (2002)	TSP, collected with cascade impactor. Sonication extraction with DCM	TA98, plate incorporation assay, with and without S9	Mutagenic potency (per mg of PM) increased with decreasing PM size; maximum for PM < 0.5μ m. Higher potency without S9. Potency (per m ³) significantly higher for smaller PM size fraction	<u>Li et al. (2005)</u>

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Lanzhou, China (1982)	TSP from industrial and commercial sites, plus rural control. Soxhlet extraction with BZ and MeOH; extract separated into 5 fractions	TA98, TA100, TA1537, plate incorporation assay, with and without S9	Mutagenicity higher with S9, and highest responses at industrial sites. Significant responses for PAH- containing fraction. Weak response for commercial site; rural site positive in winter only	<u>Tian et al. (1985)</u>
Lanzhou, China (1985)	TSP from urban sites. Soxhlet extraction with BZ and MeOH; extract separated into 5 fractions	TA98, TA100, plate incorporation assay, with and without S9	Mutagenicity much higher at downtown site, relative to suburban site. Highest activity in winter, followed by autumn and spring. Highest mutagenicity with S9 in polar aromatic fraction. Higher response for samples collected at lower elevation	<u>Tian et al. (1992)</u>
Lanzhou, China (1991)	TSP, collected with cascade impactor. Soxhlet extraction with MeOH, Ac, and DCM	TA98, TA100, plate incorporation assay, with and without S9	Mutagenic activity higher without S9, and highest response associated with PM < 1 μm	<u>Yang et al. (1994)</u>
Lanzhou, China	TSP from 5 industrial, urban, and rural sites. DCM Sonication extraction	TA98, TA100, plate incorporation assay, with and without S9	Significant responses for all sites, including suburban control. Higher activity without S9, higher at industrial/ commercial sites. Weak response for rural sample	<u>Ding & Wang</u> (1999)
Chongqing, China	TSP from 5 industrial, residential, and rural sites. BZ Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without S9	Mutagenic activity higher with S9. Similar potency (per μ g of EOM) for all 3 sites. Potency (per m ³) much higher for industrial area. Maximum potency on TA98 with S9 ~200 rev/m ³ ; without S9 ~120 rev/m ³	<u>Lu et al. (1989)</u>
Kunming, China (1986)	TSP from 5 urban and industrial sites. Ac Soxhlet extraction	TA100, plate incorporation assay, with and without S9	Significant positive response at some urban/industrial sites; others failed to elicit a positive response	<u>Hu & Yang (1988)</u>
Jinan, China (1983–1984)	TSP from 3 areas. MeOH Soxhlet extraction	TA1538, TA98, TA100, plate incorporation assay, with and without S9	Similar mutagenic potency (per mg of PM) across 3 sites. Highest mutagenic potency (per m ³) at industrial sites; winter higher than summer	<u>Li et al. (1987)</u>
Harbin, China (1981)	TSP from urban and suburban residential sites. MeOH Soxhlet extraction, solvent fractionation	TA98, TA100, plate incorporation assay, with and without S9	Higher mutagenic activity at urban sites. Maximum potency on TA98 without S9 ~20 rev/m ³	<u>Wang & Zhang</u> (1984)
Daqing, China	TSP from 4 urban, commercial, and industrial sites, plus rural control. MeOH Soxhlet extraction	TA1538, TA98, TA100, plate incorporation assay, with and without S9	Highest mutagenic activity at commercial and industrial sites. Potency (per m ³) approximately 2-fold higher during colder season	<u>Zhang et al.</u> (1991), Wang et al. (1993)
Baotou, China (2004)	PM _{2.5} from an unpolluted site during sandstorm. Sonication extraction with water or DCM, also particle suspensions	TA98, TA100, plate incorporation assay, with and without S9	None of the samples analysed (e.g. extracts or PM suspensions) elicited a significant positive response	<u>Chen & Guo</u> (2007)

Geographical location	Test article	Salmonella strainsª/assay version	Results	Reference
Kowloon, Hong Kong Special Administrative Region, China (1995–1996)	TSP. DCM Soxhlet extraction	TA98, TA100, plate incorporation assay, with and without S9	Mutagenic potency (per m ³) highest in December (winter) and lowest in August (summer). Maximum potency on TA98 without S9 ~45 rev/m ³	<u>Mo et al. (1998)</u>
Riyadh, Saudi Arabia	Airborne PM from roadside location, collected on GFFs with constant-flow sampler. Ac sonication extraction	TA98, TA100, TA102, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Mutagenic potency (per μg of EOM) on TA98 similar with and without S9	<u>al-Khodairy &</u> <u>Hannan (1997)</u>
Riyadh, Saudi Arabia	Airborne PM before, during, and after oil well fires in Kuwait, collected on GFFs. Ac sonication extraction	TA98, plate incorporation assay, with and without Aroclor 1254-induced rat liver S9	Marked increase in potency without S9 (per μ g of EOM) during oil well fires. Potency on TA98 without S9 ~8 rev/m ³ during fires; ~1 rev/m ³ before and after fires	<u>al-Khodairy et al.</u> (1998)
Jeddah, Saudi Arabia (2004)	PM_{10} from 10 urban sites and 1 reference site, collected on GFFs with high-volume sampler. Ac Soxhlet extraction	TA98, plate incorporation assay, with and without PB/5,6-BF-induced rat liver S9	Mutagenic potency (per m ³) increased with S9 activation, and highest at location near refuse incinerator and heavy diesel traffic. Maximum mutagenic potency on TA98 with S9 ~56 rev/m ³ . Potency at residential and reference sites S9 ~2–15 rev/m ³	<u>Elassouli et al.</u> (2007)
Chiang Mai, Thailand (1998–1999)	PM ₁₀ or PM _{2.5} from 5 urban sites, collected on Teflon-impregnated GFFs or Teflon membrane filters with portable samplers. DCM sonication extraction	TA100, plate incorporation assay, with and without rat liver S9	Mutagenic potency (per m ³) elevated at high-traffic sites, and higher with S9. At one site, potency elevated in winter months. Maximum potency on TA100 with S9 ~30 rev/m ³ ; without S9 ~22 rev/m ³	<u>Vinitketkumnuen</u> <u>et al. (2002)</u>

^a YG1021: TA98 with plasmid pYG216, NR-overproducing strain; YG1024: TA98 with plasmid pYG219, OAT-overproducing strain; YG1041: TA98 with plasmid pYG233, NR- and OAToverproducing strain; YG1026: TA100 with plasmid pYG216, NR-overproducing strain; YG1029: TA100 with plasmid pYG219, OAT-overproducing strain; YG1042: TA100 with plasmid pYG233, NR- and OAT-overproducing strain.

Ac, acetone; 5,6-BF, 5,6-benzoflavone; B[*a*]P, benzo[*a*]pyrene; BZ, benzene; Chl, chlorophyll; CO, carbon monoxide; CX, cyclohexane; DBA, dibenz[*a*,*h*]anthracene; DCM, dichloromethane; DMSO, dimethyl sulfoxide; DNPs, dinitropyrenes; EOM, extractable organic matter; EtOH, ethanol; FBS, fetal bovine serum; GFFs, glass-fibre filters; HPLC, high-performance liquid chromatography; MeOH, methanol; NO, nitrogen oxide; NO_s, nitrogen oxides; NR, nitroreductase; OAT, *O*-acetyltransferase; PAC, polycyclic aromatic compounds; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PCB, polychlorinated biphenyl; PM, particulate matter; PM₁₀, particulate matter with particles of aerodynamic diameter < 10 μm; PM_{2.5}, particulate matter with particles of aerodynamic diameter < 2.5 μm; PUF, polyurethane foam; rev, revertants; SVOCs, semivolatile organic compounds; TLC, thin-layer chromatography; TSP, total suspended particles.

Supplemental Table S9 Summary of studies that used bacterial mutagenicity assays (e.g. Ames assay) to assess the ability of outdoor air to induce genetic mutations – Oceania (Australia and New Zealand)

Geographical location	Test article	Salmonella strains ² /assay version	Results	Reference
South Island, New Zealand (2001–2002)	PM ₁₀ from 3 urban and residential locations, collected on quartz filters with high-volume sampler. Sequential Soxhlet extraction with DCM and MeOH	TA98, plate incorporation assay, without rat liver S9	Mutagenic potency (per m ³) substantially higher in winter, and elevated in urban areas. 74% of winter samples elicited positive responses; 25% of summer samples. Maximum potency on TA98 ~11 rev/m ³	<u>Brown et al.</u> (2005)
North Island, New Zealand (2005)	PM_{10} and $PM_{2.5}$ from 2 residential locations, collected on GFFs or quartz filters with high- volume sampler. DCM Soxhlet extraction	TA98, fluctuation assay, without S9 activation	Substantial increases in potency (per m ³) in winter; no significant seasonal difference in potency (per μ g of EOM). Potency higher for PM _{2.5} , compared with PM ₁₀ . Potency significantly correlated with PAH levels. Estimated that wood smoke made larger contribution than mobile sources. Maximum potency on ~30 rev/m ³	<u>Cavanagh et</u> al.(2009)

^a YG1021: TA98 with plasmid pYG216, NR-overproducing strain; YG1024: TA98 with plasmid pYG219, OAT-overproducing strain; YG1041: TA98 with plasmid pYG233, NR- and OAToverproducing strain; YG1026: TA100 with plasmid pYG216, NR-overproducing strain; YG1029: TA100 with plasmid pYG219, OAT-overproducing strain; YG1042: TA100 with plasmid pYG233, NR- and OAT-overproducing strain.

DCM, dichloromethane; EOM, extractable organic matter; GFFs, glass-fibre filters; MeOH, methanol; NR, nitroreductase; OAT, O-acetyltransferase; PAHs, polycyclic aromatic hydrocarbons; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; rev, revertants.

Supplemental Table S10 Summary of studies that used plant assays to assess the ability of outdoor air to induce cytogenetic damage

Geographical location	Sites examined	Test system/exposure	End-point examined	Results obtained	Reference
Perugia, Italy	3 locations: no significant local source, residential heating and traffic, high-traffic urban location	<i>Tradescantia</i> clone 4430, in situ exposures of cuttings for 24 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN (relative to indoor control) for all 3 sites during winter. Weaker but significant increase in MN also observed in March and May	<u>Villarini et al.</u> (2009)
Perugia and Brescia, Italy	5 locations: heavily industrialized district, bypass road, near railway station, car tunnels in Perugia and Brescia	<i>Tradescantia</i> clone 4430, in situ exposures of inflorescences for 24 h. Perugia tunnel exposure for 1–5 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency only for Perugia tunnel 1 h and 5 h exposures. No significant increases in MN at other sites	<u>Monarca et al.</u> (1999)
Caserta, Italy	17 locations in Caserta affected by different levels of vehicle traffic, winter and summer	<i>Tradescantia</i> clone 4430, in situ exposures of inflorescences for 24 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN for all sites in winter. Weaker response in summer, with significant increase at 2 of 17 sites	<u>Isidori et al.</u> (2003)
Stuttgart, Germany	8 locations near a municipal waste incinerator (400 m to 1400 m)	<i>Tradescantia</i> clone 4430, in situ exposures of cuttings for 6–24 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency for 9 h and 24 h exposures. Response dependent on distance from incinerator and wind direction	<u>Fomin &</u> <u>Hafner (1998)</u>
Bratislava, Slovakia	5 locations in an urban/ industrial area (vehicular traffic, municipal incinerator, and several industries)	<i>Tradescantia paludosa</i> clone 03, in situ exposures for 10–62 d	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency, with highest response near agrochemical factory, followed by glass industry and urban traffic. No significant increase near incinerator	<u>Misík et al.</u> (<u>2006)</u>
Bratislava, Slovakia	One location downwind of a municipal incinerator (150 m) and a petrochemical plant	<i>Tradescantia paludosa</i> clone 03, in situ exposures in pots for 10 d	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency for all 12 exposures in summer 1997– 2000; 1 of 9 exposures during summer 2003–2005 elicited a positive response	<u>Misík et al.</u> (2007)
10 European cites	65 urban and rural sites in Barcelona, Valencia, Copenhagen, Dusseldorf, Stuttgart, Edinburgh, Klagenfurt, Lyon, Nancy, and Verona	<i>Tradescantia</i> clone 4430, in situ exposures of cuttings for 30 h, 2–2.5 m above ground	Induction of MN (per 100 tetrads) in pollen mother cells	No relative increases in MN frequency for 85% of the assessments. Significant inter-site differences in MN frequencies within a city only observed for cities with heavy-traffic locations (Stuttgart, Lyon, and Valencia)	<u>Klumpp et al.</u> (<u>2006)</u>
Urban and rural sites in São Paulo state, Brazil	2 urban sites in the São Paulo metropolitan area and 2 rural sites outside São Paulo	<i>Tradescantia pallida</i> (Rose) Hunt. cv. <i>purpurea</i> Boom, sentinel plant monitoring and 5-month in situ exposures of potted specimens	Induction of MN (per 100 tetrads) in pollen mother cells	Significant (2–3-fold) increase in MN frequency at urban sites, relative to rural sites. Rural sites not significantly different from control	<u>Guimarães</u> et al. (2000)

upplemental Table S10 (continued)							
Geographical location	Sites examined	Test system/exposure	End-point examined	Results obtained	Reference		
Córdoba City, Argentina.	3 urban locations: city centre, university campus, and residential.	<i>Tradescantia pallida</i> , 6-month in situ exposures in pots	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency, and significant effect of site. MN frequency at residential site significantly lower than at other sites. Positive correlation between MN frequency and TSP for the city centre site	Carreras et al. (2006, 2009)		
Cities in Poland and Belgium	6 locations in Płock (Poland), 8 locations in Warsaw (Poland), 4 locations near Cour-au-Bois dump site (Belgium), 9 sites near Lubna dump site (Poland)	<i>Tradescantia</i> clone 4430, in situ exposures of cuttings for 6 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increases in MN frequency for urban/industrial sites. Elevated levels at high-traffic sites. Elevated MN frequencies at sites close to landfills	<u>Sadowska</u> <u>et al. (2001)</u>		
Sites in Poland and Belgium	3 locations in Poland near a coal- fired power plant, 3 locations in Belgium near a waste disposal site, 3 locations in Belgium near a closed waste disposal site	<i>Tradescantia</i> clone 4430, in situ exposures of cuttings for 6–8 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increases in MN frequency for all sites near the power plant, and locations at both waste sites, relative to field controls	<u>Sadowska</u> et al. (1994)		
Sites in Belgium	20 locations in Belgium at or near a large landfill	<i>Tradescantia</i> clone 4430, in situ exposures cuttings for 8 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increases in MN frequency at all sites, with highest response, relative to field controls, close to landfill and highways	<u>Sadowska</u> et al. (1999)		
Amazon region, Brazil	5 locations in Tangará da Serra region	<i>Tradescantia pallida</i> , in situ 5-month exposures of potted specimens and monitoring of sentinel specimens	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency, relative to control site (Chapada dos Guimarães) in dry season only. Positive correlation between MN frequency and PM _{2.5} level	<u>Sisenando</u> et al. (2011)		
Minas Gerais state, Brazil	4 urban locations in Uberlândia City, and a control (garden) site	<i>Tradescantia pallida</i> (Rose) Hunt. cv. <i>purpurea</i> Boom, 18 h in situ exposures in "exposure boxes" (pots)	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency relative to control site. Higher levels in winter, and positive correlation between MN frequency and traffic density (vehicles/day)	<u>Pereira et al.</u> (2013)		
Bahia state, Brazil	3 location in Feira de Santana with varying traffic density	<i>Tradescantia pallida</i> , in situ exposures of potted specimens and monitoring of sentinel specimens	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency, correlation between frequency and intensity of vehicular traffic. Active in situ exposures yielded higher MN values relative to passive sentinel exposures	<u>Meireles et al.</u> (2009)		

Geographical location	Sites examined	Test system/exposure	End-point examined	Results obtained	Reference
Rio Grande do Sul state, Brazil	4 locations in the metropolitan area of Porto Alegre	<i>Tradescantia pallida</i> var. <i>purpurea</i> , 24 h in situ exposures in pots	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency at urban site (Estância Velha), relative to rural area and indoor control	<u>Costa &</u> Droste, (2012)
Urban and rural sites in São Paulo state, Brazil	28 sites in São José dos Campos	<i>Tradescantia pallida</i> , in situ exposures in pots for 20 wk	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevations in MN frequency at sites with high traffic. Significant association between MN frequency and human cancer incidence	<u>Mariani et al.</u> (2009)
Urban and rural sites in metropolitan area of São Paulo, Brazil	4 urban locations in the city of Santo André and a reference site	<i>Tradescantia pallida</i> cv. <i>purpurea</i> , in situ exposures in pots, biweekly sample collection for 12 months	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevations in MN frequency at urban sites relative to reference. Highest levels at sites with high traffic density. Weather changes also associated with increases in MN frequency	<u>Savóia et al.</u> (2009)
Varanasi City, India	3 urban sites and 1 reference site	<i>Tradescantia pallida</i> cv. <i>purpurea</i> , in situ exposures in pots for 2, 4, or 6 months	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency, and significant site effect, with highest responses at sites with higher levels of TSP and PAHs	<u>Prajapati</u> <u>& Tripathi</u> (2008)
Žilina City, Slovakia	2 sites in proximity to a landfill (Považský Chlmec) and an industrial complex (Dubeň)	<i>Tradescantia paludosa</i> clone 03, in situ exposures in pots, weekly sample collection for 12 months	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency, with highest levels observed at the industrial area	<u>Solenská et al.</u> (2006)
Bahia state, Brazil	2 areas in the municipality of Senhor do Bonfim	<i>Tradescantia pallida</i> (Rose) Hunt. cv. <i>purpurea</i> Boom, in situ exposures in pots, monthly sampling for 1 year	Induction of MN (per 100 tetrads) in pollen mother cells	Significant increase in MN frequency relative to control site, with highest responses at location with highest traffic density	<u>Andrade Jr.</u> <u>et al. (2008)</u>
São Paulo, Brazil	Downtown São Paulo. Water extracts of PM_{10}	<i>Tradescantia pallida</i> (Rose) Hunt. cv. <i>purpurea</i> , 30 minutes exposure to water extract	Induction of MN (per 100 tetrads) in pollen mother cells	Significant dose-dependent increase in MN at 1.5 and 3.0 equiv mg of PM/L. At highest exposure, 3-fold increase in MN frequency compared with control	<u>Batalha et al.</u> (1999)
Brescia, Italy	2 locations: urban (heavy car traffic), residential (lower traffic)	Tradescantia inflorescences exposed for 24 h to PM_{10} extract in DMSO	Induction of MN (per 100 tetrads) in pollen mother cells	Extract of PM from heavy-traffic site induced significant increase in MN frequency	<u>Monarca et al.</u> (1999)
São Paulo, Brazil	Downtown São Paulo. Water extracts of $PM_{2.5}$ collected during and after a bus strike	<i>Tradescantia pallida</i> cuttings exposed to PM water extracts for 8 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant reduction in MN frequency during bus strike (i.e. for 1 mg of PM equiv/L)	<u>Carvalho-</u> <u>Oliveira et al.</u> (2005)

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Supplemental Table STV (continued)							
Geographical location	Sites examined	Test system/exposure	End-point examined	Results obtained	Reference		
Córdoba City, Argentina	University campus, 24 h TSP sample. DCM sonication extraction	<i>Tradescantia pallida</i> (Rose) Hunt. cv. <i>purpurea</i> Boom, 8 h exposure of cuttings to seasonal composites of TSP extracts (in DMSO)	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency for winter, relative to autumn or summer. However, none of the samples showed significant elevation in MN frequency relative to DMSO control	Carreras et al. (2009, 2013)		
Mato Grosso state, Brazil	PM ₁₀ collected in Tangará da Serra during biomass burning. DCM sonication extraction	<i>Tradescantia pallida</i> cuttings exposed to PM extracts for 8 h	Induction of MN (per 100 tetrads) in pollen mother cells	Significant elevation in MN frequency during the period of most intense biomass burning (i.e. August to October)	<u>de Oliveira</u> <u>Alves et al.</u> (2011)		
Slovenia	2 locations in Slovenia: 1 densely polluted (Zagreb-Žitnjak) and 1 minimally polluted (Velika Gorica)	<i>Allium cepa</i> roots exposed to rainwater or melted snow for 2 d or 5 d	Induction of CAs in root tip cells	Significant increase in CA frequency relative to laboratory control, with highest response for densely polluted site. Early collection times after start of precipitation tended to yield higher levels of CAs	<u>al-Sabti (1989)</u>		
4 regions in Slovenia	8 locations in the Šalek Valley, the Zosavje region, the Upper Meža Valley, and the Ljubljana Basin exposed to power plant, industrial, or urban emissions	Allium cepa L. var. ascalonicum, in situ exposures in pots for 3 months	Induction of CAs in metaphase and anaphase of root tip cells	Some indication of increases in CAs at sites near a major thermal power plant, and an urban site (Ljubljana)	<u>Glasenčnik</u> et al. (2004)		
L'Aquila, Italy	Airborne PM between 2.5 μ m and 10 μ m, and between 0.4 μ m and 2.5 μ m, collected with 8-stage cascade impactor	<i>Daucus carota</i> cell cultures exposed to suspended PM for 2–5 d	Induction of MN	Significant increase in MN frequency for fine and coarse PM after exposure for 5 d. Coarse PM induced a significant response for the longer exposure only. Greater response to fine PM	<u>Poma et al.</u> (2002)		
L'Aquila, Italy	Airborne PM between 2.5 μ m and 10 μ m, and between 0.4 μ m and 2.5 μ m, collected with 8-stage cascade impactor	Zea mays root tip cells exposed to suspended PM for 20 h (SCE) or 48 h (MN)	Induction of MN or SCEs	Significant increase in MN frequency for all samples of fine PM, and 3 of 12 coarse samples. Significant increase in SCE frequency for 11 of 12 fine PM samples and 6 of 12 coarse PM samples	<u>Poma et al.</u> (<u>2002)</u>		

CAs, chromosomal aberrations; d, day or days; DCM, dichloromethane; DMSO, dimethyl sulfoxide; equiv, equivalent; h, hour or hours; MN, micronuclei; PAHs, polycyclic aromatic hydrocarbons; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; SCEs, sister chromatid exchanges; TSP, total suspended particles; wk, week or weeks.

Supplemental Table S11 DNA adducts in experimental systems exposed to extracts of air particles: in vitro studies

Material studied	Experimental system	Method of analysis	Results	Reference
Extracts of air samples from Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria)	Human hepatoma HepG2 cells, human diploid lung fibroblasts HEL, and human monocytic leukaemia cell line THP-1	³² P-postlabelling	Winter samples: Prague > Sofia > Košice. Summer samples: Košice > Sofia > Prague	<u>Sevastyanova et al. (2007)</u>
Extracts of SRM 1649a dust sample	Rat liver epithelial WB-F344 cells	³² P-postlabelling	Crude extracts and non-polar fractions gave rise to DNA adducts; polar fraction did not	<u>Andrysík et al. (2011)</u>
Extracts of air particles from Teplice and Prachatice, Czech Republic	Cultured rat hepatocytes and Chinese hamster lung V79NH cells	³² P-postlabelling	Highest DNA binding activity in fractions containing PAHs and nitro-PAHs	<u>Topinka et al. (2000)</u>
Extracts of air particles from Teplice, Czech Republic	Calf thymus DNA with and without S9 liver fraction	³² P-postlabelling	Highest adduct formation with neutral (aromatic) fraction	<u>Binková et al. (1998)</u>
Extracts of air particles from Teplice and Prachatice, Czech Republic	Calf thymus DNA with and without S9 liver fraction	³² P-postlabelling	Seasonal differences significant (with and without S9). Location difference significant (without S9). DNA adducts were mainly PAH- like	<u>Binková et al. (1999)</u>
Extracts of air particles from Prague city centre and suburban area and Teplice, Czech Republic	Calf thymus DNA with and without S9 liver fraction	³² P-postlabelling	Winter > summer for all 3 locations. Differences between locations not significant	<u>Binková et al. (2003)</u>
Extracts of air particles from Ostrava region and Třeboň, Czech Republic	Calf thymus DNA with and without S9 liver fraction	³² P-postlabelling	DNA adduct-forming activity of Ostrava (industrial) > Ostrava (urban) > Třeboň (rural)	<u>Topinka et al. (2011)</u>

PAH, polycyclic aromatic hydrocarbon; SRM, standard reference mixture.

Reference Study populations Method of analysis; source of protein Results Location Sweden Urban bus drivers (26), suburban bus Immunoassay for PAH-plasma Taxi drivers had significantly higher adduct levels Hemminki drivers (21), taxi drivers (19), controls protein adducts; blood plasma than controls (P < 0.001), but bus drivers did not et al. (1994) (21, hospital workshop workers) Denmark Non-smoking men from rural (n = 29) ELISA of PAH-albumin adducts in In contrast to DNA adducts, which correlated Nielsen et al. and urban (n = 73) areas of Denmark with exposure levels, albumin adducts were non-(1996a) serum significantly elevated in the rural group ELISA of PAH-albumin adducts in Among non-smoking women, adduct levels Denmark Pregnant women resident in a rural Autrup & area (21 smokers, 30 non-smokers), in significantly lower in suburban group than in serum: maternal and cord blood cells Vestergaard suburbs (37 non-smokers), and in the city dwellers (P = 0.0173), but rural dwellers not (1996)city of Aarhus (40 non-smokers) significantly different from city dwellers. Levels in cord blood lower than in maternal blood (P < 0.000) and slightly higher in smokers and rural residents than in non-smokers and suburban and city dwellers Czech 30 women in Teplice (high pollution, ELISA of PAH-albumin adducts in No difference in albumin adducts between the 2 Binková exposed); 30 women in Prachatice (low Republic serum groups et al. (1996) pollution, control) Poland Residents living close to coke oven ELISA of PAH-albumin adducts in Mean level of adducts in rural controls significantly Kure et al. plants at Gliwice (n = 13), Bytom lower than in exposed Silesia residents (summer (1997)serum (n = 23), and Świętochłowice (n = 12)samples) (P < 0.001), but were not correlated with air (Silesia; n = 36) and rural residents of levels of B[*a*]P (stationary sampling) Biała Podlaska (n = 45) Residents of city and suburbs of Munich Germany HPLC and GC-MS of B[a]P-albumin Adduct levels did not correlate with estimated dietary Scherer et al. intake of B[*a*]P. Levels tended to be higher in suburban (n = 69)adducts and B[a]P-haemoglobin (2000)adducts in blood residents (borderline significance for B[a]P-albumin, P = 0.056) Children aged 7 years living in Munich Aromatic amine-haemoglobin Adduct levels were highest in children from Munich, Richter et al. Germany (n = 34), Augsburg (n = 126), and adducts measured by GC-MS in intermediate in children from Augsburg, and lowest (2001)Eichstätt (n = 64) (total populations, blood (4-aminobiphenyl, o-toluidine, in children from Eichstätt. Exposure to ETS did not 1.3 million, 250 000, and 13 000, *m*-toluidine, *p*-toluidine, and significantly increase adduct levels respectively) *o*-anisidine) Adduct levels significantly higher in exposed group Thailand Police officers in Bangkok: traffic police ELISA of PAH-albumin adducts in Ruchirawat (high exposure, n = 44) and office-based (P = 0.001)et al. (2002) serum police (low exposure, n = 45) Italy Newspaper vendors in Milan: high-Immunoaffinity chromatography Differences between high- and low-exposure groups Pastorelli traffic-exposed (n = 30) and low-trafficfor B[*a*]P tetrol from haemoglobin significant for non-smokers, but not for smokers et al. (1996) exposed (n = 23)hydrolysate followed by GC-MS analysis

Supplemental Table S12 Summary of studies on protein adducts in humans

B[a]P, benzo[a]pyrene; ELISA, enzyme-linked immunosorbent assay; ETS, environmental tobacco smoke; GC-MS, gas chromatography-mass spectrometry; HPLC, high-performance liquid chromatography; PAHs, polycyclic aromatic hydrocarbons.

Supplemental Table S13 DNA strand breaks in acellular test system

Particles	Substrate	Extraction	Method	Effect	Reference
PM ₁₀ from Edinburgh, United Kingdom	Phage DNA	Water (sonication)	Supercoil relaxation	Increased generation of SB, which was diminished by treatment with mannitol	<u>Donaldson et</u> al. (1997)
PM ₁₀ from Edinburgh, United Kingdom	Phage DNA	Water (sonication)	Supercoil relaxation	Increased generation of SB, which was diminished by treatment with mannitol or DFO	<u>Gilmour et al.</u> (1996)
PM _{2.5} from Baton Rouge, Louisiana, USA	Phage DNA	PBS (vortexing)	Supercoil relaxation	Increased DNA relaxation, which was decreased by superoxide or catalase treatment	<u>Dellinger et al.</u> (2001)
SRM 1648 and SRM 1649	Phage DNA with or without ascorbate	Baghouse	Supercoil relaxation	Increased level of SB when ascorbate was present. SRM 1649 more potent than SRM 1648	<u>Smith & Aust</u> (1997)
Coal fly ash from a dumping site of a thermal power plant in Aligarh, India	Calf thymus DNA	DMSO (0.5%) or water (sonication)	Alkaline unwinding	Increased generation of SB by DMSO (0.5%) and aqueous extract	<u>Dwivedi et al.</u> (2012)
TSP from an unknown site in London, United Kingdom (collected in 1958)	Plasmid DNA	Water (shaking)	Supercoil relaxation	Concentration-dependent increase in level of SB	<u>Whittaker et</u> <u>al. (2004)</u>
Different size fractions of PM ₁₀ from Leeds, United Kingdom	pBR322	Water (vortexing)	Supercoil relaxation	Small size fractions most potent in generation of SB	<u>Healey et al.</u> (2005)
Fine PM, coarse PM, or PM_{10} from an urban site 2.5 miles from a landfill site	Plasmid DNA	Not reported	Supercoil relaxation	Fine particles more potent than coarse, and $\mathrm{PM}_{\mathrm{10}}$ least potent	<u>Koshy et al.</u> (2009)
Aqueous extract of PM ₁₀ from a traffic site in Leeds, United Kingdom	Plasmid DNA	Water	Supercoil relaxation	Small particles more potent than large particles, which correlated with the level of iron	<u>Lingard et al.</u> (2005)
PM ₁₀ and PM _{2.5} from Barcelona, Spain	Plasmid DNA	Water (sonication)	Supercoil relaxation	PM_{10} more potent than $PM_{2.5}$	<u>Reche et al.</u> (2012)
$\mathrm{PM}_{\mathrm{10}}$ and $\mathrm{PM}_{\mathrm{2.5}}$ from Beijing, China	Plasmid DNA	Water (vortexing)	Supercoil relaxation	$\mathrm{PM}_{\scriptscriptstyle 2.5}$ samples generated more SB than $\mathrm{PM}_{\scriptscriptstyle 10}$ samples	<u>Shao et al.</u> (2006)
Fine and coarse particles from Cardiff, United Kingdom	Phage DNA	Water (vortexing)	Supercoil relaxation	Coarse particles more potent than fine particles	<u>Greenwell et al.</u> (2002)
Particles from Port Talbot, United Kingdom, sampled during different wind directions	Plasmid DNA	Water (vortexing)	Supercoil relaxation	Highest activity on mass basis of samples collected during wind direction from a location with hilly country and motorway. Association between strand scission activity and metal content	<u>Moreno et al.</u> (2004)

Particles	Substrate	Extraction	Method	Effect	Reference
PM _{2.5} from urban or suburban sites in Shanghai, China	Plasmid DNA	Water (vortexing)	Supercoil relaxation	Samples from urban sites most potent. Seasonal variation in potency (winter more potent than summer)	<u>Senlin et al.</u> (2008)
PM _{2.5} and PM ₁₀ samples collected at Beijing, Nankou town, and a clean site, Shisanling Reservoir, China	Plasmid DNA	Water (sonication)	Supercoil relaxation	Extracts of $PM_{2.5}$ and PM_{10} caused SB generation, with TM50 (level causing 50% of SB) ranging from 10 to 1000 µg/mL, dependent on the sites and sampling time. Generally, $PM_{2.5}$ samples more potent than PM_{10} samples. Samples collected during sandstorm less potent than non-sandstorm samples	<u>Shi et al. (2004)</u>
PM ₁₀ samples from Lanzhou city and a suburban area in China during 4 seasons	Plasmid DNA	Water (sonication)	Supercoil relaxation	Average values of TD20 (level causing 20% of SB) were 17, 625, 56, and 260 µg/mL in winter, spring, summer, and autumn, respectively, for PM_{10} suspension solution from Lanzhou city. Water extracts caused slightly lower level of SB with higher TD20. Suburban PM_{10} samples showed higher TD20 than Lanzhou sample. Particles collected during dust storm episodes or after days with rain showed lower SB generation activity with TD20 > 1000 µg/mL. TD20 values were negatively correlated with metal concentration	<u>Xiao et al.</u> (2009)
PM ₁₀ samples collected at 3 sites in Macao Special Administrative Region, China	Plasmid DNA	Water (sonication)	Superoxide relaxation	TD30 values (level causing 30% of SB) were 3, 10, and 20 μ g/mL for whole PM ₁₀ suspension samples from Sun Yat Sen Municipal Park, Avenida de Horta e Costa, and Macao University, respectively. Water extracts showed slightly high TD30 values, with 60, 63, and 80 μ g/mL for the 3 sites, respectively	<u>Shen et al.</u> (2009)

DFO, deferoxamine; DMSO, dimethyl sulfoxide; PBS, phosphate-buffered saline; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; SB, strand breaks; SRM, standard reference mixture.

Supplemental Table S14 Oxidatively damaged DNA in lung tissue of animals

Particles	Animal	Extraction	Dose and duration	Effect	Reference
PM ₁₀ collected during a dust storm from the roof of a building in Incheon, Republic of Korea	Mice	PBS	0.5 mg/mouse twice/wk for 12 wk (total dose, 12 mg/mouse), and killed at 24 h after the last exposure	8-oxodG (immunohistochemistry) in lungs	<u>Hwang et al. (2010)</u>
Intratracheal instillation of SRM 1649	ApoE ^{_/_} mice	NA	0.5 mg/kg at 26 h and 2 h before being killed	Unaltered levels of FPG-sensitive sites (comet) in lung tissue	<u>Vesterdal et al.</u> (2012)
Intratracheal instillation of PM from a town with many wood stoves and a rural area, Denmark	Rats	Mechanical collection from plates	0.64 mg/kg, and killed at 24 h	Unaltered levels of 8-oxodG and etheno adducts (HPLC)	<u>Danielsen et al.</u> (2010)

FPG, formamidopyrimidine DNA glycosylase; h, hour or hours; HPLC, high-performance liquid chromatography; NA, not applicable; 8-oxodG, 8-oxo-7,8-dihydro-2 -deoxyguanosine; PBS, phosphate-buffered saline; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; SRM, standard reference mixture; wk, week or weeks.

Supplemental Table S15 Oxidatively damaged nucleobases in cultured cells							
Particles	Cells	Extraction	Concentration and time	Effect	Reference		
Urban street (TSP), Copenhagen, Denmark	A549	Water (ultrasonication)	2.5–100 μg/mL	Concentration-dependent increase in FPG sites (comet)	<u>Danielsen</u> <u>et al. (2008)</u>		
Particles from a street tunnel in Oslo, Norway, during seasons with or without use of studded tyres	A549 and THP-1	Scraping off the filter	2.5–200 μg/mL for 3 h	Increased FPG-sensitive sites in A549 cells (comet)	<u>Danielsen</u> et al. (2009)		
PM from a town with many wood stoves and a rural area, Denmark	A549 and THP-1	Mechanical collection from plates	2.5–100 μg/mL for 3 h	Concentration-dependent increase in FPG-sensitive sites in A549 and THP-1 cells (comet). Unaltered levels of 8-oxodG and etheno adducts in A549 cells	<u>Danielsen</u> <u>et al. (2011)</u>		
PM ₁₀ from a busy street in Stockholm, Sweden	A549	Water (vortexing or sonication)	10 μg/cm² (8-oxodG) for 4 h	Unaltered 8-oxodG levels (HPLC-ECD)	<u>Karlsson et al.</u> (2005)		
PM _{2.5} or PM ₁₀ from city background site in Milan, Italy	BEAS-2B	Water (ultrasound)	$25~\mu g/cm^2$ for 24 h	Unaltered levels of FPG-sensitive sites (comet)	<u>Gualtieri et al.</u> (2011)		
Aqueous extract of PM _{2.5} from Piedmont, Italy	A549	Water (ultrasound)	15–7 m³ equiv for 24 h	Higher induction of FPG sites (comet) by aqueous extracts from industrial site compared with urban and highway sites	<u>Bonetta et al.</u> (2009)		
PM in different size fractions from urban and rural sites, United Kingdom	A549	Water (sonication)	$100~\mu g/cm^2$ for 24 h	Increased levels of FPG-sensitive sites (comet), with higher dependency on locations than on particle sizes	<u>Wessels et al.</u> (2010)		
Coarse and fine particles from Düsseldorf, Germany	A549	Water (sonication)	$50\mu g/mL$ for 2 h	Coarse and fine PM generated similar extent of 8-oxodG (antibody-based detection)	<u>Shi et al.</u> (2003)		
$\mathrm{PM}_{\mathrm{2.5}}$ from Dunkirk, France	A549	Mechanical collection from plates	6.3–31.6 μg/cm² for 24–72 h	Concentration-dependent increase in level of 8-oxodG (ELISA) at all time points	<u>André et al.</u> (2011)		
PM from Dunkirk, France	L132	Mechanical collection from plates	19–75 μg/mL for 24–72 h	Concentration- and time-dependent increase in level of 8-oxodG (ELISA)	<u>Garçon et al.</u> (2006)		
EOM of PM ₁₀ from Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria)	HepG2	DCM	10–250 μg/mL for 2–48 h	Unaltered levels of FPG-sensitive sites (comet), except EOM of PM_{10} from Kosice in summer	<u>Gábelová</u> <u>et al. (2007)</u>		
SRM 1649, ROFA, Arizona desert dust, or urban air dust from Düsseldorf, Germany	BEAS-2B	NA	400 mg/mL for 2 h	Increased 8-oxodG (HPLC-ECD)	<u>Prahalad et al.</u> (2001)		

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Particles	Cells	Extraction	Concentration and time	Effect	Reference
Coal fly ash from a power plant in Arnhem, Netherlands	RLE	Not specified	40 cm ² /mL or particles/mL for 18 h	Different potency of particles to generate 8-oxodG (HPLC-ECD)	<u>van Maanen</u> <u>et al. (1999)</u>
EOM of PM_{10} from Teplice, Czech Republic	HepG2	DCM	1–50 μg/mL for 24 h	Increased ENDOIII/FPG sites, although not concentration-dependent relationship	<u>Lazarová &</u> <u>Slamenová</u> (2004)
EOM of $PM_{2.5}$ from the traffic area in Suwon, Republic of Korea	BEAS-2B	DCM (sonication)	50 μg/mL	Increased levels of ENDOIII/FPG sites	<u>Oh et al.</u> (2011)
EOM of PM_{10} from an industrial site in France	HepG2	DCM (Soxhlet extractor)	12 μg/mL of B[<i>a</i>]P for 24 h	Increased levels of 8-oxodG (HPLC-MS/MS)	<u>Tarantini</u> <u>et al. (2009)</u>
EOM of PM ₁₀ from Prague, Czech Republic	HepG2 and lung fibroblasts	DCM	1–100 mg/mL for 24–48 h	Increased levels of 8-oxodG (ELISA) in HepG2 cells, but not in lung fibroblasts. Little variation in genotoxicity between different seasons	<u>Hanzalova</u> <u>et al. (2010)</u>
EOM of PM ₁₀ from Kaifaqu district, Dalian, China	HepG2	Sequential extraction in DCM, acetone, and methanol	7.5–30 mg/mL for 1 h	Concentration-dependent increase in 8-oxodG (antibody-based) of samples from Kaifaqu (industrial area)	<u>Jiang et al.</u> (2011)

B[a]P, benzo[a] pyrene; DCM, dichloromethane; ECD, electrochemical detection; ELISA, enzyme-linked immunosorbent assay; ENDOIII, endonuclease III; EOM, extractable organic matter; equiv, equivalent; FPG, formamidopyrimidine DNA glycosylase; h, hour or hours; HPLC, high-performance liquid chromatography; MS, mass spectrometry; NA, not applicable; 8-oxodG, 8-oxo-7,8-dihydro-2'-deoxyguanosine; PM, particulate matter; PM₁₀, particulate matter with particles of aerodynamic diameter < 10 μ m; PM_{2.5}, particulate matter with particles of aerodynamic diameter < 2.5 μ m; ROFA, residual oil fly ash; SRM, standard reference mixture.

Supplemental Table S16 Association between particles and generation of oxidatively damaged nucleobases in acellular test systems

Particles	Substrate, co- oxidant	Extraction	Method	Effect	Reference
Coarse and fine particles from locations in North Rhine-Westphalia, Germany	Calf thymus DNA with H_2O_2	Water (sonication)	8-oxodG (dot blot)	Higher generation of 8-oxodG by PM from Duisburg than from rural location. Coarse and fine particles generated the same level of 8-oxodG	<u>Shi et al.</u> (2006)
Coarse and fine particles from Düsseldorf, Germany	Calf thymus DNA with H_2O_2	Water (sonication)	8-oxodG (dot blot)	Coarse PM generated more 8-oxodG than fine PM when compared at equal mass	<u>Shi et al.</u> (2003)
TSP from a busy street in Copenhagen, Denmark	Calf thymus DNA with H ₂ O ₂	Water	8-oxodG (HPLC)	Concentration-dependent increase, but no difference between PM sampled on different days	<u>Danielsen et</u> <u>al. (2008)</u>
PM_{10} from Helsinki, Finland, collected during spring or winter	Calf thymus DNA with H_2O_2	Water and ethanol (sonication)	8-oxodG (immuno dot blot)	Concentration-dependent increase, without clear seasonal variation	<u>Salonen et</u> <u>al. (2004)</u>
Oil fly ash (from an electrostatic precipitator at a power plant in Niagara, New York, USA) and coal fly ash (from a power plant in USA)	dG or calf thymus DNA	Electrostatic precipitator	8-oxodG (HPLC- ECD)	Oil fly ash increased production of 8-oxodG, whereas coal fly ash did not. Reduced 8-oxodG production by DFO	<u>Prahalad et</u> <u>al. (2000)</u>
SRM 1649, urban air dust from Düsseldorf, Germany, Arizona desert dust, and ROFA	Calf thymus DNA	Baghouse	8-oxodG (HPLC- ECD)	Moderately increased production of 8-oxodG by SRM 1649 and urban air dust. No effect of Arizona desert dust and coal fly ash. Strong effect of ROFA and oil fly ash. Reduced effect by pre-treatment with catalase or DFO for ROFA and oil fly ash, whereas no protection against 8-oxodG generation by SRM 1649 and Düsseldorf particles	<u>Prahalad et</u> <u>al. (2001)</u>
SRM 1649 or its particles after extraction with organic solvents	dG	DCM, hexane, acetone, or DMSO	8-oxodG (HPLC)	Increased 8-oxodG production in water and lower production after extraction of organic material	<u>Karlsson et</u> <u>al. (2004)</u>
PM ₁₀ from an urban street in Stockholm, Sweden	dG with H_2O_2	Water (vortexing or sonication)	8-oxodG (HPLC- ECD)	Increased only in the presence of H_2O_2	<u>Karlsson et</u> <u>al. (2005)</u>
Fine particles from Duisburg, Germany	Calf thymus DNA with H_2O_2	Water (sonication)	8-oxodG (dot blot)	Increased only in the presence of H_2O_2 . Filtrate without particles generated same level of 8-oxodG as suspensions of particles	<u>Knaapen et</u> <u>al. (2002)</u>
TSP, PM_{10} , and $PM_{2.5}$ from Athens, Greece	dG with H_2O_2	Water (sonication)	8-oxodG (HPLC- ECD)	$PM_{2.5}$, PM_{10} , and TSP generated 8-oxodG (8-oxodG level reported in the unusual unit of μ g lesions per 10 ⁶ dG)	<u>Valavanidis</u> et al. (2005)

Particles	Substrate, co- oxidant	Extraction	Method	Effect	Reference
EOM from PM in different size fractions from the Czech Republic	Calf thymus DNA	DCM	8-oxodG (ELISA)	No difference between regions on mass concentration basis or different particle sizes (it is not possible to assess the effect of particles per se because the values in the negative control are not reported)	<u>Rossner et</u> <u>al. (2010)</u>
EOM from PM _{2.5} from different areas of the Czech Republic	Calf thymus DNA	DCM	8-oxodG (ELISA)	No difference between regions on mass concentration basis (it is not possible to assess the effect of particles per se because the values in the negative control are not reported)	<u>Topinka et</u> al. (2011)
EOM from airborne particles in Maastricht, Netherlands	Salmon testis DNA	DCM (Soxhlet)	8-oxodG (HPLC- ECD)	Similar generation of 8-oxodG by PM _{2.5} , PM ₁₀ , and TSP samples. No association between traffic intensity and potency of the extracts	<u>de Kok et al.</u> (2005)
Coal fly ash from a dumping site of a thermal power plant in Aligarh, India	Calf thymus DNA	DMSO (0.5%) or water (sonication)	8-oxodG (antibody)	Increased generation of 8-oxodG by DMSO (0.5%) and aqueous extract	<u>Dwivedi et</u> al. (2012)

DCM, dichloromethane; DFO, deferoxamine; dG, deoxyguanosine; DMSO, dimethyl sulfoxide; ECD, electrochemical detection; ELISA, enzyme-linked immunosorbent assay; EOM, extractable organic matter; H_2O_2 , hydrogen peroxide; HPLC, high-performance liquid chromatography; 8-oxodG, 8-oxo-7,8-dihydro-2'-deoxyguanosine; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; ROFA, residual oil fly ash; SRM, standard reference mixture; TSP, total suspended particles.

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
In vivo					
Netherlands	Airborne PM. MeOH sonication extraction	Relative survival of <i>Escherichia coli</i> 343/753 (<i>uvrB/recA</i>) exposed to PM extracts in vivo via host-mediated assay (Balb/c mouse, intraperitoneal or intratracheal administration)	Induction of DNA damage response	No significant host-mediated induction of DNA damage	<u>Heussen et</u> <u>al. (1990),</u> <u>Heussen &</u> <u>Alink (1990)</u>
In vitro					
Netherlands.	Airborne PM at 2 urban locations and a rural control, collected on quartz filters with high-volume sampler. Ac/ DCM (1:1) Soxtec extraction	Salmonella typhimurium TA1535/pSK1002, incubated for 2 h with extract in DMSO, with and without Aroclor 1254-induced rat liver S9	Induction of DNA damage (SOS) response as induction of <i>umu</i> operon (i.e. <i>umu</i> assay)	Significant dose-related (m ³ equiv/assay) increase in <i>umu</i> induction for both urban sites without S9. Response decreased with S9	<u>Hamers et al.</u> (2000)
Osaka area, Japan	Size-fractionated airborne PM from an urban site, collected with cascade impactor. DCM sonication extraction	Salmonella typhimurium NM3009 (NR- and OAT-enhanced) and NM2009 (OAT- enhanced), incubated for 3 h with extract in DMSO, with and without rat liver S9	Induction of DNA damage (SOS) response as induction of <i>umu</i> operon (i.e. <i>umu</i> assay)	Significant dose-related (m ³ equiv/assay) increase in <i>umu</i> induction on NM3009 without S9 and on NM2009 with S9. Greater response for NM3009, and magnitude of response increased for smaller particle size fractions (e.g. $< 2.1 \mu$ m). Some coarser fractions (e.g. $> 4.7 \mu$ m) failed to induce a significant response	<u>Funasaka et</u> <u>al. (2003)</u>
Osaka area, Japan	Airborne PM at an urban location, collected on quartz filters with high-volume sampler. DCM sonication extraction, fractionation on solid-phase columns	Salmonella typhimurium TA1535/pSK1002, NM3009 (NR- and OAT-enhanced) and NM2009 (OAT- enhanced), incubated for 2 h with extract in DMSO, with and without PB/5,6-BF- induced rat liver S9	Induction of DNA damage (SOS) response as induction of <i>umu</i> operon (i.e. <i>umu</i> assay)	Significant dose-related (m ³ equiv/assay) increase in <i>umu</i> induction. Strongest responses without S9 on NM3009 and with S9 on NM2009. Highest response without S9 associated with 2 fractions containing smaller 2–4-ring PAHs, and larger PAHs and nitrated PAHs	<u>Oda et al.</u> (2004)

Supplemental Table S17 In vivo and in vitro DNA damage response assays to assess the genotoxic activity of outdoor air

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Shanghai, China	TSP collected from 3 locations with variable levels of pollution and 1 reference site. Ac sonication extraction; extract separated into 5 fractions	Salmonella typhimurium TA1535/pSK1002, incubated for 2 h with extract in DMSO, with and without rat liver S9	Induction of DNA damage (SOS) response as induction of <i>umu</i> operon (i.e. <i>umu</i> assay)	Significant induction of DNA damage response for samples from 3 contaminated sites; higher induction at site with highest level of contamination. Higher response without S9	<u>Lu et al.</u> (1990)
Brno, Czech Republic	Airborne PM ₁₀ from an urban/ industrial location, collected with cascade impactor. 6 PM size fractions, DCM extraction with Buchi automated extractor	<i>Escherichia coli</i> PQ37, incubated for 2 h with extract in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related (m ³ equiv/mL) increase in DNA damage response. Higher response without S9, and increase in response induction with decreasing PM size; maximum for < 0.45 μ m. Highest concentration of PAHs associated with finest fraction	<u>Čupr et al.</u> (<u>2013)</u>
Bosnia and Herzegovina	Airborne PM (TSP) and SVOCs from 8 urban locations and 2 reference sites in Sarajevo and Tuzla, collected with high-volume sampler equipped with tandem GFF and PUF. DCM Soxhlet extraction	<i>Escherichia coli</i> PQ37, incubated for 2 h with pooled extracts (i.e. PUF and PM) in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related (m ³ equiv/assay) increase in DNA damage response for all samples. Higher responses without S9, higher levels at more industrialized locations in Tuzla, lowest responses at reference sites. Significant empirical relationships between sample potency, both with and without S9, and PAH concentration (ng/m ³)	<u>Škarek et al.</u> (<u>2007a)</u>
Czech Republic	Airborne PM (PM _{2.5} or TSP) and SVOCs from 2 urban locations and 2 reference sites, collected with high-volume sampler equipped with tandem quartz filter and PUF. DEE/Hx (1:9) Soxhlet extraction	<i>Escherichia coli</i> PQ37, incubated for 2 h with pooled extracts (i.e. PUF and PM) in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related (m ³ equiv/assay) increase in DNA damage response for all pooled TSP/PUF samples without S9; 2 of 4 pooled $PM_{2.5}/PUF$ samples elicited positive responses. With S9, responses marginal or negative	<u>Škarek et al.</u> (<u>2007b)</u>
Paris, France (1983–1985)	Airborne PM from an urban site, collected on GFFs with high-volume sampler. DCM or Ac sonication extraction	<i>Escherichia coli</i> PQ37, incubated for 2 h with 2 replicate extracts in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related (mg of EOM) increase in DNA damage response, without S9 only	<u>Courtois et</u> <u>al. (1988)</u>
Washington, DC, USA	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). DCM Soxhlet extraction	<i>Escherichia coli</i> PQ37, incubated for 2 h with 2 replicate extracts in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Some indication of SOS response induction both with and without S9. Erratic dose–response and high cytotoxicity with no clear, significant induction of DNA damage	<u>Nylund et al.</u> (1992)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Wrocław, Poland (2007)	PM ₁₀ from an urban site collected on sintered glass filters. DCM Soxhlet extraction	<i>Escherichia coli</i> PQ37, incubated for 2 h with 2 replicate extracts in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related (m ³ equiv) increase in DNA damage response, with and without S9. Elevated response without S9 in autumn (relative to summer)	<u>Piekarska</u> (2010)
Wrocław, Poland	Airborne PM from 2 urban sites, collected on glass filters. DCM Soxhlet extraction	<i>Escherichia coli</i> PQ37, incubated for 2 h with 2 replicate extracts in DMSO, with and without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related (m ³ equiv) increase in DNA damage response; higher with S9. Increased genotoxicity in winter compared with summer	<u>Piekarska et</u> <u>al. (2011)</u>
Taiyuan, China (1990)	Size-fractionated airborne PM from 2 urban sites. Extraction with nitric acid or SLF	<i>Escherichia coli</i> PQ35 and PQ37, incubated for 2 h with acid or SLF extracts	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related increase in DNA damage response for acid and SLF extracts for all size fractions. Increased induction of DNA damage response for extracts of smaller size fractions	<u>Lei & Xing</u> (1993), Lei et al. (1993)
Guangzhou, China (1991)	TSP, 12-month monitoring. DCM Soxhlet extraction	<i>Escherichia coli</i> PQ35 and PQ37, incubated for 2 h with extract in DMSO, without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant dose-related increase in DNA damage response for all samples; highest response observed in February and correlated with maximum TSP level. Sample potency affected by meteorological conditions	<u>Qian et al.</u> (1996)
Shanghai, China	TSP samples from 4 locations in industrial Baoshan district. DEE Soxhlet extraction	<i>Escherichia coli</i> PQ35 and PQ37, incubated for 2 h with extract in DMSO, without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Significant increase in DNA damage response for urban location; responses for residential/ suburban sites marginal or equivocal	<u>Lu et al.</u> (1997)
Montreal, Canada (1989)	Snow pack samples from 14 urban locations. DCM liquid– liquid extraction	<i>Escherichia coli</i> PQ35 and PQ37, incubated for 2 h with extract in DMSO, without S9 metabolic activation	Induction of DNA damage (SOS) response (SOS chromotest)	Only 1 sample elicited a positive response without S9; half the samples elicited a positive response with S9. Mean CO and NO ₂ significantly higher at locations that yielded genotoxic samples. Significant association between sample potency and outdoor PM levels	<u>White et al.</u> (1995)

Supplemental	Table S17	(continued)
Supprentation		(continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Netherlands	Airborne PM. MeOH sonication extraction	Differential survival of <i>Escherichia coli</i> 343/753 (<i>uvrB/recA</i>) and 343/765 (<i>uvr+/rec+</i>), exposed to PM extracts in vitro, with and without S9	Induction of DNA damage response	Significant dose-related (m ³ equiv/assay) induction of DNA damage response. Maximum response without S9	<u>Heussen et</u> <u>al. (1990),</u> <u>Heussen &</u> <u>Alink (1990)</u>
Houston, Texas, USA	Airborne TSP or size- fractionated PM, collected with high-volume sampler or cascade impactor. PM collected on GFFs. Soxhlet extraction with BZ:MeOH:DCM (1:1:1)	<i>Escherichia coli</i> UTH8177, overnight incubation with PM extracts	Induction of DNA damage response. Prophage induction assay (i.e. λ phage plaque counts)	No significant response despite strong response on <i>Salmonella</i> reverse mutation assay	<u>Preidecker</u> (<u>1980a)</u>
Several sites, urban Rio de Janeiro, Brazil, and Camden, New Jersey, USA	Airborne PM, collected on GFFs with cascade impactor. Sequential Soxhlet extraction with CX, DCM, and Ac	Escherichia coli WP2 _s λ^+ , overnight incubation with PM extracts	Induction of DNA damage response. Microscreen prophage induction assay (i.e. λ phage plaque counts)	Potency of most samples increased with S9, particularly Camden DCM extract. DCM extract for Rio de Janeiro greater than Ac and CX extracts. Similar potency for Camden and Rio samples.	<u>Miguel et al.</u> (1990)
Flanders, Belgium (2000)	PM_{10} from urban, rural, and industrial sites, collected on GFFs with low-volume sampler. ASE with THF/Hx (20:80)	Salmonella typhimurium TA104 recN2-4, incubation of log-phase cells with PM_{10} extract	Induction of DNA damage (i.e. SOS) response. Recorded as luminescence (Vitotox assay)	Significant induction of DNA damage response for 20 m ³ equiv/mL of urban site with S9. High cytotoxicity and no observable induction of DNA damage response for other samples	<u>Brits et al.</u> (2004)
Flanders, Belgium (2000–2001)	PM_{10} and SVOCs from urban, rural, and industrial sites; PM collected on quartz filters and SVOCs on PUFs with high- volume sampler. Ac Soxhlet extraction	Salmonella typhimurium TA104 recN2-4, incubation of log-phase cells with PM_{10} extract	Induction of DNA damage (i.e. SOS) response. Recorded as luminescence (Vitotox assay)	Significant induction of DNA damage response for PM extracts. Greater potency (per m ³) without S9, and higher potency in winter compared with summer. PUF extracts did not induce a positive response	<u>Du Four et</u> <u>al. (2004)</u>
Flanders, Belgium (2002)	PM ₁₀ and SVOCs from 15 urban, rural, and industrial sites; PM collected on quartz filters and SVOCs on PUFs with high-volume sampler. Ac Soxhlet extraction	Salmonella typhimurium TA104 recN2-4, incubation of cells with PM_{10} and PUF extract with and without S9	Induction of DNA damage (i.e. SOS) response. Recorded as luminescence (Vitotox assay)	Significant induction of DNA damage response for PM extracts. Greater potency (per m ³) without S9, and only 2 of 15 sites yielded positive samples with S9. Only 1 site yielded a PUF extract that elicited a significant positive response	<u>Du Four et</u> <u>al. (2005)</u>

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Taiyuan, China (1991)	TSP samples from a residential area. Nitric acid or SLF sonication extraction	Exposure of human amnion cells for 5 h to acid or SLF extracts	Unscheduled DNA synthesis (incorporation of radio-labelled thymidine)	Significant dose-related increase in DNA damage for 0.5 m ³ equiv of acid extract and 21 m ³ equiv of SLF extract. Lead, zinc, chromium, manganese, and nickel suspected as contributors to DNA damage response	<u>Yuan & Xun</u> (1994)
Taiyuan, China (1987)	Size-fractionated TSP from a residential area. Nitric acid or SLF sonication extraction	Exposure of human amnion cells for 5 h to acid or SLF extracts	Unscheduled DNA synthesis (incorporation of radio-labelled thymidine)	Significant dose-related increase in DNA damage for all acid extracts. Significant DNA damage response for SLF extracts of PM < 2 μ m only. Extracts of smaller size classes induced responses	<u>Yuan et al.</u> (1994)
Shanghai, China	TSP from 13 locations. NM sonication extraction	Exposure of primary hepatocytes from SD rats	Unscheduled DNA synthesis (incorporation of radio-labelled thymidine)	Significant dose-related increases in DNA damage response induced by most TSP extracts. Winter samples induced a stronger response relative to summer samples	<u>Zhao & Zhu</u> (1996)
Shanghai, China	TSP from 4 locations. NM sonication extraction; extract separated into 5 fractions	Exposure of primary hepatocytes from SD rats	Unscheduled DNA synthesis (incorporation of radio-labelled thymidine)	Significant dose-related increases in DNA damage induced by all fractions of all samples, except residential area during summer. Residential area induced the weakest response. Highest potency for PAH-containing fraction; in contrast, <i>Salmonella</i> potency highest for polar fraction	<u>Zhao & Zhu</u> (1997)
Leeds, United Kingdom	Size-fractionated PM from 3 urban sites in Leeds city centre, collected with cascade impactor. Water extraction	Incubation of plasmid DNA with PM extracts for 60 h	Plasmid DNA strand break assay	Significant induction of strand breaks; highest activity associated with smallest size fractions	<u>Lingard et al.</u> (2005)
Novara Province, Italy	In situ exposures at 19 sites, including urban, rural, and industrial locations	AFLPs in <i>Trifolium</i> <i>repens</i> L. after 6 wk exposure	DNA sequence changes measured as alterations in restriction sites	Significant increase in percentage of total polymorphisms at 4–7 sites, depending on season. Elevated ozone at locations with elevated polymorphisms	<u>Piraino et al.</u> (2006)

Ac, acetone; AFLP, amplified fragment length polymorphism; ASE, accelerated solvent extraction; 5,6-BF, 5,6-benzoflavone; BZ, benzene; CO, carbon monoxide; CX, cyclohexane; DCM, dichloromethane; DEE, diethyl ether; DMSO, dimethyl sulfoxide; EOM, extractable organic matter; equiv, equivalent; GFFs, glass-fibre filters; h, hour or hours; Hx, hexane; MeOH, methanol; NO₂, nitrogen dioxide; NR, nitroreductase; OAT, *O*-acetyltransferase; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PM, particulate matter; PM₁₀, particulate matter with particles of aerodynamic diameter < 10 μm; PM_{2.5}, particulate matter with particles of aerodynamic diameter < 2.5 μm; PUF, polyurethane foam; SD, Sprague-Dawley; SLF, surrogate lung fluid; SRM, standard reference mixture; SVOCs, semivolatile organic compounds; THF, tetrahydrofuran; TSP, total suspended particles; wk, week or weeks.

Supplemental Table S18 Changes in gene expression in experimental animals associated with exposure to polluted outdoor air

Animals	Exposure	Assay	Results	Call	Reference
Male SD rats	Intratracheal installation of PM_{10} from Cardiff, United Kingdom, for 3 d	Lung RNA evaluated with Atlas Rat 1.2 macroarrays	Altered expression of genes coding for hormone receptors, growth factors, cytokines, IL-2, chemokines, and ligand-gated ion channels	+	<u>Wise et al.</u> (2006)
Male hypertensive rats	Intratracheal installation of EHC-93 urban particulate for 2–20 h	Lung RNA evaluated with Affymetrix U34A	Altered expression of genes in the oxidative stress, inflammation, transcriptional regulation, and cardiovascular system pathways	+	<u>Kooter et al.</u> (2005)
Female BALB/cJ mice	Inhalation exposure to $380 \ \mu\text{g/m}^3$ for 4 h and 24 h of ultrafine carbon particles generated by an electric spark motor	Lung RNA evaluated with Affymetrix U74Av2 GeneChip	Increased expression of heat shock genes after 4 h, but increased expression of other immunomodulatory proteins after 24 h of inhalation exposure	+	<u>André et al.</u> (2006)
Male C57BL/6 mice	Inhalation exposure to < 2.5 μm CAPs from an urban area near San Joaquin Valley, California, USA, for 6 h/d, 5 d/wk, for 2 wk. Winter CAPs, 39.01 μg/m ³ ; summer CAPs, 21.7 μg/m ³	Lung RNA evaluated by qRT-PCR (TaqMan, Applied Biosystems)	Winter CAPs increased expression of <i>IL-2</i> , <i>MIP-</i> <i>1α</i> , <i>TNFα</i> , <i>CYP1a1</i> , <i>ICAM-1</i> , and <i>Nox-2</i>	+	<u>Tablin et al.</u> (2012)
Male C57BL6 mice	Inhalation exposure in Craeybeckx tunnel in Antwerp, Belgium, for 5 d; 55.1 μ g/m ³ of PM _{2.5} ; EC = 13.9 μ g/m ³	Olfactory bulb and hippocampus tissue from brain evaluated by RT-PCR	Increased expression of inflammatory response (<i>COX2</i> , <i>NOS2</i> , <i>NOS3</i> , and <i>NFE2L2</i>) in the hippocampus, and decreased expression of <i>IL-1</i> α , <i>COX2</i> , <i>NFE2L2</i> , <i>IL-6</i> , and <i>BDNF</i> in the olfactory bulb	+	<u>Bos et al.</u> (2012)
Male Brown Norway rats	Rats first given ovalbumin by intranasal installation for 3 d, then exposed to 1–2.5 μ m CAPs from Grand Rapids, Michigan, USA, for 8 h/d for 13 d at 493 μ g/m ³	Lung RNA evaluated with Affymetrix R230 2.0 whole-genome arrays	Altered expression of genes in the inflammation and immune-response pathways	+	<u>Heidenfelder</u> et al. (2009)
Male C57BL/CBA mice	Inhalation exposure in situ near a busy highway and 2 steel mills in Hamilton, Ontario, Canada	Lung RNA evaluated with Agilent G4121B microarrays	Altered expression of genes in lipid droplet synthesis and antioxidant defence	+	<u>Rowan-</u> <u>Carroll et al.</u> (2013)
Male F344 rats	Inhalation exposure for 5 h/d, 4 d/wk, up to 10 months to CAPs from Riverside, California, USA. Mass (μ g/m ³) was 63 (UFP), 149 (FP), and 58 (CP)	Brain RNA evaluated with Affymetrix GeneChip Rat Genome 230 microarray	Increased expression of <i>Arc</i> up to 3-month exposure to CP, then decreased. Increased expression of <i>Rac1</i> after 10-month exposure to CP	+	<u>Ljubimova et</u> al. (2013)
Male C57Bl/6 mice	Inhalation exposure to $PM_{2.5}$ CAPs from unspecified urban area for 8 h/d for 9 wk	Lung RNA evaluated by RT-PCR	Increased expression of DNMT1	+	<u>Soberanes et</u> al. (2012)
Male C57BL/6 mice	Inhalation exposure to PM _{2.5} CAPs from Columbus, Ohio, USA, for 6 h/d, 5 d/wk, for 10 months	RNA from epididymal mouse adipose tissues evaluated by RT-PCR	Increased ERAD and RIDD	+	<u>Mendez et al.</u> (2013)

CAPs, concentrated ambient particles; CP, coarse particles; d, day or days; FP, fine particles; h, hour; IL-2, interleukin-2; PCR, polymerase chain reaction; PM_{10} , particulate matter with particles of aerodynamic diameter < 2.5 μ m; SD, Sprague-Dawley; UFP, ultrafine particles; wk, week.

Supplemental Table S19 Changes in gene expression in vitro induced by polluted outdoor air

Cells	Exposure	Assay	Results	Call	Reference
Human breast cancer cell line MCF7	Extract of urban dust particulate SRM 1649a + B[<i>a</i>]P	Affymetrix U133A GeneChip arrays	Increased expression of <i>CYP1A1</i> and <i>CYP1B1</i> , as well as genes in DNA repair, tissue growth factor, and oncogene pathways	+	<u>Mahadevan et</u> al. (2005)
Primary mouse alveolar type II cells	PM _{2.5} NIST SRM 1649 standard	RT-qPCR	Increased DNMT1	+	<u>Soberanes et al.</u> (2012)
Human lung epithelial cell line L132	PM _{2.5} from Dunkirk, France	RT-qPCR	Increased apoptosis genes: <i>TNFα</i> , <i>caspase-3</i> , -8, -9	+	<u>Dagher et al.</u> (2006)
Human lung cell line A549	PM _{2.5} from Dunkirk, France	RT-qPCR	Increased <i>CYP1A1</i> , <i>CYP2E1</i> , <i>CYP2F1</i> , <i>NQO1</i> , and <i>GST-π1</i>	+	<u>Billet et al.</u> (2007)
Human AMs	PM _{2.5} from Dunkirk, France	RT-qPCR	Increased <i>CYP1A1</i> , <i>CYP2E1</i> , <i>NQO1</i> , and <i>GST-π1/μ3</i>	+	Saint-Georges et al. (2008)
Human AMs and human lung cell line L132	PM _{2.5} from Dunkirk, France	RT-qPCR	In AMs, AMs co-cultured with L132 cells, and L132 cells, PM increased expression of <i>CYP1A1</i> , <i>CYP2E1</i> , <i>NQO1</i> , <i>GST</i> - π 1, and/or <i>GST</i> - μ 3. In L132 cells co-cultured with AMs, no changes in gene expression	+ -	<u>Abbas et al.</u> (2009)
Human lung cell line BEAS- 2B	PM _{2.5-3.0} from Dunkirk, France	RT-qPCR	Increased CYP1A1, CYP1B1, IL-6, and IL-8	+	<u>Dergham et al.</u> (2012)
Human monocyte- macrophage line U937	PM_{10} from Rome, Italy (direct exposure)	Clontech Atlas Human Toxicology 1,2 cDNA Expression Array	Changes in 87 of 1176 genes; 9 were associated with lung cancer. Increased DNA repair and apoptosis genes, such as <i>ERCC1</i> , <i>TDG</i> , <i>DAD1</i> , and <i>MCL1</i>	+	<u>Bastonini et al.</u> (2011)
Human airway epithelial cells	Coarse, fine, and ultrafine PM from Chapel Hill, North Carolina, US	Affymetrix HGU133A microarray	Changes in expression of NRF2-mediated oxidative- stress response genes, cell-cycle genes, DNA damage checkpoint genes, and polo kinase genes	+	<u>Huang et al.</u> (2011)
Human lung cell line HEL12469	EOM from PM _{2.5} from 4 cities in Czech Republic	Illumina Human- HT12v3 Expression Bead Chips	Increased <i>CYP1B1</i> ; decreased <i>ABC Transporters</i> , <i>Wnt</i> , <i>TGF-</i> β , steroid biosynthesis, glycerolipid metabolism	+	<u>Líbalová et al.</u> (2012)
Normal human tracheobronchial epithelium 3-D cell constructs	PM from Swansea, United Kingdom	RT-PCR and Biorad Human Stress and Toxicity Pathway Finder PCR Array	Increased <i>CYP1A1</i> ; decreased <i>MT2A</i> , <i>NFKB1</i> , <i>RAD50</i> , <i>UNG</i> , <i>ANXA5</i> , <i>BCL2L1</i> , <i>MUC1</i> , and heat shock genes	+	<u>Hoogendoorn et</u> <u>al. (2012)</u>

AMs, alveolar macrophages; B[a]P, benzo[a]pyrene; EOM, extractable organic matter; PCR, polymerase chain reaction; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; STM, standard reference mixture.

Supplemental Table S20 Lur	g inflammation in anim	hals after exposure to	air pollution particles
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Particles	Species	Extraction	Exposure time and dose	Effect	Reference
Inhalation of CAPs from Bilthoven, Netherlands	Spontaneously hypertensive rats and ozone-pre- exposed rats	NA	1.2 mg/m ³ for 6 h, and killed 2 d later	Increased PMNs in BALF. Unaltered levels of IL- 6, MIP2, and TNF	<u>Cassee et al.</u> (2005)
Inhalation of CAPs from Boston, Massachusetts, USA	SD rats (normal or SO ₂ -pre- treated rats)	NA	500 μg/m³, 5 h/d for 3 consecutive days, and killed at 24 h after the last exposure	Increased number of PMNs in BALF	<u>Clarke et al.</u> (1999)
Inhalation of CAPs from Boston, Massachusetts, USA	Young and old F344 rats	NA	100 μ g/m ³ , 5 h/d for 3 consecutive days, and killed at 24 h after the last exposure	Substantially higher pulmonary inflammation (total cells and PMNs in young compared with old rats)	<u>Clarke et al.</u> (2000a)
Inhalation of CAPs from Boston, Massachusetts, USA	Dogs	NA	200 µg/m ³ , 6 h/d for 3 consecutive days, and killed at 24 h after the last exposure	Statistically non-significant percentage of PMNs in BALF	<u>Clarke et al.</u> (2000b)
Inhalation of CAPs from Tuxedo, New York, USA	Rats	NA	110–350 μg/m ³ for 3 h, and killed after 3 h or 24 h	Increased number of neutrophils in BALF after 3 h, but not 24 h	<u>Gordon et al.</u> (1998)
Inhalation of CAPs from Boston, Massachusetts, USA, by inhalation	SD rats	NA	1.1 mg/m ³ for 5 h, and killed at 24 h after the exposure	Increased levels of PMNs in BALF. NAC pre- treatment inhibited pulmonary inflammation	<u>Rhoden et al.</u> (2004)
Inhalation of CAPs from Boston, Massachusetts, USA	SD rats (normal or pre-exposed to SO_2 to develop chronic bronchitis)	NA	255 μg/m³, 5 h/d for 3 consecutive days, and killed at 24 h after the last exposure	Increased inflammation (PMNs) in both the normal and chronic bronchitis rats	<u>Saldiva et al.</u> (2002)
Inhalation of CAPs from Manhattan, New York, USA	C3H and C57/BL6 mice	NA	300 μ g/m ³ for 6 h, and killed at 24 h after the exposure	Increased expression of <i>IL-6</i> , <i>TNF</i> , <i>TGF</i> , <i>IFN-y</i> , and <i>MIF</i> in lung tissue	<u>Shukla et al.</u> (2000)
Inhalation of PM from Porto Alegre, Brazil	Wistar rats	NA	110–140 μ g/m ³ for 6, 20, or 4 × 5 h, and killed at 24 h after the exposure	Increased number of leukocytes in BALF of rats exposed for 20 h, and unaltered response in rats after 6 h. Intermittent $(4 \times 5 h \text{ on } 4 d)$ did not generate inflammation	<u>Pereira et al.</u> (2007)
Inhalation of CAPs from Grand Rapids, Michigan, USA	Brown Norway rats	NA	493 μg/m³, 8 h/d for 13 d, and killed 24 h after the last exposure	Unaltered total or differential cell count (including PMNs) in BALF	<u>Heidenfelder et</u> <u>al. (2009)</u>
Inhalation of CAPs from Chapel Hill, North Carolina, USA, or ROFA	SD rats with or without bronchitis	NA	$475-907 \ \mu g/m^3$ (or $1 \ m g/m^3$ for ROFA), 6 h/d for 2–3 d, and killed immediately after the last exposure	Unaltered total or neutrophils in BALF of healthy rats, whereas rats with bronchitis had increased influx of neutrophils after exposure to CAPs and ROFA	<u>Kodavanti et al.</u> (2000)

Particles	Species	Extraction	Exposure time and dose	Effect	Reference
Inhalation of CAPs from a city background location in Bilthoven and a freeway tunnel, Netherlands	Spontaneously hypertensive rats	NA	399–3612 μg/m ³ , 6 h/d for 2 d, and killed 18 h after the last exposure	Unaltered cell count, MIP2, and TNF in BALF	<u>Kooter et al.</u> (2006)
Inhalation of EHC93	F344 rats	NA	57 μg/m³ for 4 h, and killed 33 h later	Increased number of neutrophils in air space and tissue	<u>Adamson et al.</u> <u>(1999b)</u>
Inhalation or intratracheal instillation of ROFA	SD rats	Saline	$12~mg/m^3$ for 6 h or 110 $\mu g/rat$ for 24–94 h	Highest level of neutrophils in BALF at 24 h. Same level of inflammation by intratracheal instillation and inhalation	<u>Costa et al.</u> (2006)
Intratracheal instillation of EHC93	Spontaneously hypertensive rats	Saline	10 mg/kg, and killed at 4 h or 24 h	Increased percentage of PMNs in BALF	<u>Bagate et al.</u> (2004)
Intratracheal instillation of PM from a motorway tunnel or EHC93	Spontaneously hypertensive rats	Water, ethanol, and methanol (sonication)	3 mg/kg or 10 mg/kg	Dose-dependent increase in the number of neutrophils in BALF at 24 h (lower levels at 48 h). Increased levels of IL-6, TNF, and MIP2 in BALF (protein)	<u>Gerlofs-Nijland</u> et al. (2005)
Intratracheal instillation of EHC93	Wistar rats	NA	5 mg/rat, and killed at day 2–7 after the exposure	Increased number of cells in BALF at day 2, with decreased levels at day 4 and 7 after exposure. Increased expression of <i>MIP2</i> , <i>TNF</i> , and <i>Inos</i>	<u>Ulrich et al.</u> (2002)
Intratracheal instillation of EHC93	Swiss mice	NA	1 mg/mouse, and killed at 72 h–8 wk	Increased PMNs in BALF. Water-soluble fraction more inflammogenic than insoluble fraction. Inflammogenicity was mainly explained by the content of zinc	<u>Adamson et al.</u> (1999a, 2000)
ROFA	CD1 mice and transgenic mice overexpressing SOD	NA	50 μg/mouse, and killed at 24 h	Higher levels of PMNs, TNF, and MIP2 in BALF from CD1 mice compared with mice with overexpression of SOD	<u>Ghio et al.</u> (2002)
Intratracheal instillation of SRM 1648	C57BL/6	PBS	1.6 mg/lung, and killed at 20 h	Increased content of IL-6, TNF, and MIP2 in BALF (protein)	<u>Becher et al.</u> (2007)
Intratracheal instillation of fine and coarse particles from 4 cities in Europe, collected in different seasons, and EHC93	Wistar rats	Saline (stirring)	2.5 mg/rat, and killed at 24 h	Fine particles more potent than coarse particles. Influx of neutrophils associated with season. TNF and MIP2 responses dependent on both city and season	<u>Halatek et al.</u> (2011)

Particles	Species	Extraction	Exposure time and dose	Effect	Reference
Fine and coarse particles from cities in Europe	Spontaneously hypertensive rats	Saline	3 or 10 mg/kg, and killed at 24 h	Increased cell count, TNF, and MIP2 in BALF. Tendency towards a more pronounced inflammation response by coarse particles than fine particles. Tendency that particles from sites with high intensity of traffic were most inflammogenic	<u>Gerlofs-Nijland</u> <u>et al. (2007)</u>
Intratracheal instillation of coarse, fine, and ultrafine particles from 6 different cities in Europe	C57B1/6J mice	Methanol	1–10 mg/kg for 4, 12, or 24 h	Dose-dependent increase for coarse particles, which showed stronger effect compared with fine and ultrafine particles on mass basis. Same tendency for TNF, IL-6, and KC in BALF	<u>Happo et al.</u> (2007)
Intratracheal aerosolization of PM_{10} or $PM_{2.5}$ from Milan, Italy, collected during summer or winter at a heavy-traffic site	Balb/c mice	Water (sonication)	100 μg/mouse, and killed at 3 h, 24 h, or 1 wk	PM_{10} samples slightly more potent than $PM_{2.5}$ samples, and winter samples slightly more potent than summer samples (percentage of PMNs at 24 h). $PM_{2.5}$ from summer most potent at 3 h and unaltered effect at 1 wk. TNF (3 h) highest for PM_{10} , and summer samples more potent than winter samples	<u>Farina et al.</u> (2011)
Fine and coarse particles from Duisburg (industrial area) and Borken (rural area) in Germany	Wistar rats	Water (sonication)	0.32 mg/rat, and killed at 18 h	Coarse particles increased number of neutrophils in BALF. Only coarse particles from Borken increased level of TNF and MIP2 in BALF. Strong association between inflammatory response and endotoxin level	<u>Schins et al.</u> (2004)
Intratracheal instillation of $PM_{2.5}$ or PM_{10} from locations near to or far away from traffic in Beijing, China	Wistar rats	Water (sonication)	7.5 mg/kg once/d for 14 d, and killed at 24 h after the last instillation	Higher levels of TNF, IL-6, and IL-1 (protein) in lung homogenate $PM_{2.5}$ generated higher levels of inflammation than PM_{10} . Particles collected closest to traffic generated highest levels of inflammation	<u>Zhang et al.</u> (2011)
Dust storm particles from Shapotou, China, or Arizona, USA	ICR mice	Saline	0.1 mg/mouse 4 times over 8 wk, and killed at 24 h after the last exposure	Increased number of cells in BALF (predominantly macrophages). Increased TNF and IFN-γ (protein); unaltered levels of IL-5, IL- 6, IL-12, and IL-13 in BALF	<u>Ichinose et al.</u> (2008)
Fine particles from World Trade Center dust, Mt Saint Helens dust, ROFA, and SRM 1649 by oropharyngeal aspiration	CD-1 mice	Water	32–100 μg/mouse, and killed at 24 h	Increased neutrophils in BALF by fine particles. ROFA (and SRM 1649) more potent than World Trade Center fine particles	<u>Gavett et al.</u> (2003)
Intratracheal instillation of ROFA	SD rats	Saline	500 μg/rat, and killed at 3 h or 24 h	Increased number of neutrophils in BALF (24 h) and IL-6, TNF, CCL2, and IL-1 β (3 h) gene expression, whereas MIP2 was unaltered	<u>Roberts et al.</u> (2003)
Supplemental Table S20 (continued)

Particles	Species	Extraction	Exposure time and dose	Effect	Reference
Intranasal instillation of PM from a heavy-traffic location in Buenos Aires, Argentina, or ROFA	Balb/c mice	PBS (sonication)	0.17 mg/kg 3 times/d on days 1, 4, and 7, and killed 1 h after the last exposure	Increased pulmonary inflammation (histology and neutrophils in BALF)	<u>Martin et al.</u> (2007), Martin <u>et al. (2010)</u>
Intranasal instillation of PM _{2.5} from São Paulo, Brazil	Balb/c mice	Water (sonication)	5–15 μg/mouse, and killed at 24 h	Increased number of neutrophils, macrophages, IL-6, and TNF in lung tissue sections	<u>Riva et al.</u> (2011)
Intranasal instillation of CAPs from Boston, Massachusetts, USA	Balb/c mice with or without IFN-γ pre-treatment	Saline (sonication)	50 μg/mouse, and killed at 24 h	Increased number of PMNs in BALF. Macrophages and PMNs had increased ex vivo production of ROS (DCFH assay)	<u>Sigaud et al.</u> (2007)
Intratracheal instillation of aqueous extract or insoluble fraction of TSP from Provo, Utah, USA	SD rats	Water (agitation)	100–1000 μg/rat, and killed 24 h after the exposure	Dose-dependent increase in neutrophils in BALF. Lower response with soluble fraction compared with insoluble fraction	<u>Ghio et al.</u> (1999)
Intratracheal instillation of coarse, fine, and ultrafine particles from Chapel Hill, North Carolina, USA	CD-1 mice	Collected on an electrostatic precipitator	10–100 μg/mouse, and killed at 18 h	Increased influx of neutrophils (dose-dependent for fine and ultrafine particles). Increased levels of TNF and IL-6 in BALF. Decreased inflammation by treatment with DMTU	<u>Dick et al.</u> (2003)

BALF, bronchoalveolar lavage fluid; CAPs, concentrated ambient particles; d, day or days; DCFH, 2',7'-dichlorodihydrofluorescein; DMTU, dimethylthiourea; h, hour or hours; IFN- γ , interferon-gamma; IL-6, interleukin-6; NA, not applicable; NAC, *N*-acetylcysteine; PBS, phosphate-buffered saline; PM, particulate matter; PM₁₀, particulate matter with particles of aerodynamic diameter < 10 µm; PM_{2.5}, particulate matter with particles of aerodynamic diameter < 2.5 µm; PMNs, polymorphonuclear leukocytes; ROFA, residual oil fly ash; ROS, reactive oxygen species; SD, Sprague-Dawley; SRM, standard reference mixture; TNF, tumour necrosis factor; wk, week or weeks.

Particles	Cells	Extraction	Exposure conditions	Effect	Reference
PM _{2.5} from Porte d'Auteuil, France	Human nasal epithelial cells	Water (sonication)	10-80 μg/cm ² for 24 h	Concentration-dependent increase in release of GM-CSF, IL-6, and IL-8. Weak and inconsistent increase in TNF (protein)	<u>Auger et al.</u> (2006)
PM _{2.5} from a school playground in Vitry-sur- Seine, France, or SRM 1648	Human bronchial epithelial cells (16- HBE)	Sonication	10–30 µg/cm² for 4 h	Concentration-dependent increase in release of GM-CSF (protein)	<u>Baulig et al.</u> (2003)
SRM 1648	Alveolar type 2 cells or macrophages	NA	20 μg/cm² (or 200 μg/mL) for 20 h	Increased release of IL-6 and MIP2 (protein)	<u>Becher et al.</u> (2007)
PM ₁₀ from a street in London, United Kingdom	J774 macrophages	PBS (vortexing)	5–100 μg/mL for 4 h	Concentration-dependent increase in TNF secretion (protein)	<u>Brown et al.</u> (2004)
PM ₁₀ from a street in London, United Kingdom	Mononuclear blood cells	PBS (vortexing)	10 μ g/mL for 4 h	Increased TNF (protein) secretion	<u>Brown et al.</u> (2007)
EHC93	Human alveolar and bronchial epithelial cells	NA	100 μg/mL for 24 h	Increased secretion of GM-CSF, IL-6, IL-8, IL-1 β , and TNF (protein and mRNA) in macrophages, whereas there was less effect in bronchial epithelial cells	<u>Fujii et al.</u> (2001, 2002)
PM from Dunkirk, France	L132	Mechanical collection from plates	19 μg/mL and 75 μg/mL for 24–72 h	Increased TNF (protein), iNOS activity, and NO production (Griess)	<u>Garçon et al.</u> (2006)
PM _{2.5} from Helsinki, Finland, EHC93, or SRM 1649	RAW 264.7	Methanol or water (sonication)	150 μg/mL for 12 h or 24 h	EHC93 was more inflammogenic than SRM 1649. PM _{2.5} was the least inflammogenic based on TNF, IL-1, IL-6 (protein), and NO (Griess). Water and methanol extraction generated the same effect	<u>Jalava et al.</u> (2005)
PM ₁₀ from a busy street in Stockholm, Sweden.	A549	Water (sonication)	$40 \ \mu g/cm^2$ for $4 \ h$	Increased levels of IL-6, IL-8, and TNF (protein)	<u>Karlsson et al.</u> (2006)
SRM 1648	RAW 264.7	NA	62.5 μg/cm² for 4 h	Increased PGE ₂ release (protein)	<u>Schneider</u> <u>et al. (2005)</u>
SRM 1649	Mouse macrophages and Kupffer cells	NA	100 μg/mL or 200 μg/mL for 24 h	Concentration-dependent increases in IL-6 (protein). Also increased mRNA of IL-6, TNF, and IL-12	<u>Tan et al.</u> (2009)
EHC93 or ROFA	AMs	NA	0.01–0.1 mg/mL for 24 h	Concentration-dependent TNF response by EHC93. ROFA less potent than EHC93. Also increased cytokine profile by EHC93 (increased IL-8, IL-6, GM-CSF, MIP-1a, MIP-1b, IL-1 α , IL-1 β , IL-10, and TNF)	<u>van Eeden</u> et al. (2001)
PM _{2.5} from Cache Valley, Utah, USA	BEAS-2B	Water (sonication)	0.8–3.9 μg/mL for 24 h	Increased IL-6 excretion (protein). Excretion of TNF was unaltered	Watterson et al. (2007)

Supplemental Table S21 Markers of inflammation in cultured cells exposed to air pollution particles

Supplemental Table S21 (continued)

Particles	Cells	Extraction	Exposure conditions	Effect	Reference
SRM 1649 or ROFA	RLE, AMs, or co- culture	NA	1–50 μg/mL for 24 h	Unaltered secretion of MIP2 and TNF in mono-cultures, whereas there were concentration-dependent increases in co-cultures	<u>Tao & Kobzik</u> (2002)
PM ₁₀ , PM _{2.5} , PM ₁ , and PM _{0.4} from Milan, Italy, collected during summer or winter	THP-1 or THP-1/ A549 co-cultures	Water (sonication)	10 $\mu g/cm^2$ for 24 h	Increased IL-6 and IL-1 β secretion (protein) in THP-1 or co-cultures (and IL-8 in co-cultures) exposed to PM ₁₀ collected during summer (correlated with endotoxin level)	<u>Longhin et al.</u> (2013)
Fine and coarse fraction of PM from 4 cities in Europe	Rat lung type 2 cells and AMs	Methanol	10–50 mg/mL for 24 h	Increased MIP2, IL-6, and TNF (protein) with both spatial and temporal variability. Coarse fraction more potent than fine fraction	<u>Dybing</u> et al. (2004), <u>Hetland et al.</u> (2005)
Coarse, fine, and ultrafine particles from 6 cities in Europe	RAW 264.7	Methanol (sonication)	15–300 μg/mL for 24 h	Concentration-dependent increases in NO (Griess), TNF, IL-6, and MIP2 (protein). Coarse particles more potent than fine and ultrafine particles. Inter-city variability in inflammation potency	<u>Jalava et al.</u> (2007)
PM _{2.5} from rural and urban (Dakar city) sites in Senegal	BEAS-2B	Mechanically recovered	3–12 μg/mL for 24–48 h	Increased TNF, IL-8, and IL-6, and unaltered IL-1 β secretion. Increased IL-1 β , IL-6, and IL-8 mRNA levels, and unaltered TNF. Urban sites more potent than rural site	<u>Dieme</u> et al.(2012)
PM ₁₀ from different locations in Mexico City, Mexico	J774A.1 and RLE	Mechanically recovered	40–80 μg/cm² for 24 h	Concentration-dependent increases in IL-6 and TNF (protein) in J774A.1 cells and PGE_2 secretion by RLE cells. Particles from one location (business area) more potent than those from other areas (industry or residential areas)	<u>Alfaro-</u> <u>Moreno</u> <u>et al.(2002)</u>
PM _{2.5} collected at a busy street or urban background in Copenhagen, Denmark	A549	Water	25 μg/mL for 24 h	Increased expression level of IL-6 and IL-8, although not a clear difference between street and background particles	<u>Sharma</u> et al.(2007)
PM from a town with many wood stoves and a rural area, Denmark	A549 and THP-1	Mechanical collection from plates	2.5–100 μg/mL for 3 h	Unaltered expression of CCL2, I-L8, and IL-6 (mRNA) in A549 cells. Increased expression levels of CCL2, IL-8, and TNF (mRNA) in THP-1 cells	<u>Danielsen</u> et al.(2011)
PM ₁₀ from Utah Valley, Utah, USA	BEAS-2B	Water (agitation)	125–500 μg/mL for 2 h or 24 h	Concentration-dependent increase in IL-6 and IL-8 (protein or protein) production. Lowest potency had samples that were collected during a strike at the steel mill in the area	Frampton et al.(1999)
PM from a street tunnel in Oslo, Norway	THP-1	Scraping off the filter	20–280 μg/mL for 12 h	Increased TNF, IL-1 β , and IL-8 (protein) secretion. Undetectable levels of IL-4, IL-6, and IL-10. No difference between particles collected in seasons with or without use of studded tyres	<u>Kocbach</u> <u>et al.(2008)</u>

Supplemental Table S21 (continued)

Particles	Cells	Extraction	Exposure conditions	Effect	Reference
Water-soluble fraction of PM_{10} from cities in Switzerland	Rat AMs (NR8383)	Water (shaking)	Concentration not reported. Incubation for 40 h	Highest potency of particles collected during summer and autumn compared with other seasons	<u>Monn et al.</u> (2003)
PM ₁₀ from Helsinki, Finland, collected during spring or winter	RAW 264.7	Methanol (sonication)	15–1000 μg/mL for 24 h	Increased TNF, IL-6 production (protein), and NO (Griess). Particle fraction had stronger response than water-soluble fraction. No effect of DFO treatment. Spring samples more potent than winter samples	<u>Salonen et al.</u> (2004)
Outdoor and indoor PM _{2.5} samples from Boston, Massachusetts, USA, or SRM 1649	Rat AMs	Water (sonication)	100 μg/mL for 20 h	Indoor samples more potent than outdoor samples in TNF secretion (protein) after normalization for endotoxin content	<u>Long et al.</u> (2001)
PM from Chapel Hill, North Carolina, USA, and ROFA	Bronchial epithelial cells and AMs	Water (sonication)	11 μg/m³ or 50 μg/m³ for 18 h	Increased IL-6 and IL-8 release (protein). Coarse particles more potent than fine and ultrafine particles. Low temporal variability in cytokine secretion	<u>Becker et al.</u> (2005)
Fine and coarse particles from Chapel Hill, North Carolina, USA, SRM 1649, volcanic ash, or Mount St Helens dust	Human AMs	Water (sonication)	50–100 μg/mL for 24 h	SRM 1649 increased the production of TNF, IL-6, and IL-8. Fine and coarse particles increased TNF and IL-8, with a tendency to be size- and season-dependent	<u>Sawyer et al.</u> (2010)
Fine and coarse particles from Duisburg (industrial area) and Borken (rural area), Germany	WBCs	Water (sonication)	< 0.7 mg/mL for 4 h	Coarse particles generated more IL-8 and TNF than fine particles. Particles from Borken were most inflammogenic. Inflammogenicity correlated with endotoxin content in sample	<u>Schins et al.</u> (2004)
Coarse, fine, and ultrafine samples from the Netherlands	RAW 264.7	VACES	6.3–100 μg/mL for 16 h	Increased secretion of TNF, IL-6, and MIP2. Fine particles more inflammogenic than ultrafine particles (coarse particles were not included because of high endotoxin content)	<u>Steenhof et al.</u> (2011)
Coarse, fine, or ultrafine particles from Tuxedo, New York, USA, or SRM 1649	HMCEC-LB1	Water (outdoor air particles)	25–200 mg/mL for 24 h	Concentration-dependent increases in IL-6 secretion (protein). Decreased IL-6 production of fine particles by DFO pre-treatment	<u>Qu et al.</u> (2010)
PM in different size fractions from urban and rural sites, United Kingdom	A549	Water (sonication)	100 µg/cm² for 24 h	Increased IL-8 release, with some dependency on locations and particle sizes	<u>Wessels et al.</u> (2010)
Aqueous extract or insoluble fraction of TSP from North Provo, Utah, USA	BEAS-2B	Water (agitation)	500 µg for 24 h	Increased IL-8 release (protein). Higher response for soluble extract compared with insoluble fraction	<u>Ghio et al.</u> (1999)

Supplemental Table S21 (continued)

Particles	Cells	Extraction	Exposure conditions	Effect	Reference
PM _{2.5} from Toronto, Canada, or SRM 1648	A549	Water	10–1000 μg/mL for 24 h	Water-insoluble content of SRM 1648 more potent than water-soluble content for IL-8 secretion (protein). Only increased IL-8 secretion at high concentration of PM _{2.5} samples	<u>Akhtar et al.</u> (2010)
SRM 1649, ROFA, or CAPs from Boston, Massachusetts, USA	AMs (LPS- primed)	Saline (sonication) for CAPs	25–100 μg/mL for 20 h	Substantial difference in potency to promote TNF secretion by CAPs collected on different days. TNF secretion was attributed mainly to the particle fraction, whereas the water-soluble fraction had little effect	<u>Imrich et al.</u> (1999)
ROFA	Bronchial epithelial cells	NA	5–200 μg/mL for 2 h or 24 h	Concentration-dependent increase in IL-6, IL-8, and TNF (protein and mRNA). Decreased cytokine secretion by treatment with DFO or DMTU	<u>Carter et al.</u> (1997)
ROFA	RAW 264.7 and hamster AMs	Saline (sonication)	100–400 μg/mL for 0.5 h and 16 h	Concentration-dependent increase in TNF production (protein), which was inhibited by DFO. <i>MIP2</i> gene expression increased after 30-minute exposure	<u>Goldsmith</u> et al. (1998)
SRM 1649 or CAPs from Boston, Massachusetts, USA	AMs (LPS- primed)	Water (sonication)	100 μg/mL for 20 h	Increased TNF release (protein), which was diminished by treatment with NAC, catalase, or DMTU	<u>Imrich et al.</u> (2007)
PM ₁₀ from an urban street in Stockholm, Sweden	RAW 264.7	Water (sonication)	1–100 μg/mL for 18 h	Concentration-dependent increase in IL-6 and TNF (protein), which was inhibited by DFO or NAC. Increased NO release. Increased mRNA expression of <i>IL-6</i> and <i>iNOS</i> (non-significant for TNF)	<u>Lindbom</u> et al. (2007)
$\mathrm{PM}_{2.5}$ from Vermont, USA	Murine alveolar type 2 cells	Water and sonication	10 μg/cm² for 1–8 h	Increased NFκB binding to DNA, which was ameliorated by pre-treatment with catalase, but not DFO	<u>Shukla et al.</u> (2000)
PM from Düsseldorf, SRM 1648, EHC93, volcanic and oil fly ash	Human or rat alveolar cells	Not specified	18.5–500 μg/mL for 18–20 h	Increased TNP and IL-6 secretion (protein) by EHC93, PM from Düsseldorf, and SRM 1648. Unaltered secretion by volcanic and oil fly ash. DFO pre-treatment had no effect on SRM 1648-mediated IL-6 secretion	<u>Becker et al.</u> (1996)
EOM of coarse and fine particles from Paso del Norte Air Basin, Texas, USA	BEAS-2B	DCM	50 μg/mL for 18 h	Unaltered gene expression of IL-6 and IL-8	<u>Lauer et al.</u> (2009)
EOM of road tunnel particles from Shanghai, China	A549	DCM (sonication)	1–400 µg/mL for 24 h	Bell-shaped concentration effect curve with increased gene expression of <i>IL-6</i> and <i>IL-8</i> (mRNA). Unaltered expression of <i>CCL2</i> and <i>RANTES</i>	<u>Shang et al.</u> (2013)

AMs, alveolar macrophages; CAPs, concentrated ambient particles; DCM, dichloromethane; DFO, deferoxamine; DMTU, dimethylthiourea; EOM, extractable organic matter; GM-CSF, granulocyte macrophage colony-stimulating factor; h, hour or hours; IL-6, interleukin-6; NA, not applicable; NAC, *N*-acetylcysteine; PBS, phosphate-buffered saline; PGE₂, prostaglandin E2; PM, particulate matter; PM₁₀, particulate matter with particles of aerodynamic diameter < 10 μ m; PM_{2.5}, particulate matter with particles of aerodynamic diameter < 2.5 μ m; ROFA, residual oil fly ash; SRM, standard reference mixture; TNF, tumour necrosis factor; VACES, versatile aerosol concentration enrichment system; WBCs, white blood cells; wk, week or weeks.

Supplemental Table S22 Production of reactive oxygen species in cells exposed to air pollution particles

Particles	Cells	Extraction	Exposure conditions	Effect	Reference
PM _{2.5} from Porte d'Auteuil, France	Human nasal epithelial cells	Water (sonication)	10-80 μg/cm ² for 3 h	Increased ROS production (DCFH)	<u>Auger et al. (2006)</u>
PM _{2.5} from Vitry-sur- Seine, France, or SRM 1648	Human bronchial epithelial cells (16-HBE)	Sonication	10 µg/cm² for 4 h	Increased ROS production (DCFH)	<u>Baulig et al. (2003)</u>
PM from Düsseldorf, SRM 1648, EHC93, volcanic and oil fly ash	Human or rat alveolar cells	Not specified	18.5–500 μg/mL for 0.5 h	Increased ROS production (luminol and chemiluminescence) by all particles. Oil fly ash particles more potent than other particles	<u>Becker et al. (1996)</u>
PM from Chapel Hill, North Carolina, USA, and ROFA	Bronchial epithelial cells and AMs	Water (sonication)	11 or 50 μg/m ³ for 1 h	Increased ROS production (DCFH and DHR). Coarse, fine, and ultrafine particles generated the same level of ROS on mass basis. High temporal variability in ROS production	<u>Becker et al. (2005)</u>
PM from a town with many wood stoves and a rural area, Denmark	A549 and THP-1	Water	2.5–100 μg/mL for 3 h	Concentration-dependent increase in ROS production (DCFH)	<u>Danielsen et al.</u> (2010)
ROFA and CAPs from Boston, Massachusetts, USA	Hamster AMs	Water (sonication)	25–200 μg/mL and 4–20 μg/mL for 0.5 h	Concentration-dependent increase in ROS production (DCFH). ROFA had a stronger effect than CAPs	<u>Goldsmith et al.</u> (1997)
Airborne PM from Düsseldorf, Germany	Human bronchial epithelial cells (IB3–1 and S-9 CF)	Baghouse	25 μg/cm² for 1 h	Increased ROS production (DCFH)	<u>Kamdar et al.</u> (2008)
PM ₁₀ from a busy street in Stockholm, Sweden	A549	Water (sonication)	20 μg/cm² for 2 h	Increased ROS production (DCFH)	<u>Karlsson et al.</u> (2008)
PM from Osaka, Japan	Human mononuclear blood cells	Aqueous solution	50–650 μg/mL for 200 minutes	Bell-shaped ROS production (lucigenin chemiluminescence assay)	<u>Ohyama et al.</u> (2007)
PM _{2.5} from Vermont, USA	Murine alveolar type 2 cells	Water (sonication)	10 μg/cm² for 1–8 h	Increased ROS (DCFH)	<u>Shukla et al. (2000)</u>
PM ₁₀ from Beijing, China	A549	Water (sonication)	10 μg/mL for 24 h	Increased ROS production (DCFH and DHE) by particles as well as soluble and insoluble fractions thereof	<u>Yi et al. (2014)</u>
PM _{2.5} from Denver, Colorado, USA	NR8383 AMs	Water (shaking)	20–200 pg/cell for 2 h	Increased ROS production. Positive association with iron, soil dust sources, and water-soluble carbon	<u>Zhang et al. (2008)</u>
SRM 1648	AMs and type 2 cells	NA	20 μg/cm ² (or 200 μg/mL) for 20 h	Increased ROS production in type 2 cells, but not macrophages	<u>Becher et al. (2007)</u>

Supplemental Table S22 (continued)

Particles	Cells	Extraction	Exposure conditions	Effect	Reference
Oil fly ash from a power plant in Sicily, Italy	A549	Water	17.2–68.8 of VOSO ₄ equiv for 0.5–4 h	Concentration-dependent increase in ROS production (DCFH) Ameliorated by treatment with DFO	<u>Di Pietro et al.</u> (2009)
Coal fly ash from a dumping site of a thermal power plant in Aligarh, India	Mononuclear blood cells	DMSO (0.5%) or water (sonication)	600–2400 ppm for 24 h	Increased generation of ROS (DCFH) by DMSO (0.5%) and ROS production by aqueous extract	<u>Dwivedi et al.</u> (2012)
SRM 1648	Human pulmonary artery endothelial cells	NA	1–100 μg/mL for 5–120 minutes	Concentration- and time-dependent increase in ROS production (Amplex Red)	<u>Li et al. (2006)</u>
SRM 1648	RAW 264.7	NA	62.5 μg/cm² for 4 h	Increased ROS (DCFH)	<u>Schneider et al.</u> (2005)
PM_{10} , $PM_{2.5}$, PM_1 , and $PM_{0.4}$ from Milan, Italy, collected during summer or winter	A549 and THP-1	Water (sonication)	10 μg/cm² for 1 h	All size fractions increased ROS production. Samples collected during winter generally slightly more potent than samples collected during summer	<u>Longhin et al.</u> (<u>2013)</u>
PM ₁₀ from Utah Valley, Utah, USA, dust from 3 different years	AMs	Water (sonication)	≤ 1 mg/mL for 20 minutes or 24 h	Increased ROS production (chemiluminescence) by samples from the year of collection, which was not affected by DFO treatment. Unaltered (or decreased) ROS production by dihydrorhodamine 123 assay	<u>Soukup et al.</u> (2000)
ROFA and CAPs from Boston, Massachusetts, USA	Hamster AMs	Water (sonication)	25–200 μg/mL for 0.5 h	Concentration-dependent increase in ROS production (DCFH), which was inhibited by DFO. Substantial variation in ROS production by CAPs collected on different days	<u>Goldsmith et al.</u> (1998)
EOM of PM from Taiwan, China	MCF-7	Hexane/acetone (ultrasound)	0.04–0.05 m ³ - air equiv for 24–72 h	No clear difference between PM from urban and rural sites in regard to ROS production (DCFH)	<u>Chen et al. (2013)</u>
EOM of PM ₁₀ from Kaifaqu district, Dalian, China	HepG2	Sequential extraction in DCM, acetone, and methanol	7.5-30 mg/mL for 1 h	Concentration-dependent increase in ROS production (DCFH)	<u>Jiang et al. (2011)</u>
EOM of road tunnel particles from Shanghai, China	A549	DCM (sonication)	10–400 μg/mL for 1 h	Concentration-dependent increase	<u>Shang et al. (2013)</u>

AMs, alveolar macrophages; CAPs, concentrated ambient particles; DCFH, 2',7'-dichlorodihydrofluorescein; DCM, dichloromethane; DFO, deferoxamine; DHE, dihydroethidium; DHR, dihydrorhodamine 123; DMSO, dimethyl sulfoxide; EOM, extractable organic matter; equiv, equivalent; h, hour or hours; NA, not applicable; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 μ m; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μ m; ROFA, residual oil fly ash; ROS, reactive oxygen species; SRM, standard reference mixture.

Particles	Extraction	Method	Effect	Reference
Fine particles from Duisburg, Germany	Water (sonication)	ESR with H_2O_2 and DMPO as spin trap	Concentration-dependent increase in ROS production (DMPO-OH signal), which was inhibited by DFO or catalase	<u>Knaapen et al. (2002)</u>
TSP from the city centre of Athens, the suburb of Piraeus, or street dust, Greece	PBS (sonication)	ESR with H ₂ O ₂ and without DMPO	Little difference in potency between TSP from different locations, whereas street dust generated lower levels of ROS. Decreased ROS production by treatment with DFO	<u>Valavanidis et al.</u> (2000)
TSP, PM_{10} , and $PM_{2.5}$ from Athens, Greece	Water (sonication)	ESR with H ₂ O ₂ and without DMPO	Detection of DMPO-OH signals. No difference between size fractions	<u>Valavanidis et al.</u> (2005)
Coal fly ash from a dumping site of a thermal power plant in Aligarh, India	DMSO (0.5%) or water (sonication)	NTB	Increased generation of ROS by DMSO (0.5%) and aqueous extract	<u>Dwivedi et al. (2012)</u>
Coal fly ash from a power plant in Arnhem, Netherlands	Not specified	ESR with DMPO	Detection of DMPO-OH signals. Association between ROS production and release of iron	<u>van Maanen et al.</u> <u>(1999)</u>
PM _{2.5} from San Joaquin Valley, California, USA	Salt solution (shaking)	Benzoate as chemical probe of hydroxyl radicals	Particles from urban area more potent than particles from rural area. Treatment with DFO decreased ROS production	<u>Shen et al. (2011),</u> <u>Shen & Anastasio</u> (2011, 2012)
SRM 1648 or SRM 1649	Water	Deoxyribose assay	Concentration-dependent increase in ROS production. SRM 1649 more potent than SRM 1648. Inhibition of ROS production by DFO	<u>Ball et al. (2000)</u>
PM from Utah Valley, Utah, USA	Water (agitation)	Deoxyribose assay with H_2O_2	Higher ROS production by samples collected during activity at a steel mill. Decreased ROS production by DFO or antioxidant (DMTU or DMSO) treatment	<u>Frampton et al.</u> (1999)
Aqueous extract from Utah Valley, Utah, USA, before, during, and after a strike at the steel mill	Water (agitation)	Deoxyribose assay	Increased ROS production in samples before and after the strike compared with samples collected during the strike. Decreased ROS production by treatment with DFO or DMTU	<u>Ghio & Devlin</u> (2001)
Water-soluble or insoluble fraction of CAPs from Boston, Massachusetts, USA	Water (sonication)	DCFH with H ₂ O ₂	Only ROS production in the presence of H_2O_2 . Water- soluble fraction more potent than insoluble fraction. Diminished ROS production by treatment with DFO	<u>Imrich et al. (2007)</u>
PM ₁₀ from an urban street in Stockholm, Sweden	Water (sonication)	DTT	Concentration-dependent increase, which was slightly inhibited by DFO	<u>Lindbom et al.</u> (2007)
PM _{2.5} from urban air particulates at 20 different sites (19 European cities)	Water	ESR and H_2O_2 with DMPO as spin trap	Different ROS production potential between sites and over time. Heterogeneous association between ROS production and PM constituents across locations	<u>Künzli et al. (2006),</u> <u>Nawrot et al. (2009)</u>
$\mathrm{PM}_{2.5}$ from 5 cities in the USA	PBS (vortexing)	ESR	Similar ESR spectra of samples collected in different cities, whereas no ESR signal observed in blank filters	<u>Dellinger et al.</u> (2001)

Supplemental Table S23 Acellular production of reactive oxygen species

Supplemental Table S23 (continued)

Particles	Extraction	Method	Effect	Reference
PM from a town with many wood stoves and a rural area, Denmark	Mechanical collection from plates	ESR (with ascorbate and DMPO) and DCFH	Rural particles more potent than city particles by ESR. Same ROS production by particles for the DCFH assay	<u>Danielsen et</u> <u>al.(2011)</u>
Fine and coarse particles from Duisburg (industrial area) and Borken (rural area), Germany	Water (sonication)	ESR with H_2O_2 and DMPO as spin trap	Fine and coarse particles from Borken generated the same level of ROS. Coarse particles from Duisburg generated more ROS than fine particles	<u>Schins et al. (2004)</u>
Fine and coarse particles from various cities in Germany	Water (sonication)	ESR with H ₂ O ₂ and DMPO as spin trap	Coarse particles generated higher level of ROS than fine particles. Samples from industrial area (Dortmund and Duisburg) generated higher level of ROS than samples from rural site (Borken)	<u>Shi et al. (2006)</u>
PM _{2.5} from a smelter site and non-industrialized site in Germany	Water (sonication)	ESR	Higher levels of ROS production in samples from industrial area	<u>Schaumann et al.</u> (2004)
PM_{10} from an urban or suburban site in Mexico City, Mexico	Mechanically recovered	ESR with H_2O_2 and DMPO as spin trap	Particles from urban site more potent than particles from suburban site. Temporal variation related to wind direction at sampling site	<u>Quintana et al.</u> (2011)
PM ₁₀ from a busy street in Maastricht, Netherlands	Not specified	ESR with DMPO as spin trap	Presence of oxygen radicals, but no carbon-centred radicals	<u>de Kok et al. (2004)</u>
$\mathrm{PM}_{\mathrm{10}}$ from Edinburgh, Scotland	Water (sonication)	2.3-dihydroxybenzoic acid production	Increased ROS production	<u>Donaldson et al.</u> (1997)
Aqueous extract or insoluble fraction of TSP from North Provo, Utah, USA	Water (agitation)	Deoxyribose assay with H_2O_2	Increased ROS production. Higher response by soluble extract compared with insoluble fraction	<u>Ghio et al. (1999)</u>
PM ₁₀ from Helsinki, Finland, collected during spring or winter	Methanol (sonication)	ESR with H_2O_2	Increased production, without clear seasonal variation	<u>Salonen et al. (2004)</u>
$PM_{2.5}$ and PM_{10} from different sites in the Netherlands	Water	ESR with H_2O_2 and DMPO as spin trap	PM_{10} from major streets more potent than urban background. PM_{10} more potent than $PM_{2.5}$	<u>Boogaard et al.</u> (2012)
Fine and coarse particles from Düsseldorf, Germany	Water (sonication)	ESR with H_2O_2 and DMPO as spin trap	Coarse PM generated more ROS (DMPO-OH signal) than fine PM when compared at equal mass. Samples collected during summer had stronger ROS generating potency than autumn/winter samples	<u>Shi et al. (2003)</u>
PM _{2.5} and PM ₁₀ from Maastricht, Netherlands	Not reported	ESR with DMPO as spin trap	No difference in ROS generating ability between $PM_{2.5}$ and PM_{10} samples. Season-dependent variation in ROS generating ability of outdoor PM_{10} samples	<u>Briedé et al. (2005)</u>
PM _{2.5} ,PM ₁₀ , or TSP from school playgrounds in Maastricht, Netherlands	No extraction	ESR with DMPO as spin trap	Differences in ROS generation in samples collected at different locations. No clear difference between different size modes	<u>Hogervorst et al.</u> (2006)

Supplemental Table S23 (continued)

Particles	Extraction	Method	Effect	Reference
PM in different size fractions from urban and rural sites, United Kingdom	Water (sonication)	EST with H ₂ O ₂ and DMPO	ROS production highest for sites near traffic. Little effect related to size fractions	Wessels et al. (2010)
PM _{2.5} from San Joaquin Valley, California, USA	Not specified	DTT	Temporal variation in ROS production, although no clear effect of summer and winter	<u>Charrier &</u> <u>Anastasio (2012)</u>
EOM of TSP from Fresno, California, USA	DCM (sonication)	DTT (measured as H_2O_2 production)	Increased ROS production, which correlated with content of 3 active quinones	<u>Chung et al. (2006)</u>
PM _{2.5} from Los Angeles, California, USA, during wildfires	Water	DTT	Increased ROS production by samples collected during wildfire compared with samples collected after wildfire	<u>Verma et al. (2009)</u>
PM _{2.5} from Atlanta, Georgia, USA	Water or methanol	DTT	Day-to-day variation in potency of particles. Methanol extract more potent than water extract	<u>Verma et al. (2012)</u>
Urban and rural sites in Norfolk, Virginia, USA	Water (sonication)	DTT	Ultrafine particles more potent on mass basis than $\mathrm{PM}_{\mathrm{2.5}}$	<u>Jeng (2010)</u>
Coarse, fine, and ultrafine particles from Los Angeles basin, California, USA	VACES	DTT	Ultrafine particles had higher redox activity than fine and coarse particles on weight basis	<u>Cho et al. (2005)</u>
Coarse or fine particles from Mexico City, Mexico	Water	DTT	Fine particles more potent than coarse particles. Temporal and spatial differences in potency	<u>De Vizcaya-Ruiz et</u> <u>al. (2006)</u>
Coarse, fine, and ultrafine particles from Los Angeles basin, California, USA	VACES	DTT	Ultrafine particles generated higher DDT oxidation than coarse and fine particles on mass basis. Substantial spatial and temporal variation in ROS production ability of ultrafine particles	<u>Li et al. (2003)</u>
PM from Los Angeles and San Francisco, California, USA	VACES	DTT	Higher ROS production on mass basis for ultrafine particles compared with fine and coarse particles. Strong correlation between organic carbon content and ROS production	<u>Ntziachristos et al.</u> (2007)
Coarse, fine, and ultrafine samples from the Netherlands	VACES	DTT	Increased ROS production, with generally the same potency for all size fractions	Steenhof et al. (2011)
$\mathrm{PM}_{2.5}$ from Toronto, Canada, and SRM 1648	Water	DTT	Similar level of ROS production of PM _{2.5} and SRM 1648. Water-soluble fraction of SRM 1648 more potent than water-insoluble fraction	<u>Akhtar et al. (2010)</u>

CAPs, concentrated ambient particles; DCFH, 2',7'-dichlorodihydrofluorescein; DCM, dichloromethane; DFO, deferoxamine; DMPO, 5,5-dimethyl-1-pyrroline-*N*-oxide; DMSO, dimethyl sulfoxide; DMTU, dimethylthiourea; DTT, dithiothreitol; EOM, extractable organic matter; ESR, electron spin resonance; H_2O_2 , hydrogen peroxide; PBS, phosphate-buffered saline; PM, particulate matter; PM_{10} , particulate matter with particles of aerodynamic diameter < 10 µm; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 µm; ROS, reactive oxygen species; SRM, standard reference mixture; TSP, total suspended particles; VACES, versatile aerosol concentration enrichment system.

Location	Animals	Experimental design/ exposure	End-point	Results	Call	Reference
Hamilton, Canada	Herring gull	Feral gulls collected from a steel mill environment and rural reference locations	Heritable minisatellite DNA mutation by multilocus DNA fingerprinting	Significant > 2-fold increase in mutation rate observed in offspring from the industrial site compared with rural locations	+	<u>Yauk &</u> Quinn (1996)
Urban and industrial sites, Canada and USA	Herring gull	Feral gulls collected from 4 steel mill environments, 2 urban sites, and 3 rural reference locations	Heritable minisatellite DNA mutation by multilocus DNA fingerprinting	Significant 2-fold increase in mutation rate observed in offspring from the industrial sites compared with rural locations. No difference between urban sites and rural sites. Minisatellite mutation rates decreased with increasing distance from industrial coking oven and urbanization site	+	<u>Yauk et al.</u> (2000)
Hamilton, Canada	Swiss Webster mice, outbred	In situ, 10 wk exposure to outdoor air near steel mills and a rural reference site	Heritable mutation at ESTR loci in offspring conceived 6 wk after exposure	Significant 1.5–2-fold increase in mutation frequency at several loci observed in offspring from the steel mills site compared with rural locations	+	<u>Somers et al.</u> (2002)
Hamilton, Canada	Swiss Webster mice, outbred	In situ, 10 wk exposure to outdoor air near steel mills and a rural reference site	Heritable mutation at ESTR loci in offspring conceived 9 wk after exposure	Offspring of mice from the urban/industrial site showed 1.9–2.1-fold increased mutation frequency in ESTR of paternal origin, compared with offspring from rural locations. HEPA filtration of outdoor air significantly reduced heritable mutation rates	+	<u>Somers et al.</u> (2004)
Hamilton, Canada	C57BL/CBA F1 mice, inbred	In situ, mice exposed to outdoor air near steel mills or HEPA-filtered air for 3, 10, or 16 wk, followed by 6 wk in the laboratory	ESTR loci mutation in sperm; DNA adducts in testes; DNA damage in sperm	Significant 1.6-fold increase in germline mutation frequency at Ms6-hm observed in mice exposed to outdoor air at the steel mills site compared with HEPA-filtered air after 16 wk, but not at 3 wk or 10 wk. DNA strand breaks in mature sperm were significantly elevated in the outdoor air-exposed group after 3 wk. No detectable DNA adducts observed in testes samples	+	<u>Yauk et al.</u> (2008)
Rome, Italy	Wild mice Mus domesticus	Feral mice collected from 3 sites of low, medium, and high air pollution from traffic	Abnormality on the sperm cells of cauda epididymis	Significant increase in frequency of abnormal sperm cells from sites with medium ($P < 0.05$) and heavy ($P < 0.01$) pollution compared with low-air-pollution control site. Sperm abnormality includes banana-like form, narrow form without hook, with 2 tails and triangular head, amorphous, and folded form	+	<u>Ieradi et al.</u> (1996)

Supplemental Table S24 Genotoxic effects in germ cells of animals exposed to polluted outdoor air

Supplemental Table S24 (continued)

Location	Animals	Experimental design/ exposure	End-point	Results	Call	Reference
Shanghai, China	Male mice (strain not specified)	Mice treated with extracts of TSP collected from 13 sites in Shanghai	Sperm morphology, shape of normal and abnormal heads	Increased germ-cell deformations observed for most sites in winter, especially for the sites of Yang Pu, Huang Pu, and Western Suburb Park	+	<u>Mao et al.</u> (1993)
Kunming, China	Oregon R male Drosophila	<i>Drosophila</i> fed with particulate extracts for 3 days at concentrations of 0.1% and 0.2% of stock solution of TSP extract from Kunming	Germ-cell mutation in <i>Drosophila</i> by sex-linked recessive lethal test (SLRL)	Treatment of $\frac{1}{2}$ LC ₅₀ in male <i>Drosophila</i> caused mutation with frequency of 0.76% and 0.92%, respectively. No mutation observed in the control	+	<u>Yang & Lu</u> (1989)
Magnitogorsk, Russian Federation	Drosophila melanogaster	Exposure to outdoor air and snow samples from different Magnitogorsk areas, 4 yr monitoring	Dominant lethal induction in gametes	Positive induction of DLM in the gametes of <i>Drosophila</i> ; positive correlation between all genotoxic chemicals found in the outdoor air and the rate of DLM	+	<u>Legostaeva</u> et al. (2010)

DLM, dominant lethal mutation; ESTR, expanded simple tandem repeat; HEPA, high-efficiency particulate air; LC₅₀, median lethal concentration; TSP, total suspended particulates; wk, week or weeks; yr, year or years.

Supplemental Table S25 Studies that used cultured animal cells to assess the ability of outdoor air to induce malignant cell transformation

Geographical location	Test article Assay/exposure system		End-point(s) examined	Results	Reference
Rhine-Ruhr region, Germany	"City smog" collected at heavily industrialized location (17), collected on GFFs. MeOH extraction, followed by CX and Pro fractionation	Syrian hamster kidney cells, 18 h exposure to extract in DMSO and subsequently infected with SV40	Cell transformation	Significant dose-related (equiv m ³ /assay) increase in transformation frequency induced by total extract (up to 20-fold vs control). MeOH and CX fractions also elicited significant increase in transformation frequency. Inoculation of cells transformed in vitro in Syrian golden hamsters elicited malignant tumours	<u>Seemayer. et</u> <u>al. (1981, 1984</u>)
Duisburg, Germany	Airborne PM from industrialized Rhine–Ruhr region, Draeger Box Micron filter. CX extraction	Syrian hamster kidney cells, 18 h exposure to extract in DMSO and subsequently infected with SV40	Cell transformation	Significant dose-related (m ³ equiv/mL) increase in transformation frequency. As little as 0.25–0.5 m ³ equiv elicited a significant enhancement in transformation frequency	<u>Seemayer et</u> <u>al. (1987a), Seemayer et al. (1988)</u>
Rhine–Ruhr region, Germany	20 samples of "city smog" collected between 1975 and 1986 at highly industrialized locations, collected on GFFs. Organic extraction	Syrian hamster kidney cells, 18 h exposure to extract in DMSO and subsequently infected with SV40	Cell transformation	Düsseldorf provided as an example. Significant dose-related (equiv m ³ /mL) increase in transformation frequency. As little as 0.5 m ³ equiv elicited a significant enhancement in transformation frequency	<u>Seemayer et al.</u> (<u>1987b)</u>
Rhine–Ruhr region, Germany	41 samples of "city smog" collected between 1975 and 1990 at highly industrialized locations, collected on GFFs. Organic extraction	Syrian hamster kidney cells, 18 h exposure to extract in DMSO and subsequently infected with SV40	Cell transformation	Site 54 (Düsseldorf) shown as an example. Significant dose-related (equiv m ³ /mL) increase in transformation frequency. As little as 0.5 m ³ equiv elicited a significant enhancement in transformation frequency	<u>Seemayer et al.</u> (1990a)
Rhine-Ruhr region, Germany.	Airborne PM from Duisburg and Düsseldorf, collected on GFFs with high-volume sampler. DCM extraction	Syrian hamster kidney cells, 18 h exposure to extract in DMSO and subsequently infected with SV40	Cell transformation	Significant dose-related (equiv m ³ /assay) increase in transformation frequency. Less than 1 m ³ equiv required to elicit a significant enhancement in transformation frequency. Higher responses in March compared with October	<u>Seemayer et al.</u> (1990a)
Rhine–Ruhr region, Germany.	Airborne PM from Duisburg and Düsseldorf, collected on GFFs with high-volume sampler. DCM extraction	Syrian hamster kidney cells, 18 h exposure to extract in DMSO and subsequently infected with SV40	Cell transformation	Significant dose-related (equiv m ³ /assay) increase in transformation frequency. As little as 0.5 m ³ equiv elicited a significant enhancement in transformation frequency. Inoculation of cells transformed in vitro in Syrian golden hamsters produced malignant tumours (mostly sarcomas)	<u>Seemayer &</u> <u>Hornberg</u> (1998)

Supplemental Table S25 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Athens, Greece (1983)	Monthly PM samples collected on cellulose filters with high-volume sampler. Hx sonication extraction	BALB/c 3T3 cells, 48 h treatment with extract in DMSO	Cell transformation (type III foci)	Significant dose-related (µg of EOM/mL) increase in transformation frequency	<u>Athanasiou et</u> <u>al. (1987)</u>
Shanghai, China	TSP from 4 locations in industrial Baoshan industrial district. DEE Soxhlet extraction	CHO fibroblasts, treated with extracts in DMSO	Cell transformation	3 of the 4 samples examined induced a significant increase in transformation frequency	<u>Lu et al. (1997)</u>
Tokyo, Japan (1980–1983)	Airborne PM from single urban site, collected on quartz filters with high- volume sampler. DCM sonication extraction	Bhas42 cells (BALB/c 3T3 transfected with v-Ha- <i>ras</i>), treated for 3, 7, and 10 days with extracts in DMSO	Cell transformation	Significant dose-related (µg of PM/mL) increase in transformation frequency. Higher potency (per mg of PM) in autumn compared with spring	<u>Ezoe et al.</u> (2004)

CHO, Chinese hamster ovary; CX, cyclohexane; DCM, dichloromethane; DEE, diethyl ether; DMSO, dimethyl sulfoxide; EOM, extractable organic matter; h, hour or hours; equiv, equivalent; GFFs, glass-fibre filters; Hx, hexane; MeOH, methanol; PM, particulate matter; SV40, simian virus 40.

Supplemental Table 526 influence of genotype and lung cancer risk							
Exposure group	Country	Genotype/phenotype method	Result	<i>P</i> value	Reference		
GenAir is a case– control study nested within the EPIC cohort	EPIC, Europe	NAT1, NAT2, GSTM1, GSTM3, GSTT1, GSTPi, CYP1A1, CYP1B1, MnSOD, MPO, NQO1	A polymorphism for NQO1 (involved in oxidative damage scavenging) was strongly associated with lung cancer	Odds ratio of 8.06 (95% CI, 1.74–37.41) for the homozygous variant	<u>Vineis et al. (2006)</u>		
GenAir	EPIC, Europe	Several genes	XRRC and BRCA variants in gene-environment interactions in lung cancer. More genes reported associated with bladder cancer and leukaemia		<u>Manuguerra et al.</u> (2007)		
GenAir	EPIC, Europe	GSTM1, XRCC1	Profile regression analysis of DNA adducts showed increased risk from <i>GSTM1</i> null		<u>Papathomas et al.</u> (2011)		
Indoor air pollution Coal exposure	Asia: 5 studies from China and 1 from Thailand	GSTM1, GSTT1, GSTP1	<i>GSTM1</i> null genotype associated with cancer risk in 4 studies	0.001	Hosgood et al. (2007)		

Supplemental Table S26 Influence of genotype and lung cancer risk

CI, confidence interval; EPIC, European Prospective Investigation into Cancer and Nutrition.

Sup	plemental	Table S27	Influence of	aenotype	and biomark	ers in humans
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Exposure group	Country	Genotype/ phenotype method	Result	<i>P</i> value	Reference
Bus drivers and garage workers	Czech Republic	Several genes	The carriers of at least one variant hOGG1 (Cys) allele tended to higher oxidative damage to lymphocyte DNA than those with the wild genotype, whereas XPD23 (Gln/ Gln) homozygotes were more susceptible to the induction of DNA strand breaks. In contrast, <i>GSTM1</i> null variant seemed to protect DNA integrity		<u>Bagryantseva et al.</u> (2010)
Police officers	Czech Republic	Several genes	The carriers of at least one variant allele, CYP1A1*2C (Ile/ Val), MTHFR 2656, or MS 2656, and the EPHX1-medium phenotype appeared to be more susceptible specifically to the induction of oxidative damage to DNA		<u>Novotna et al. (2007)</u>
Outdoor air pollution, traffic police	Czech Republic	GSTM1	<i>GSTM1</i> null genotype associated with increased sperm DNA damage		<u>Rubes et al. (2010)</u>
Traffic police	India	CYP1A1, GSTM1, GSTT1	CYP1A1 m1 and GSTM1 null showed increased 8-OHdG		<u>Prasad et al. (2013)</u>
Outdoor air pollution Bus drivers and mail carriers	Denmark	GSTM1 NAT2	DNA adducts: no significant effects CAs: increased in bus drivers with <i>GSTM1</i> null and <i>NAT2</i> slow acetylator CAs: increased in mail carriers with <i>NAT2</i> slow acetylator	0.0005	<u>Nielsen et al. (1996b),</u> <u>Knudsen et al. (1999)</u>
Bus drivers	Denmark	Urinary <i>CYP1A2</i> activity	Urinary 8-OHdG: correlation with <i>CYP1A2</i> activity	0.05	<u>Loft et al. (1999)</u>
Bus drivers and mail carriers	Denmark	NAT2	Mutagenic activity in urine increased in bus drivers with <i>NAT2</i> fast acetylator (see Section 4.2.1a)		<u>Hansen et al. (2004)</u>
Workers and residents exposed to traffic	Italy	XPD, XRCC1, XRCC3	DNA adducts: no significant effects		<u>Palli et al. (2001)</u>
Children exposed to urban air pollution	Bangkok, Thailand	CYP1A1, GSTM1, GSTT1	DNA adducts and 1-OHP: no significant effects		Ruchirawat et al. (2007)

CAs, chromosomal aberrations; NAT2, N-acetyltransferase 2; 1-OHP, 1-hydroxypyrene; 8-OHdG, 8-hydroxydeoxyguanosine.

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