

# 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<br>examined                                | Results obtained  | Reference                         |
|---|---|--|--|---|-----------------------------------|
| São Paulo<br>metropolitan<br>area, Brazil | 3 sites in the<br>vicinity of a<br>solid waste<br>incinerator in<br>São Paulo, and a<br>rural reference<br>site | <i>Tradescantia</i> clone 4430, in situ exposures in pots for 5 months   | <i>Tradescantia</i><br>stamen-hair<br>mutation assay | Significant increase in<br>mutation frequency<br>relative to reference site.<br>Negative correlation<br>between mutation<br>frequency and distance<br>from incinerator source   | <u>Ferreira et al.</u><br>(2000)  |
| São Paulo<br>metropolitan<br>area, Brazil | Ibirapuera Park<br>in the southern<br>area of the city<br>of São Paulo  | <i>Tradescantia</i> clones<br>4430 and KU-20,<br>in situ exposures in<br>pots, biweekly sample<br>collection for 1 yr  | <i>Tradescantia</i><br>stamen-hair<br>mutation assay | No significant increase<br>in mutation frequency<br>in 4430. Mutation<br>frequency in KU-20<br>significantly increased<br>in summer months and<br>associated with elevated<br>$NO_2$ and NO   | <u>Ferreira et al.</u><br>(2007)  |
| São Paulo<br>metropolitan<br>area, Brazil | Two urban sites<br>in the city of<br>São Paulo, and a<br>rural reference<br>site                                | <i>Tradescantia</i> 4430,<br>in situ exposures in<br>pots, monthly sample<br>collection after initial<br>2-month exposure  | <i>Tradescantia</i><br>stamen-hair<br>mutation assay | Significant increase in<br>mutation frequency at<br>the high-traffic site only.<br>Positive correlation<br>between mutation<br>frequency and suspended<br>PM levels   | <u>Ferreira et al.</u><br>(2003)  |
| São Paulo<br>metropolitan<br>area, Brazil | High-traffic,<br>urban site in<br>São Paulo, and a<br>rural reference<br>site                                   | Tradescantia clone<br>KU-20, chamber<br>exposures of<br>inflorescences to<br>outdoor air or filtered<br>(0.8 μm porosity)<br>outdoor air, daily<br>sampling for 35 d | <i>Tradescantia</i><br>stamen-hair<br>mutation assay | São Paulo outdoor air<br>induced a significant<br>increase in mutation<br>frequency relative to<br>reference site. Particle<br>removal contributed to<br>a reduction in mutation<br>frequency, but frequency<br>still elevated relative to<br>reference site. Mutation<br>frequency at São Paulo<br>significantly correlated<br>with PM <sub>10</sub> | <u>Guimarães et al.</u><br>(2004) |

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

d, day or days; DCM, dichloromethane; DMSO, dimethyl sulfoxide; h, hour or hours; NO, nitrogen oxide; NO<sub>2</sub>, nitrogen dioxide; PM, particulate matter;  $PM_{10}$ , particulate matter with particles of aerodynamic diameter < 10  $\mu$ m; PUF, polyurethane foam; SO<sub>2</sub>, 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<br>location         | Test article   | Assay/exposure<br>system   | End-point(s)<br>examined   | Results  | Reference                             |
|----------------------------------|--|--|--|--|---------------------------------------|
| Po Valley, Italy<br>(1990)       | Airborne PM from<br>residential/commercial<br>locations in Parma,<br>collected on GFFs with low-<br>volume sampler. Tl Soxhlet<br>extraction       | Diploid D7 strain<br>of <i>S. cerevisiae</i> , 2 h<br>exposure to extract<br>in DMSO, with and<br>without endogenous<br>metabolic activation<br>(i.e. elevated P450 in<br>log-phase cells) | Mitotic gene<br>conversion of <i>trp5</i><br>locus, reversion of<br><i>ilv1–92</i> mutants,<br>mitochondrial<br>DNA mutations<br>("petite" colonies) | 6 of 9 24 h samples collected over a 9-month period<br>induced significant dose-related increases in <i>trp5</i><br>conversions without endogenous activation. 5 of 9<br>samples tested with activation elicited significant<br>responses. Only one sample with endogenous<br>activation induced a significant dose-related<br>increase in <i>ilv1</i> reversion. Increase in petite colonies<br>for spring and summer samples   | <u>Poli et al.</u><br>( <u>1992)</u>  |
| Po Valley, Italy<br>(1990–1994)  | Airborne PM from<br>residential/commercial<br>locations in Parma,<br>collected on GFFs with low-<br>volume sampler. Tl or Ac<br>Soxhlet extraction | Diploid D7 strain<br>of <i>S. cerevisiae</i> , 2 h<br>exposure to extract<br>in DMSO, with and<br>without endogenous<br>metabolic activation<br>(i.e. elevated P450 in<br>log-phase cells) | Mitotic gene<br>conversion of <i>trp5</i><br>locus, reversion of<br><i>ilv1–92</i> mutants,<br>mitochondrial<br>DNA mutations<br>("petite" colonies) | 48 monthly samples analysed. Several samples<br>induced increases in point mutations and gene<br>conversions both with and without endogenous<br>activation. High variability across year and season.<br>No evidence of temporal decline in outdoor air<br>mutagenicity between 1990 and 1994. Several<br>samples induced > 10-fold increase over control.<br>Stronger response for Tl extracts. Several Tl extracts<br>induced increases in mitochondrial DNA mutations | <u>Rossi et al.</u><br>( <u>1995)</u> |
| Po Valley, Italy<br>(1991–1998)  | Airborne PM from<br>residential/commercial<br>locations in Parma,<br>collected on GFFs with low-<br>volume sampler. Tl Soxhlet<br>extraction       | Diploid D7 strain<br>of <i>S. cerevisiae</i> , 2 h<br>exposure to extract<br>in DMSO, with and<br>without endogenous<br>metabolic activation<br>(i.e. elevated P450 in<br>log-phase cells) | Mitotic gene<br>conversion of <i>trp5</i><br>locus, reversion of<br><i>ilv1–92</i> mutants,<br>mitochondrial<br>DNA mutations<br>("petite" colonies) | 96 pooled monthly samples analysed. With<br>endogenous activation, several samples induced<br>significant dose-related (equiv m <sup>3</sup> /mL) increases in<br>point mutations and gene conversions. No evidence<br>of temporal decline in outdoor air mutagenicity<br>between 1991 and 1998  | <u>Poli et al.</u><br>(1999)          |
| Po Valley, Italy<br>(1996, 1997) | Airborne PM from<br>residential/commercial<br>locations in Parma,<br>collected on GFFs with low-<br>volume sampler. Tl or Ac<br>Soxhlet extraction | Diploid D7 strain<br>of <i>S. cerevisiae</i> , 2 h<br>exposure to extract<br>in DMSO, with and<br>without endogenous<br>metabolic activation<br>(i.e. elevated P450 in<br>log-phase cells) | Mitotic gene<br>conversion of <i>trp5</i><br>locus, reversion of<br><i>ilv1–92</i> mutants   | Significant induction of gene conversion and point mutations, and potency values (rev/equiv m <sup>3</sup> ) 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><br>(2001)      |

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

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  $\mu$ m;  $PM_{2.5}$ , particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ 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 <sup>3</sup> ) |                            |                  |                            |
|--------|-------------------|---|----------------------------|------------------|----------------------------|
|        |                   | 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 <sup>b</sup>                              | 1.1                        | 9.4 <sup>b</sup> | 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 <sup>3</sup> ) |                            |                   |                            |
|-------------|-------------------|---|----------------------------|-------------------|----------------------------|
|             |                   | 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                        | 10.0 <sup>a</sup> | 1.1                        |
| Residential | 259               | 7.8 <sup>b</sup>                              | 1.1                        | 6.4 <sup>b</sup>  | 1.1                        |
| Rural       | 441               | 5.0°  | 1.3                        | 6.0ь              | 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<br>location | Test article   | Salmonella strains <sup>a</sup> , assay version  | Results  | Reference                               |
|--------------------------|--|--|--|---|
| Washington, DC,<br>USA   | Time-integrated<br>(baghouse) urban PM<br>collected in 1976 and<br>1977 (SRM 1649). DCM<br>sonication extraction   | TA98, TA100, standard plate<br>incorporation and pre-incubation<br>assays, with and without S9 from<br>Wistar rat liver, Aroclor 1254<br>induction, or PB/5,6-BF induction | S9 source had no effect on TA100 mutagenicity<br>(per µg of EOM). Stronger response with<br>TA98, and TA98 mutagenicity increased with<br>Aroclor-induced S9. Similar responses for plate<br>incorporation and pre-incubation versions   | <u>Tokiwa et al. (1992)</u>             |
| Washington, DC,<br>USA   | Time-integrated<br>(baghouse) urban PM<br>collected in 1976 and 1977<br>(SRM 1649). Sequential<br>Soxhlet extraction<br>with DCM and MeOH,<br>fractionation on silica,<br>acid-base-neutral<br>fractionation | TA98, TA100, standard plate<br>incorporation assay, with and without<br>rat liver S9   | Higher potency (per µg of EOM or per µg of<br>PM) without S9, and DCM extracts more potent<br>compared with MeOH extracts. Highest activity<br>(92% of direct acting) in polar (MeOH) eluate.<br>Acid–base–neutral fractionation revealed strong<br>activity in acid fraction  | <u>Nishioka et al.</u><br>(1985)        |
| Washington, DC,<br>USA   | Time-integrated<br>(baghouse) urban PM<br>collected in 1976 and<br>1977 (SRM 1649). DCM<br>sonication extraction,<br>separate, sequential<br>extraction with Hx,<br>Hx:DEE (9:1), Hx:DEE<br>(1:1), MeOH      | TA98, TA100, standard plate<br>incorporation assay, with and without<br>Aroclor 1254-induced rat liver S9  | Stronger response on TA98 (per $\mu$ L of extract),<br>and clear increase in potency without S9. Clear<br>increase in potency with increasing solvent<br>polarity. PAHs, nitro-PAHs, and heterocyclic<br>compounds detected in DCM extract. Polar<br>extract contained some known mutagens, but<br>compounds that contribute to activity without S9<br>largely unknown | <u>Savard et al. (1992)</u>             |
| Washington, DC,<br>USA   | Time-integrated<br>(baghouse) urban<br>PM collected in 1976<br>and 1977 (SRM 1649).<br>Two sequential DCM<br>extractions followed by Ac<br>Soxhlet extraction  | TA98, TA100, standard plate<br>incorporation assay, with and without<br>Aroclor 1254-induced rat liver S9  | Addition of S9 increased activity of DCM extract<br>(per mg of PM) on TA98 and TA100. Substantial<br>fraction of total mutagenic activity associated<br>with Ac extract; 11–13% with S9, 20–21%<br>without S9  | <u>Nielsen (1992)</u>                   |
| Washington, DC,<br>USA   | Time-integrated<br>(baghouse) urban PM<br>collected in 1976 and<br>1977 (SRM 1649). DCM<br>sonication extraction   | TA98, TA100, microsuspension<br>assay, with and without Aroclor<br>1254-induced rat liver S9   | Highest potency (per μg of EOM) on TA98<br>without S9. No significant effect of S9 for TA100.<br>Microsuspension technique contributed to<br>3–9-fold increases in potency, relative to plate<br>incorporation   | <u>Agurell &amp; Stensman</u><br>(1992) |

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

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

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

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

#### Test article Geographical Salmonella strains<sup>a</sup>, 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) m<sup>3</sup>) for finest material (< 1 $\mu$ 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 m<sup>3</sup> 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 m<sup>3</sup>) 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 m<sup>3</sup>) with S9. Significant correlations between potency extraction (per m<sup>3</sup>) and lead in fine PM ( $< 2.5 \mu m$ ) as well as atmospheric NO<sub>2</sub>. 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 m<sup>3</sup>) 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/m<sup>3</sup> Diurnal analyses showed rapid changes (over TA98, TA98NR, standard plate Pitts et al. (1985) Los Angeles, PM<sub>10</sub> from 2 sites near San Diego Freeway, collected several hours) in mutagenic potency (per m<sup>3</sup>). 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 NO<sub>2</sub>. Maximum potency on TA98 with S9 ~140 rev/m3; without S9 ~140 rev/m3

| <b>Supplemental Table S5</b> | (continued) |
|------------------------------|-------------|
|------------------------------|-------------|

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

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

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

| Supplementa                            | in Table 55 (continued)   |  |  |  |
|--|---|--|--|--|
| Geographical location                  | Test article  | Salmonella strains <sup>a</sup> , assay version  | Results  | Reference  |
| Mexico City,<br>Mexico (1989–<br>1990) | PM <sub>10</sub> and TSP from 5<br>locations, collected on<br>GFFs with high-volume<br>sampler. MeOH Soxhlet<br>extraction              | TA98, TA100, plate incorporation<br>assay, with and without Aroclor<br>1254-induced rat liver S9 | Mutagenic potency (per m <sup>3</sup> ) generally higher for PM <sub>10</sub> compared with TSP; response significantly increased with S9  | <u>Villalobos-Pietrini</u><br><u>et al. (1995)</u> |
| Mexico City,<br>Mexico (1998)          | PM <sub>10</sub> from south-western<br>Mexico City, collected on<br>GFFs with high-volume<br>sampler. Sonication<br>extraction with DCM | TA98, YG1021, plate incorporation<br>assay, with and without rat liver S9                        | Mutagenic potency (per m <sup>3</sup> ) higher with S9 in<br>summer; opposite for colder months. Highest<br>potency during a dry period with many fires.<br>Significant increases on YG1021 relative to TA98<br>only for colder months with ground-based<br>inversions. Some positive associations between<br>mutagenic activity and atmospheric levels of CO,<br>NO <sub>2</sub> , and SO <sub>2</sub> ; negative association with rainfall.<br>Maximum potency on TA98 without S9 ~25 rev/m <sup>3</sup> ;<br>with S9 ~52 rev/m <sup>3</sup> | <u>Villalobos-Pietrini</u><br><u>et al. (2006)</u> |
| Mexico City,<br>Mexico (1990)          | Airborne PM from 2 urban<br>locations, collected on<br>GFFs with high-volume<br>sampler. DCM extraction<br>of shredded filter           | TA1538, TA98, YG1024, YG1021,<br>TA98NR, plate incorporation assay,<br>without rat liver S9      | Mutagenic potency (per m <sup>3</sup> ) increased on both<br>YG1021 and YG1024, and reduced on TA98NR,<br>relative to TA98. Maximum potency on TA98<br>without S9 ~21 rev/m <sup>3</sup>   | <u>Espinosa-Aguirre</u><br>et al. (1993)           |
| Mexico City,<br>Mexico (1993)          | $PM_{10}$ and TSP from a<br>residential area, collected<br>on GFFs. Sequential<br>Soxhlet extraction with<br>CX, DCM, and Ac            | TA98, plate incorporation assay, with and without rat liver S9                                   | Mutagenic potency (per m <sup>3</sup> ) highest for Ac extract and frequently higher with S9. TSP extracts frequently more potent than $PM_{10}$ extracts.<br>Maximum potency on TA98 without S9 ~4.7 rev/m <sup>3</sup> ; with S9 ~4.2 rev/m <sup>3</sup>   | <u>Amador-Muñoz</u><br><u>et al. (2001)</u>        |

<sup>a</sup> 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; NO<sub>2</sub>, nitrogen dioxide; NO<sub>3</sub>, nitrogen oxides; NR, nitroreductase; OAT, *O*-acetyltransferase; PACs, polycyclic aromatic compounds; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PM, particulate matter; PM<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10  $\mu$ m; PM<sub>2.5</sub>, particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ m; PUF, polyurethane foam; rev, revertants; SO<sub>2</sub>, 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<br>location                          | Test article  | Salmonella strains <sup>a</sup> /assay version  | Results   | Reference                    |
|---|---|---|---|------------------------------|
| Rio de Janeiro,<br>Brazil (1984)                  | Airborne PM collected<br>on GFFs with cascade<br>impactor. Sequential<br>Soxhlet extraction with<br>CX, DCM, and Ac   | TA98, TA100, standard plate<br>incorporation assay, with and without<br>Aroclor 1254-induced rat liver S9       | For Rio de Janeiro, potency higher (per $m^3$ or per $\mu$ 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 $\mu$ g of EOM) similar across sites. Maximum potency on TA98 without S9 ~11.8 rev/m <sup>3</sup> ; with S9 ~8.5 rev/m <sup>3</sup>   | <u>Miguel et al. (1990)</u>  |
| Rio Grande do<br>Sul state, Brazil<br>(2004–2005) | Airborne PM from<br>2 sites, collected on<br>GFFs with high-volume<br>sampler. DCM sonication<br>extraction   | TA98, TA100, YG1021, YG1024,<br>microsuspension assay, with and<br>without Aroclor 1254-induced rat<br>liver S9 | Elevated mutagenic potency (per m <sup>3</sup> ) at urban/<br>industrial site, relative to non-industrial reference.<br>PAH concentrations at urban/industrial site ~25-<br>fold higher than at non-industrial site. Potency<br>increased with S9. Potency for urban/industrial<br>sample (per µg of PM) elevated on both NR- and<br>OAT-enhanced strains. Maximum potency on<br>TA98 without S9 ~19 rev/m <sup>3</sup> ; with S9 ~32 rev/m <sup>3</sup>      | <u>Pereira et al. (2013)</u> |
| Rio Grande do<br>Sul state, Brazil<br>(2004–2005) | TSP from an urban/<br>industrial site and a<br>rural site, collected on<br>GFFs with high-volume<br>sampler. DCM sonication<br>extraction   | TA98, TA100, YG1021, YG1024,<br>microsuspension assay, with and<br>without Aroclor 1254-induced rat<br>liver S9 | Mutagenic potency (per $\mu$ g of EOM) increased on<br>YG1021 and YG1024, relative to TA98; generally<br>highest on YG1024. Potency (per m <sup>3</sup> ) substantially<br>higher for urban/industrial site. Maximum<br>potency on TA98 without S9 ~3 rev/m <sup>3</sup> ; with S9<br>~4 rev/m <sup>3</sup>   | Pereira et al. (2010)        |
| Rio Grande do<br>Sul state, Brazil<br>(2009–2010) | TSP and PM <sub>2.5</sub> from 2<br>urban/industrial sites,<br>collected on GFFs and<br>Teflon membrane filters<br>with high-volume sampler.<br>Sequential sonication<br>extraction with DCM and<br>water | TA98, YG1021, YG1024,<br>microsuspension assay, with and<br>without rat liver S9                                | Potency (per $\mu$ g of PM) of all DCM extracts<br>higher for PM <sub>2.5</sub> , relative to TSP. Water extracts all<br>negative for mutagenic activity. Enhanced activity<br>on metabolically enhanced strains, particularly<br>NR-enhanced strain YG1021. High temporal<br>variability in effect of S9 on TA98 mutagenic<br>potency (per m <sup>3</sup> ). Maximum potency on TA98<br>without S9 ~2.5 rev/m <sup>3</sup> ; with S9 ~2.3 rev/m <sup>3</sup> | <u>Lemos et al. (2012)</u>   |

#### Supplemental Table S6 (continued) Test article Geographical Salmonella strains<sup>a</sup>/assay version Results Reference location TA98, YG1021, YG1024, São Paulo, Brazil Airborne PM<sub>10</sub> from a Mutagenic potency (per m<sup>3</sup>) 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. (2008)(2004)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 m<sup>3</sup>) 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 m<sup>3</sup> without S9; ~120 rev/m<sup>3</sup> with S9. Reductions in potency on TA98NR and TA98/1,8-DNP, relative to TA98 São Paulo state, PM<sub>10</sub> from Araraquara, YG1024, pre-incubation assay, with Mutagenic potency (per m<sup>3</sup>) 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<br>location          | Test article   | Salmonella strains <sup>a</sup> /assay version  | Results  | Reference                                 |
|-----------------------------------|--|---|--|---|
| São Paulo state,<br>Brazil (2003) | PM <sub>10</sub> from Araraquara,<br>Piracicaba, and São Paulo<br>city, collected on GFFs<br>with high-volume sampler.<br>DCM Soxhlet extraction                     | TA98, TA100, YG1041, YG1042,<br>microsuspension assay, with and<br>without Aroclor 1254-induced rat liver<br>S9 | Mutagenic potency (per m <sup>3</sup> ) on TA98 higher<br>without S9; highest in areas affected by cane-<br>burning activities. Dramatic increase in potency<br>on YG1041 without S9, relative to TA98.<br>Maximum potency on TA98 ~320 rev/m <sup>3</sup> without<br>S9; ~57 rev/m <sup>3</sup> with S9 (cane-burning area);<br>~450 rev/m <sup>3</sup> without S9; ~140 rev/m <sup>3</sup> with S9 (São<br>Paulo City). Strong contributions from nitro-<br>PAHs and oxy-aromatics | <u>Umbuzeiro et al.</u><br>(2008)         |
| Santiago, Chile<br>(1996)         | $PM_{10}$ and TSP from an<br>urban, heavy-traffic site,<br>collected on Teflon-coated<br>filters and GFFs with high-<br>volume sampler. DCM<br>sonication extraction | TA98, plate incorporation assay, with<br>and without Aroclor 1254-induced rat<br>liver S9                       | Mutagenic potency varied during the year,<br>reached up to 700 rev/m <sup>3</sup> during summer (without<br>S9). Potency often elevated without S9. Maximum<br>potency on TA98 with S9 ~620 rev/m <sup>3</sup> . Potency<br>of TSP elevated relative to $PM_{10}$ . PAHs elevated<br>during summer months  | <u>Adonis &amp; Gil</u><br>( <u>2000)</u> |
| Santiago, Chile<br>(1990–1991)    | Airborne PM from an<br>urban, heavy-traffic site,<br>collected on GFFs with<br>high-volume sampler.<br>DCM sonication<br>extraction                                  | TA98, TA98NR, TA98/1,8-DNP $_6$ , plate incorporation assay, with and without rat liver S9                      | Mutagenic potency higher with S9, and potency<br>on TA98 reached maximum of 309 rev/m <sup>3</sup> .<br>Maximum potency on TA98 without S9 ~106 rev/m <sup>3</sup> .<br>Potency without S9 dramatically reduced on TA98NR<br>and TA98/1,8-DNP <sub>6</sub> compared with parent strain   | <u>Adonis &amp; Gil (1993)</u>            |
| Santiago, Chile<br>(1991)         | Airborne PM from an<br>urban, site, collected on<br>GFFs with high-volume<br>sampler. DCM sonication<br>extraction   | TA98, plate incorporation assay, with<br>and without Aroclor 1254-induced rat<br>liver S9                       | Higher potency (per m <sup>3</sup> ) with S9. Maximum potency without S9 177 rev/m <sup>3</sup> ; with S9 166 rev/m <sup>3</sup>   | <u>Adonis et al. (1997)</u>               |
| Santiago, Chile<br>(1991)         | Airborne PM from<br>downtown area, collected<br>on GFFs with high-volume<br>sampler. DCM sonication<br>extraction  | TA98, plate incorporation assay, with<br>and without Aroclor 1254-induced rat<br>liver S9                       | Higher potency (per m <sup>3</sup> ) with S9 in winter;<br>opposite in summer. Corresponds with elevated<br>levels of PAHs in cold season. Maximum potency<br>with S9 ~300 rev/m <sup>3</sup> ; without S9 ~100 rev/m <sup>3</sup>   | <u>Gil &amp; Adonis (1996)</u>            |
| Santiago, Chile<br>(1994)         | Airborne PM from<br>downtown area and rural<br>control, collected on<br>Teflon filters with low-flow<br>sampler. DCM sonication<br>extraction                        | TA98, microsuspension assay, with and without rat liver S9  | Mutagenic potency (per m <sup>3</sup> ) at urban site higher<br>without S9; at rural site higher with S9. Maximum<br>potency with S9 ~2100 rev/m <sup>3</sup> ; without S9<br>~2800 rev/m <sup>3</sup>   | <u>Gil et al. (1997)</u>                  |

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

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

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

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

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

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

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

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

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

| Geographical<br>location                    | Test article   | Salmonella strainsª/assay<br>version   | Results  | Reference  |
|---|--|--|--|--|
| Berlin,<br>Germany<br>(1983–1985)           | Airborne PM from 2 urban sites<br>and a highway tunnel, collected<br>with cascade impactor. CX:DEE<br>(2:8) sonication extraction,<br>acid-base-neutral and TLC<br>fractionation | TA98, TA98NR, plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9   | For urban sites and highway tunnel, mutagenic potency (per $\mu$ g 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 &amp;</u><br><u>Rüden (1988)</u>          |
| Berlin,<br>Germany<br>(1981–1982)           | Airborne PM collected with<br>cascade impactor. DEE:BZ (9:1)<br>sonication extraction  | TA98, TA100, plate<br>incorporation assay, with<br>and without rat liver S9  | Mutagenic potency (per m <sup>3</sup> ) of $0.4-0.7 \mu m$ size fraction elevated<br>with S9; higher in autumn and winter compared with spring<br>and summer. Maximum potency on TA98 with S9 ~22 rev/m <sup>3</sup> ;<br>without S9 ~11 rev/m <sup>3</sup>  | <u>Wullenweber</u><br>et al. (1982,<br><u>1984</u> ) |
| Berlin,<br>Germany<br>(1981)                | Airborne PM collected with<br>cascade impactor. DEE<br>sonication extraction, polar<br>organic and PAH fractions   | TA98, TA100, plate<br>incorporation assay, with<br>and without rat liver S9  | Mutagenic potency (per $\mu$ g of EOM) much higher for polar<br>aromatics compared with PAH fraction. Polar aromatics slightly<br>elevated without S9; PAH fraction higher with S9. Higher<br>mutagenic activity for smaller particles (0.4–1.3 $\mu$ m), compared<br>with particles of 1.3–10.2 $\mu$ m. Maximum potency on TA98 with<br>S9 ~8 rev/m <sup>3</sup> ; without S9 ~6 rev/m <sup>3</sup>                                  | <u>Moriske et al.</u><br>( <u>1982)</u>              |
| Berlin,<br>Germany<br>(1981–1982)           | Airborne PM collected with<br>cascade impactor. DEE:BZ (9:1)<br>sonication extraction, acid-<br>base-neutral fractionation   | TA98, plate incorporation<br>assay, with and without rat<br>liver S9   | Mutagenic potency (per m <sup>3</sup> ) of ultrafine PM (< $0.4 \mu$ m)<br>marginally higher without S9; significantly elevated in autumn<br>and winter compared with summer. Some indication of reduced<br>mutagenicity on weekends. Substantial portion of mutagenic<br>activity in acidic (18–31%) and neutral (41–66%) fractions.<br>Maximum potency on TA98 with S9 ~15 rev/m <sup>3</sup> ; without S9<br>~17 rev/m <sup>3</sup> | <u>Gottlieb et al.</u><br>( <u>1983)</u>             |
| Herne,<br>Germany<br>(1977)                 | Airborne PM from urban site<br>in Ruhr area, collected on<br>polystyrene or cellulose nitrate<br>filters. DCM or DEE extraction  | TA98, plate incorporation<br>assay, with and without<br>PCB-induced mouse liver<br>S9  | Mutagenic potency (per $\mu$ 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><br><u>al. (1980)</u>              |
| Baden-<br>Württemberg,<br>Germany<br>(1995) | Airborne PM from an urban,<br>site, collected on GFFs with<br>high-volume sampler. Ac<br>Soxhlet extraction  | Plate incorporation assay<br>with Ames II base-pair<br>substitution strains<br>TA7001, TA7002, TA7003,<br>TA7004, TA7005, TA7006,<br>without S9 activation | Mutagenic potency (per m <sup>3</sup> ) 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><br>(2004)                     |

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

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

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

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

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

| Geographical<br>location               | Test article  | Salmonella strainsª/assay<br>version  | Results   | Reference                        |
|--|---|---|---|----------------------------------|
| Athens,<br>Greece (1984)               | TSP from 4 urban sites and 1<br>rural site, collected on GFFs<br>with cascade impactor. Soxhlet<br>extraction with BZ or CX   | TA98, standard plate<br>incorporation assay,<br>without S9  | Highest potency (per m <sup>3</sup> ) for city centre locations, and increase<br>in winter relative to summer. Correlation between mutagenic<br>potency without S9 and B[ <i>a</i> ]P concentration. Maximum potency<br>~35 rev/m <sup>3</sup> . Largest fraction of mutagenic activity associated<br>with PM < 1.1 $\mu$ m. Reduced potency (per m <sup>3</sup> ) at increased<br>sampling heights | <u>Viras et al.</u><br>(1990)    |
| Paris, France<br>(1979–1980)           | Airborne PM from 2 urban sites,<br>collected on GFFs or Teflon<br>filters. Sequential extractions<br>with Chl and Ac, with agitation,<br>or Chl extraction with agitation<br>and sonication | TA1535, TA1537, TA1538,<br>TA98, TA100, plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9                                    | Highest potency (per m <sup>3</sup> ) on frameshift strains without S9. Maximum potency on TA98 without S9 ~190 rev/m <sup>3</sup> ; with S9 ~40 rev/m <sup>3</sup>   | <u>Courtois et al.</u><br>(1981) |
| Paris, France<br>(1983–1985)           | Airborne PM from an urban<br>site, collected on GFFs with<br>high-volume sampler. DCM or<br>Ac sonication extraction  | TA1538, TA98, TA1537,<br>TA97, TA1535, TA100,<br>TA102, TA98NR, plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9            | Mutagenic potency (per $\mu$ g of EOM) on TA98 and TA100<br>increased with S9 for winter samples; S9 effect much lower or<br>negligible for spring samples. Decrease in activity on TA98NR,<br>relative to TA98. Maximum potency on TA98 without S9<br>~60 rev/m <sup>3</sup> ; with S9 ~100 rev/m <sup>3</sup>   | <u>Courtois et al.</u><br>(1988) |
| Paris, France<br>(1980–1981)           | Size-fractionated PM from<br>central Paris, collected with<br>cascade impactor. ACN<br>extraction   | TA98, TA98NR, plate<br>incorporation assay, with<br>and without Aroclor<br>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 $\mu$ m. 75% of activity associated with PM < 1.1 $\mu$ m, 90% with PM < 2 $\mu$ m. Strong decrease in mutagenicity of TA98NR, relative to TA98   | <u>Festy et al.</u><br>(1984)    |
| Paris region,<br>France<br>(1978–1979) | Airborne PM from 5 sites in city<br>centre and suburbs, collected<br>on Teflon filters. Agitation<br>and sonication with Chl, or<br>sequential extraction with Chl<br>and Ac                | TA98, TA100, plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9   | Highest potency (per mg of PM) without S9, and substantial<br>elevations in winter months. Close correspondence between<br>mutagenic potency and PAH concentration  | <u>Festy (1980)</u>              |
| Dunkirk,<br>France                     | Airborne PM, collected with<br>cascade impactor. Organic<br>fraction thermally desorbed   | TA98, YG1041, plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver<br>S9, suspensions of PM or<br>thermally desorbed PM in<br>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><br>(2011)    |
| Geographical location               | Test article   | Salmonella strainsª/assay<br>version  | Results  | Reference                        |
|-------------------------------------|--|---|--|----------------------------------|
| Barcelona,<br>Spain (1989–<br>1990) | Airborne PM from 2 urban<br>locations, collected on GFFs<br>with high-volume sampler.<br>DCM sonication extraction,<br>fraction by GPC   | TA98, standard plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9   | Mutagenic potency (per m <sup>3</sup> ) 13–32% higher with S9; similar<br>potency for PM from 2 locations. Maximum potency with<br>S9 ~100 rev/m <sup>3</sup> ; without S9 ~78 rev/m <sup>3</sup> . Potency without<br>S9 corresponds with levels of nitroarenes; activity with S9<br>corresponds with levels of aromatic ketones and quinones.<br>Seasonal variation in ketones, quinones, and lactones indicate<br>strong contribution of atmospheric transformation processes | <u>Bayona et al.</u><br>(1994)   |
| Barcelona,<br>Spain (1990)          | Airborne PM from urban<br>locations, collected on GFFs<br>with high-volume sampler.<br>DCM sonication extraction,<br>fraction by GPC and<br>subfractionation                   | TA98, TA98NR, TA98/1,8-<br>DNP <sub>6</sub> , standard plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9 | Mutagenic potency higher without S9. Several potent fractions reduced on TA98NR and TA98/1,8-DNP <sub>6</sub> , relative to TA98. Active fractions contain PAHs, nitro-PAHs, aromatic ketones and quinones, and N-heterocyclics. Maximum potency with S9 $\sim$ 45 rev/m <sup>3</sup> ; without S9 $\sim$ 100 rev/m <sup>3</sup>   | <u>Casellas et al.</u><br>(1995) |
| Caserta, Italy                      | Atmospheric contamination at<br>5 locations in Caserta affected<br>by different levels of vehicle<br>traffic, in situ 3 wk deployments<br>of semipermeable membrane<br>devices | Reverse mutation of dark<br>mutants of <i>Vibrio fischeri</i> ,<br>with and without rat liver<br>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><br>(2003)  |

<sup>a</sup> 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; NO<sub>2</sub>, 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  $\mu$ m;  $PM_{2.5}$ , particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ m; PUF, polyurethane foam; rev, revertants; SO<sub>2</sub>, 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<br>location     | Test article  | Salmonella strainsª/assay<br>version  | Results  | Reference                                     |
|------------------------------|---|---|--|---|
| Mumbai, India                | Airborne PM (> 0.8 μm) collected<br>on GFFs. DMSO or water<br>extraction using mortar and<br>pestle                                       | TA1537, TA1535, TA1538,<br>TA100, TA98, standard<br>plate incorporation assay,<br>with and without Aroclor<br>1254-induced rat liver S9 | Only TA100 results recorded. Highest potency (per $\mu$ L of extract) for DMSO extracts with S9. Highest response for site with high population density, traffic, and industrialization  | <u>Shenoy &amp; Chaubal</u><br>(1984)         |
| Agra, India<br>(2010)        | $PM_{10}$ and $PM_{2.5}$ from a single site, collected on GFFs. DCM sonication extraction   | TA98, TA100, plate<br>incorporation assay,<br>without rat liver S9  | Weak responses with modest increase for $\mathrm{PM}_{2.5}$ relative to $\mathrm{PM}_{10}$   | <u>Singla et al. (2012)</u>                   |
| Tokyo, Japan<br>(1988, 1990) | Airborne PM from an urban site<br>during winter, collected on GFFs<br>with high-volume sampler. DCM<br>sonication extraction              | TA98, TA100, YG1024,<br>YG1029, pre-incubation<br>assay and Spiral assay,<br>without S9 activation                                      | Mutagenic activity higher with Spiral assay; enhanced<br>on YG1024 and YG1029 compared with parent strains.<br>Maximum potency on TA98 ~450 rev/m <sup>3</sup> for Spiral and<br>318 rev/m <sup>3</sup> for pre-incubation   | <u>Houk et al. (1992)</u>                     |
| Tokyo, Japan<br>(1980–1981)  | Airborne PM, collected on GFFs<br>with high-volume sampler.<br>BZ:EtOH (4:1) sonication<br>extraction, acid-base-neutral<br>fractionation | TA98, TA100, pre-<br>incubation assay, with and<br>without PCB-induced rat<br>liver S9  | Mutagenic potency (per mg of EOM or m <sup>3</sup> ) on TA98<br>and TA100 higher with S9; highest activity for neutral<br>fraction. Potency on TA98 without S9 ~11 rev/m <sup>3</sup> ; with<br>S9 ~12 rev/m <sup>3</sup> . Neutral fraction induced tumours in ICR<br>mice  | <u>Sasaki et al. (1987)</u>                   |
| Tokyo, Japan<br>(1983)       | Airborne PM <sub>10</sub> , collected on<br>GFFs with high-volume sampler.<br>MeOH extraction and extract<br>fractionation                | TA98 pre-incubation assay,<br>with and without PCB-<br>induced rat liver S9   | Potency (per $\mu g$ of EOM) higher without S9 activation. Highest potency for crude MeOH extract and NM fraction  | <u>Sakitani &amp; Suzuki</u><br><u>(1986)</u> |
| Tokyo, Japan<br>(1978–1979)  | Airborne PM, collected on GFFs<br>with high-volume sampler. NM<br>extraction  | TA98, pre-incubation assay,<br>with and without rat liver<br>S9   | Mutagenic potency (per m <sup>3</sup> ) higher with S9; elevated in<br>autumn compared with spring and summer. With S9,<br>daytime higher than night-time, but only for summer<br>and autumn. Maximum potency without S9 ~8 rev/m <sup>3</sup> ;<br>with S9 ~16 rev/m <sup>3</sup>   | <u>Sakitani &amp;</u><br>Hayashi (1986)       |
| Tokyo, Japan                 | Airborne PM, collected on GFFs<br>with high-volume sampler. MeOH<br>extraction, solvent fractionation                                     | TA98, pre-incubation assay,<br>with and without rat liver<br>S9   | Mutagenic potency (per µg of EOM) higher without S9;<br>highest activity in NM fraction, compared with CX. No<br>response for MeOH/water fraction  | <u>Sakitani &amp; Suzuki</u><br><u>(1986)</u> |
| Tokyo, Japan<br>(1978–1979)  | Airborne PM from city centre,<br>collected on GFFs with high-<br>volume sampler. MeOH<br>extraction                                       | TA98, pre-incubation assay,<br>with PCB-induced rat liver<br>S9   | Mutagenic potency (per m <sup>3</sup> ) frequently higher during<br>the day, especially in autumn. Seasonal average potency<br>higher without S9; progressive potency decline from<br>winter to autumn, to summer and spring. Some<br>correlation between mutagenic activity and B[ <i>a</i> ]P<br>concentration. Maximum potency without S9 ~10 rev/m <sup>3</sup> ;<br>with S9 ~8 rev/m <sup>3</sup> | <u>Shimizu et al.</u><br>(1983)               |

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

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

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

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

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

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

| Geographical location             | Test article   | Salmonella strains²/assay<br>version  | Results   | Reference                              |
|-----------------------------------|--|---|---|--|
| Shanghai,<br>China<br>(1992–1993) | Airborne PM from 13 urban sites,<br>collected on GFFs with high-<br>volume sampler. DCM sonication<br>extraction, acid-base-neutral<br>fractionation | TA98, TA100, plate<br>incorporation assay, with<br>and without Aroclor<br>1254-induced rat liver S9 | No significant responses on TA100. Mutagenic potency<br>on TA98 generally higher without S9, and substantially<br>increased in winter. Substantial temporal and spatial<br>variation in potency (per m <sup>3</sup> ). Maximum potency on<br>TA98 without S9 ~20 rev/m <sup>3</sup> ; with S9 ~25 rev/m <sup>3</sup> . With<br>S9, high activity in basic and polar fraction; without S9,<br>high activity in numerous fractions, including acidic,<br>basic, neutral, and aromatic | <u>Zhao et al. (2002)</u>              |
| Shanghai,<br>China                | Airborne PM from residential and<br>industrial locations affected by<br>coking facility. NM extraction   | TA98, TA100, plate<br>incorporation assay, with<br>and without S9                                   | Mutagenic potency (per m <sup>3</sup> ) higher without S9; elevated<br>in residential and industrial areas when coke oven<br>in operation (10–23-fold above control site). When<br>coke oven not operating, 3-fold decrease in potency.<br>Maximum potency on TA98 without S9 (factory<br>entrance) ~2600 rev/m <sup>3</sup> ; with S9 ~2000 rev/m <sup>3</sup>   | <u>Yu et al. (1989)</u>                |
| Shanghai,<br>China                | TSP from 10 representative areas.<br>NM sonication extraction  | TA98, TA100, plate<br>incorporation assay, with<br>and without S9                                   | Mutagenic potency (per m <sup>3</sup> ) higher without S9; similar<br>results for TA98 and TA100. Maximum potency on TA98<br>without S9 ~260 rev/m <sup>3</sup>   | <u>Zhu et al. (1991)</u>               |
| Shanghai,<br>China (1988)         | TSP from 5 representative areas.<br>NM sonication extraction; extract<br>separated into 5 fractions  | TA98, TA100, plate<br>incorporation assay, with<br>and without S9                                   | Mutagenic potency (per m <sup>3</sup> ) marginally higher without<br>S9. Maximum potency on TA98 without S9 ~205 rev/m <sup>3</sup> .<br>One polar fraction accounted for up to 86% of mutagenic<br>activity. PAH-containing fraction accounted for only<br>2.6–11% of mutagenicity   | <u>Zhu et al. (1990)</u>               |
| Shanghai,<br>China                | TSP from 13 representative areas.<br>DCM sonication extraction   | TA98, TA100, plate<br>incorporation assay, with<br>and without S9                                   | Positive response for all sites, including control<br>(suburban park). Higher response without S9; higher<br>activity at industrial and commercial sites with heavy<br>vehicle traffic  | <u>Zhao et al. (1996)</u>              |
| Shanghai,<br>China                | TSP from 4 commercial and<br>suburban areas. DCM sonication<br>extraction; extract separated into<br>5 fractions                                     | TA98, plate incorporation assay, with and without S9  | Mutagenic activity generally higher with S9, and higher<br>in winter compared with summer. Strong responses for<br>fractions containing PAHs and polar aromatics  | <u>Zhao &amp; Zhu</u><br><u>(1997)</u> |
| Beijing, China<br>(1981)          | TSP from 1 commercial site, 2<br>residential sites, and control site.<br>BZ Soxhlet extraction   | TA98, plate incorporation assay, without S9   | Mutagenic potency (per m <sup>3</sup> ) highest at commercial<br>site, followed by residential site. Negative at control.<br>Maximum potency TA98 ~2.3 rev/m <sup>3</sup>   | <u>Chen et al. (1982)</u>              |
| Beijing, China<br>(1982)          | TSP from 1 commercial site, 2<br>residential sites, and 1 industrial<br>site. BZ Soxhlet extraction  | TA97, TA98, plate<br>incorporation assay, with<br>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<br>location                  | Test article  | Salmonella strainsª/assay<br>version  | Results   | Reference                                     |
|---|---|---|---|---|
| Beijing, China<br>(1984)                  | TSP from downtown and<br>industrial area. BZ:EtOH (3:1)<br>Soxhlet extraction   | TA100, TA98, plate<br>incorporation assay, with<br>and without S9                   | Higher potency (per mg of EOM) in downtown area,<br>relative to site with petrochemical industry. Lower or<br>equivalent potency with S9  | <u>Chen et al. (1987)</u>                     |
| Beijing, China<br>(1980)                  | TSP from 7 urban, suburban, and<br>industrial sites. BZ:MeOH (4:1)<br>Soxhlet extraction, fractionation<br>on silica                          | TA98, plate incorporation assay, with and without S9                                | Mutagenic potency (per m <sup>3</sup> ) highest at downtown (urban)<br>site and lowest at suburban site. Higher response with S9.<br>Fractionation showed highest S9-activated response in<br>PAH-containing fractions and highest response without<br>S9 in polar fractions. Maximum potency on TA98 with<br>S9 ~66 rev/m <sup>3</sup> | <u>Zhao et al. (1983)</u>                     |
| Beijing, China<br>(2005)                  | $PM_{10}$ and $PM_{2.5}$ from urban/<br>commercial and industrial<br>sites. Chl Soxhlet extraction,<br>fractionation on silica                | TA98, plate incorporation assay, with and without S9                                | Higher mutagenic activity with S9, and higher in<br>industrial area. Winter samples more potent than<br>summer samples for industrial area only   | <u>Che et al. (2008)</u>                      |
| Several cities<br>in China<br>(1983–1984) | TSP from residential, industrial,<br>and suburban sites in Shenyang,<br>Guangzhou, Xi'an, Beijing,<br>and Shanghai. DCM Soxhlet<br>extraction | TA98, plate incorporation assay, with and without S9                                | Mutagenic activity higher in winter compared with<br>summer. Potency (per mg of PM) similar across 5<br>cities. Highest potency (per m <sup>3</sup> ) in Shenyang in winter.<br>Maximum potency on TA98 ~90 rev/m <sup>3</sup>  | <u>Li et al. (1985)</u>                       |
| Shenyang,<br>China (1990)                 | TSP from 3 residential, industrial,<br>and commercial sites, plus<br>control. DCM Soxhlet extraction  | TA98, TA98NR, TA98/1,8-<br>DNP <sub>6</sub> , plate incorporation assay, without S9 | Significant mutagenicity at all sites, including control.<br>Reduced responses on enzymatically deficient strains<br>without S9 confirm contributions from nitro-PAHs.<br>Similar potency (per mg of PM) across all sites   | <u>Kong et al. (1994)</u>                     |
| Shenyang,<br>China (1990)                 | TSP from 3 residential, industrial,<br>and commercial sites, plus<br>control. DCM Soxhlet extraction  | TA98, YG1021, YG1024,<br>plate incorporation assay,<br>without S9                   | Significant mutagenicity at all sites, including control.<br>Enhanced responses on enzymatically enhanced strains<br>without S9 confirm contributions from nitro-PAHs   | <u>Kong et al. (1995)</u>                     |
| Shenyang,<br>China (2000–<br>2002)        | TSP from 4 urban, industrial, and suburban sites. DCM sonication extraction   | TA98, TA100, plate<br>incorporation assay, with<br>and without S9                   | Positive responses for all sites, and higher mutagenicity<br>with S9. Similar potency (per mg of PM) across sites<br>investigated   | <u>Piao et al. (2007)</u>                     |
| Guangzhou,<br>China (1991)                | TSP. DCM Soxhlet extraction   | TA98, plate incorporation assay, without S9   | Mutagenic potency (per m <sup>3</sup> ) higher in February (winter)<br>and lowest in June (summer). Maximum potency on<br>TA98 without S9 ~23 rev/m <sup>3</sup>  | Qian & Zhang<br>(1997), Qian et al.<br>(1997) |
| Guangzhou,<br>China (2002)                | TSP, collected with cascade<br>impactor. Sonication extraction<br>with DCM  | TA98, plate incorporation assay, with and without S9                                | Mutagenic potency (per mg of PM) increased with decreasing PM size; maximum for PM < 0.5 $\mu$ m. Higher potency without S9. Potency (per m <sup>3</sup> ) significantly higher for smaller PM size fraction  | <u>Li et al. (2005)</u>                       |

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

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

<sup>a</sup> 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<sub>s</sub>, nitrogen oxides; NR, nitroreductase; OAT, *O*-acetyltransferase; PAC, polycyclic aromatic compounds; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PCB, polychlorinated biphenyl; PM, particulate matter; PM<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10 μm; PM<sub>2.5</sub>, 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 strainsª/assay version                        | Results   | Reference                       |
|---|--|--|---|---------------------------------|
| South Island,<br>New Zealand<br>(2001–2002) | PM <sub>10</sub> from 3 urban and<br>residential locations,<br>collected on quartz<br>filters with high-volume<br>sampler. Sequential Soxhlet<br>extraction with DCM and<br>MeOH | TA98, plate incorporation assay,<br>without rat liver S9 | Mutagenic potency (per m <sup>3</sup> ) substantially higher in<br>winter, and elevated in urban areas. 74% of winter<br>samples elicited positive responses; 25% of summer<br>samples. Maximum potency on TA98 ~11 rev/m <sup>3</sup>  | <u>Brown et al.</u><br>(2005)   |
| North Island,<br>New Zealand<br>(2005)      | PM <sub>10</sub> and PM <sub>2.5</sub> from 2<br>residential locations,<br>collected on GFFs or<br>quartz filters with high-<br>volume sampler. DCM<br>Soxhlet extraction        | TA98, fluctuation assay, without S9 activation           | Substantial increases in potency (per m <sup>3</sup> ) in winter;<br>no significant seasonal difference in potency (per<br>$\mu$ g of EOM). Potency higher for PM <sub>2.5</sub> , compared<br>with PM <sub>10</sub> . Potency significantly correlated with<br>PAH levels. Estimated that wood smoke made<br>larger contribution than mobile sources. Maximum<br>potency on ~30 rev/m <sup>3</sup> | <u>Cavanagh et</u><br>al.(2009) |

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

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

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

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

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  $\mu$ m;  $PM_{2.5}$ , particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ 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<br>Prague (Czech Republic), Košice<br>(Slovakia), and Sofia (Bulgaria)   | Human hepatoma HepG2 cells,<br>human diploid lung fibroblasts HEL,<br>and human monocytic leukaemia<br>cell line THP-1 | <sup>32</sup> P-postlabelling | Winter samples: Prague > Sofia<br>> Košice. Summer samples: Košice<br>> Sofia > Prague  | <u>Sevastyanova et al. (2007)</u> |
| Extracts of SRM 1649a dust sample   | Rat liver epithelial WB-F344 cells   | <sup>32</sup> P-postlabelling | Crude extracts and non-polar<br>fractions gave rise to DNA adducts;<br>polar fraction did not   | <u>Andrysík et al. (2011)</u>     |
| Extracts of air particles from<br>Teplice and Prachatice, Czech<br>Republic                           | Cultured rat hepatocytes and<br>Chinese hamster lung V79NH cells   | <sup>32</sup> P-postlabelling | Highest DNA binding activity in<br>fractions containing PAHs and<br>nitro-PAHs  | <u>Topinka et al. (2000)</u>      |
| Extracts of air particles from<br>Teplice, Czech Republic   | Calf thymus DNA with and without S9 liver fraction   | <sup>32</sup> P-postlabelling | Highest adduct formation with neutral (aromatic) fraction   | <u>Binková et al. (1998)</u>      |
| Extracts of air particles from<br>Teplice and Prachatice, Czech<br>Republic                           | Calf thymus DNA with and without<br>S9 liver fraction  | <sup>32</sup> P-postlabelling | Seasonal differences significant<br>(with and without S9). Location<br>difference significant (without S9).<br>DNA adducts were mainly PAH-<br>like | <u>Binková et al. (1999)</u>      |
| Extracts of air particles from<br>Prague city centre and suburban<br>area and Teplice, Czech Republic | Calf thymus DNA with and without S9 liver fraction   | <sup>32</sup> P-postlabelling | Winter > summer for all 3<br>locations. Differences between<br>locations not significant  | <u>Binková et al. (2003)</u>      |
| Extracts of air particles from<br>Ostrava region and Třeboň,<br>Czech Republic                        | Calf thymus DNA with and without<br>S9 liver fraction  | <sup>32</sup> P-postlabelling | DNA adduct-forming activity of<br>Ostrava (industrial) > Ostrava<br>(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.

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

| Particles  | Substrate      | Extraction            | Method                   | Effect  | Reference                      |
|--|----------------|-----------------------|--------------------------|---|--------------------------------|
| PM <sub>2.5</sub> from urban or<br>suburban sites in Shanghai,<br>China  | Plasmid<br>DNA | Water<br>(vortexing)  | Supercoil relaxation     | Samples from urban sites most potent. Seasonal variation<br>in potency (winter more potent than summer)   | <u>Senlin et al.</u><br>(2008) |
| PM <sub>2.5</sub> and PM <sub>10</sub> samples<br>collected at Beijing, Nankou<br>town, and a clean site,<br>Shisanling Reservoir, China | Plasmid<br>DNA | Water<br>(sonication) | Supercoil<br>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 <sub>10</sub> samples from Lanzhou<br>city and a suburban area in<br>China during 4 seasons   | Plasmid<br>DNA | Water<br>(sonication) | Supercoil<br>relaxation  | Average values of TD20 (level causing 20% of SB) were 17,<br>625, 56, and 260 $\mu$ g/mL in winter, spring, summer, and<br>autumn, respectively, for PM <sub>10</sub> suspension solution from<br>Lanzhou city. Water extracts caused slightly lower level of SB<br>with higher TD20. Suburban PM <sub>10</sub> samples showed higher<br>TD20 than Lanzhou sample. Particles collected during dust<br>storm episodes or after days with rain showed lower SB<br>generation activity with TD20 > 1000 $\mu$ g/mL. TD20 values<br>were negatively correlated with metal concentration | <u>Xiao et al.</u><br>(2009)   |
| PM <sub>10</sub> samples collected at<br>3 sites in Macao Special<br>Administrative Region,<br>China                                     | Plasmid<br>DNA | Water<br>(sonication) | Superoxide<br>relaxation | TD30 values (level causing 30% of SB) were 3, 10, and 20 $\mu$ g/mL for whole PM <sub>10</sub> 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 $\mu$ g/mL for the 3 sites, respectively  | <u>Shen et al.</u><br>(2009)   |

DFO, deferoxamine; DMSO, dimethyl sulfoxide; PBS, phosphate-buffered saline; PM, particulate matter;  $PM_{10}$ , particulate matter with particles of aerodynamic diameter < 10  $\mu$ m;  $PM_{2.5}$ , particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ 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 <sub>10</sub> collected during a dust<br>storm from the roof of a<br>building in Incheon, Republic<br>of Korea | Mice                     | PBS                                     | 0.5 mg/mouse twice/wk<br>for 12 wk (total dose,<br>12 mg/mouse), and<br>killed at 24 h after the<br>last exposure | 8-oxodG (immunohistochemistry) in lungs                        | <u>Hwang et al. (2010)</u>        |
| Intratracheal instillation of SRM 1649  | ApoE <sup>_/_</sup> mice | NA                                      | 0.5 mg/kg at 26 h and<br>2 h before being killed  | Unaltered levels of FPG-sensitive sites (comet) in lung tissue | <u>Vesterdal et al.</u><br>(2012) |
| Intratracheal instillation of<br>PM from a town with many<br>wood stoves and a rural area,<br>Denmark             | Rats                     | Mechanical<br>collection<br>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><br>(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<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10 μm; SRM, standard reference mixture; wk, week or weeks.

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

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| Particles  | Cells                            | Extraction   | Concentration and time                             | Effect  | Reference                                 |
|--|----------------------------------|--|--|---|---|
| Coal fly ash from a power plant<br>in Arnhem, Netherlands                  | RLE                              | Not specified  | 40 cm <sup>2</sup> /mL or<br>particles/mL for 18 h | Different potency of particles to generate 8-oxodG (HPLC-ECD)   | <u>van Maanen</u><br><u>et al. (1999)</u> |
| EOM of PM <sub>10</sub> from Teplice,<br>Czech Republic                    | HepG2                            | DCM  | 1–50 µg/mL for 24 h                                | Increased ENDOIII/FPG sites, although not concentration-dependent relationship  | <u>Lazarová &amp; Slamenová</u><br>(2004) |
| EOM of PM <sub>2.5</sub> 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><br>(2011)                |
| EOM of $PM_{10}$ from an industrial site in France                         | HepG2                            | DCM (Soxhlet<br>extractor)                                   | 12 μg/mL of B[ <i>a</i> ]P for<br>24 h             | Increased levels of 8-oxodG (HPLC-MS/MS)  | <u>Tarantini</u><br><u>et al. (2009)</u>  |
| EOM of PM <sub>10</sub> from Prague,<br>Czech Republic                     | HepG2<br>and lung<br>fibroblasts | DCM  | 1–100 mg/mL for<br>24–48 h                         | Increased levels of 8-oxodG (ELISA) in HepG2<br>cells, but not in lung fibroblasts. Little variation in<br>genotoxicity between different seasons | <u>Hanzalova</u><br><u>et al. (2010)</u>  |
| EOM of PM <sub>10</sub> from Kaifaqu<br>district, Dalian, China            | HepG2                            | Sequential<br>extraction in<br>DCM, acetone, and<br>methanol | 7.5–30 mg/mL for 1 h                               | Concentration-dependent increase in 8-oxodG<br>(antibody-based) of samples from Kaifaqu<br>(industrial area)                                      | <u>Jiang et al.</u><br>(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; PM<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10  $\mu$ m; PM<sub>2.5</sub>, particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ 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-<br>oxidant                                | Extraction                            | Method                       | Effect   | Reference                                |
|--|--|---------------------------------------|------------------------------|--|--|
| Coarse and fine particles<br>from locations in North<br>Rhine-Westphalia, Germany  | Calf thymus<br>DNA with<br>H <sub>2</sub> O <sub>2</sub> | Water<br>(sonication)                 | 8-oxodG (dot blot)           | Higher generation of 8-oxodG by PM from Duisburg than<br>from rural location. Coarse and fine particles generated<br>the same level of 8-oxodG   | <u>Shi et al.</u><br>(2006)              |
| Coarse and fine particles<br>from Düsseldorf, Germany  | Calf thymus<br>DNA with<br>H <sub>2</sub> O <sub>2</sub> | Water<br>(sonication)                 | 8-oxodG (dot blot)           | Coarse PM generated more 8-oxodG than fine PM when compared at equal mass  | <u>Shi et al.</u><br>(2003)              |
| TSP from a busy street in<br>Copenhagen, Denmark   | Calf thymus<br>DNA with<br>H <sub>2</sub> O <sub>2</sub> | Water                                 | 8-oxodG (HPLC)               | Concentration-dependent increase, but no difference<br>between PM sampled on different days  | <u>Danielsen et</u><br><u>al. (2008)</u> |
| PM <sub>10</sub> from Helsinki, Finland,<br>collected during spring or<br>winter   | Calf thymus<br>DNA with<br>H <sub>2</sub> O <sub>2</sub> | Water and<br>ethanol<br>(sonication)  | 8-oxodG (immuno<br>dot blot) | Concentration-dependent increase, without clear seasonal variation   | <u>Salonen et</u><br><u>al. (2004)</u>   |
| Oil fly ash (from an<br>electrostatic precipitator at a<br>power plant in Niagara, New<br>York, USA) and coal fly ash<br>(from a power plant in USA) | dG or calf<br>thymus DNA                                 | Electrostatic<br>precipitator         | 8-oxodG (HPLC-<br>ECD)       | Oil fly ash increased production of 8-oxodG, whereas coal<br>fly ash did not. Reduced 8-oxodG production by DFO  | <u>Prahalad et</u><br><u>al. (2000)</u>  |
| SRM 1649, urban air dust<br>from Düsseldorf, Germany,<br>Arizona desert dust, and<br>ROFA  | Calf thymus<br>DNA                                       | Baghouse                              | 8-oxodG (HPLC-<br>ECD)       | Moderately increased production of 8-oxodG by SRM<br>1649 and urban air dust. No effect of Arizona desert dust<br>and coal fly ash. Strong effect of ROFA and oil fly ash.<br>Reduced effect by pre-treatment with catalase or DFO<br>for ROFA and oil fly ash, whereas no protection against<br>8-oxodG generation by SRM 1649 and Düsseldorf particles | <u>Prahalad et</u><br><u>al. (2001)</u>  |
| SRM 1649 or its particles<br>after extraction with organic<br>solvents   | dG   | DCM, hexane,<br>acetone, or<br>DMSO   | 8-oxodG (HPLC)               | Increased 8-oxodG production in water and lower production after extraction of organic material  | <u>Karlsson et</u><br><u>al. (2004)</u>  |
| $PM_{10}$ from an urban street in Stockholm, Sweden  | dG with $H_2O_2$   | Water<br>(vortexing or<br>sonication) | 8-oxodG (HPLC-<br>ECD)       | Increased only in the presence of $\rm H_2O_2$   | <u>Karlsson et</u><br><u>al. (2005)</u>  |
| Fine particles from<br>Duisburg, Germany   | Calf thymus<br>DNA with<br>H <sub>2</sub> O <sub>2</sub> | Water<br>(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><br><u>al. (2002)</u>   |
| TSP, PM <sub>10</sub> , and PM <sub>2.5</sub> from Athens, Greece  | dG with $H_2O_2$   | Water<br>(sonication)                 | 8-oxodG (HPLC-<br>ECD)       | $PM_{2.5}, PM_{10}, and TSP generated 8-oxodG (8-oxodG level reported in the unusual unit of \mu g lesions per 10^6 dG)$   | <u>Valavanidis</u><br>et al. (2005)      |

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

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

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

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

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

| Supplemental Table S17 | (continued)  |
|------------------------|--------------|
| Suppremental Table SI/ | (contentaca) |

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

| Geographical location        | Test article   | Assay/exposure system  | End-point(s)<br>examined  | Results  | Reference                               |
|------------------------------|--|--|---|--|---|
| Taiyuan,<br>China (1991)     | TSP samples from a residential<br>area. Nitric acid or SLF<br>sonication extraction                                      | Exposure of human<br>amnion cells for 5 h to<br>acid or SLF extracts | Unscheduled<br>DNA synthesis<br>(incorporation<br>of radio-labelled<br>thymidine) | Significant dose-related increase in DNA<br>damage for 0.5 m <sup>3</sup> equiv of acid extract<br>and 21 m <sup>3</sup> equiv of SLF extract. Lead, zinc,<br>chromium, manganese, and nickel suspected as<br>contributors to DNA damage response  | <u>Yuan &amp; Xun</u><br>(1994)         |
| Taiyuan,<br>China (1987)     | Size-fractionated TSP from a<br>residential area. Nitric acid or<br>SLF sonication extraction                            | Exposure of human<br>amnion cells for 5 h to<br>acid or SLF extracts | Unscheduled<br>DNA synthesis<br>(incorporation<br>of radio-labelled<br>thymidine) | Significant dose-related increase in DNA damage for all acid extracts. Significant DNA damage response for SLF extracts of PM < 2 $\mu$ m only. Extracts of smaller size classes induced responses   | <u>Yuan et al.</u><br>(1994)            |
| Shanghai,<br>China           | TSP from 13 locations. NM sonication extraction  | Exposure of primary<br>hepatocytes from SD<br>rats                   | Unscheduled<br>DNA synthesis<br>(incorporation<br>of radio-labelled<br>thymidine) | Significant dose-related increases in DNA<br>damage response induced by most TSP extracts.<br>Winter samples induced a stronger response<br>relative to summer samples   | <u>Zhao &amp; Zhu</u><br>( <u>1996)</u> |
| Shanghai,<br>China           | TSP from 4 locations. NM<br>sonication extraction; extract<br>separated into 5 fractions                                 | Exposure of primary<br>hepatocytes from SD<br>rats                   | Unscheduled<br>DNA synthesis<br>(incorporation<br>of radio-labelled<br>thymidine) | Significant dose-related increases in DNA<br>damage induced by all fractions of all samples,<br>except residential area during summer.<br>Residential area induced the weakest response.<br>Highest potency for PAH-containing fraction;<br>in contrast, <i>Salmonella</i> potency highest for polar<br>fraction | <u>Zhao &amp; Zhu</u><br>(1997)         |
| Leeds, United<br>Kingdom     | Size-fractionated PM from<br>3 urban sites in Leeds city<br>centre, collected with cascade<br>impactor. Water extraction | Incubation of plasmid<br>DNA with PM extracts<br>for 60 h            | Plasmid DNA<br>strand break assay   | Significant induction of strand breaks; highest activity associated with smallest size fractions   | <u>Lingard et al.</u><br>(2005)         |
| Novara<br>Province,<br>Italy | In situ exposures at 19 sites,<br>including urban, rural, and<br>industrial locations                                    | AFLPs in <i>Trifolium</i><br><i>repens</i> L. after 6 wk<br>exposure | DNA sequence<br>changes measured<br>as alterations in<br>restriction sites        | Significant increase in percentage of total<br>polymorphisms at 4–7 sites, depending on<br>season. Elevated ozone at locations with elevated<br>polymorphisms  | <u>Piraino et al.</u><br>(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; NO<sub>2</sub>, nitrogen dioxide; NR, nitroreductase; OAT, *O*-acetyltransferase; PAHs, polycyclic aromatic hydrocarbons; PB, phenobarbital; PM, particulate matter; PM<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10 μm; PM<sub>2.5</sub>, 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<br>with Atlas Rat 1.2<br>macroarrays                         | Altered expression of genes coding for hormone<br>receptors, growth factors, cytokines, IL-2,<br>chemokines, and ligand-gated ion channels   | +    | <u>Wise et al.</u><br>(2006)                     |
| Male<br>hypertensive<br>rats | Intratracheal installation of EHC-93 urban<br>particulate for 2–20 h   | Lung RNA evaluated with Affymetrix U34A   | Altered expression of genes in the oxidative<br>stress, inflammation, transcriptional regulation,<br>and cardiovascular system pathways  | +    | <u>Kooter et al.</u><br>(2005)                   |
| Female<br>BALB/cJ<br>mice    | Inhalation exposure to 380 $\mu$ g/m <sup>3</sup> for 4 h and 24 h of ultrafine carbon particles generated by an electric spark motor  | Lung RNA evaluated<br>with Affymetrix<br>U74Av2 GeneChip                        | Increased expression of heat shock genes<br>after 4 h, but increased expression of other<br>immunomodulatory proteins after 24 h of<br>inhalation exposure   | +    | <u>André et al.</u><br>(2006)                    |
| Male<br>C57BL/6<br>mice      | Inhalation exposure to < 2.5 μm CAPs from an<br>urban area near San Joaquin Valley, California,<br>USA, for 6 h/d, 5 d/wk, for 2 wk. Winter CAPs,<br>39.01 μg/m <sup>3</sup> ; summer CAPs, 21.7 μg/m <sup>3</sup> | Lung RNA evaluated<br>by qRT-PCR (TaqMan,<br>Applied Biosystems)                | Winter CAPs increased expression of <i>IL-2</i> , <i>MIP-</i><br><i>1α</i> , <i>TNFα</i> , <i>CYP1a1</i> , <i>ICAM-1</i> , and <i>Nox-2</i>  | +    | <u>Tablin et al.</u><br>(2012)                   |
| Male<br>C57BL6<br>mice       | Inhalation exposure in Craeybeckx tunnel in<br>Antwerp, Belgium, for 5 d; 55.1 $\mu$ g/m <sup>3</sup> of PM <sub>2.5</sub> ;<br>EC = 13.9 $\mu$ g/m <sup>3</sup>   | Olfactory bulb and<br>hippocampus tissue<br>from brain evaluated by<br>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> $\alpha$ , <i>COX2</i> , <i>NFE2L2</i> , <i>IL-6</i> , and <i>BDNF</i> in the olfactory bulb | +    | <u>Bos et al.</u><br>(2012)                      |
| Male Brown<br>Norway rats    | Rats first given ovalbumin by intranasal installation for 3 d, then exposed to 1–2.5 $\mu$ m CAPs from Grand Rapids, Michigan, USA, for 8 h/d for 13 d at 493 $\mu$ g/m <sup>3</sup>                               | Lung RNA evaluated<br>with Affymetrix R230<br>2.0 whole-genome<br>arrays        | Altered expression of genes in the inflammation<br>and immune-response pathways  | +    | <u>Heidenfelder</u><br>et al. (2009)             |
| Male<br>C57BL/CBA<br>mice    | Inhalation exposure in situ near a busy highway<br>and 2 steel mills in Hamilton, Ontario, Canada  | Lung RNA evaluated<br>with Agilent G4121B<br>microarrays                        | Altered expression of genes in lipid droplet synthesis and antioxidant defence   | +    | <u>Rowan-</u><br><u>Carroll et al.</u><br>(2013) |
| Male F344<br>rats            | Inhalation exposure for 5 h/d, 4 d/wk, up to<br>10 months to CAPs from Riverside, California,<br>USA. Mass ( $\mu$ g/m <sup>3</sup> ) was 63 (UFP), 149 (FP), and<br>58 (CP)                                       | Brain RNA evaluated<br>with Affymetrix<br>GeneChip Rat Genome<br>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><br>al. (2013)                |
| Male<br>C57Bl/6<br>mice      | Inhalation exposure to $PM_{2.5}$ CAPs from<br>unspecified urban area for 8 h/d for 9 wk   | Lung RNA evaluated by<br>RT-PCR   | Increased expression of DNMT1  | +    | Soberanes et<br>al. (2012)                       |
| Male<br>C57BL/6<br>mice      | Inhalation exposure to PM <sub>2.5</sub> CAPs from<br>Columbus, Ohio, USA, for 6 h/d, 5 d/wk, for<br>10 months   | RNA from epididymal<br>mouse adipose tissues<br>evaluated by RT-PCR             | Increased ERAD and RIDD  | +    | <u>Mendez et al.</u><br>(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  $\mu$ 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<br>line MCF7                              | Extract of urban dust<br>particulate SRM 1649a<br>+ B[ <i>a</i> ]P           | Affymetrix U133A<br>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><br><u>al. (2005)</u>     |
| Primary mouse alveolar type<br>II cells                            | PM <sub>2.5</sub> NIST SRM 1649<br>standard                                  | RT-qPCR   | Increased DNMT1  | +      | <u>Soberanes et al.</u><br>(2012)            |
| Human lung epithelial cell<br>line L132                            | PM <sub>2.5</sub> from Dunkirk,<br>France                                    | RT-qPCR   | Increased apoptosis genes: <i>TNFα</i> , <i>caspase-3</i> , -8, -9   | +      | <u>Dagher et al.</u><br><u>(2006)</u>        |
| Human lung cell line A549  | PM <sub>2.5</sub> from Dunkirk,<br>France                                    | RT-qPCR   | Increased CYP1A1, CYP2E1, CYP2F1, NQO1, and GST-π1   | +      | <u>Billet et al.</u><br>(2007)               |
| Human AMs  | PM <sub>2.5</sub> from Dunkirk,<br>France                                    | RT-qPCR   | Increased CYP1A1, CYP2E1, NQO1, and GST-π1/μ3  | +      | <u>Saint-Georges et</u><br><u>al. (2008)</u> |
| Human AMs and human<br>lung cell line L132                         | PM <sub>2.5</sub> from Dunkirk,<br>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> - $\pi$ 1, and/or <i>GST</i> - $\mu$ 3. In L132 cells co-cultured with AMs, no changes in gene expression | +<br>- | <u>Abbas et al.</u><br>(2009)                |
| Human lung cell line BEAS-<br>2B                                   | PM <sub>2.5-3.0</sub> from Dunkirk,<br>France                                | RT-qPCR   | Increased CYP1A1, CYP1B1, IL-6, and IL-8   | +      | <u>Dergham et al.</u><br>(2012)              |
| Human monocyte-<br>macrophage line U937                            | PM <sub>10</sub> from Rome, Italy<br>(direct exposure)                       | Clontech Atlas Human<br>Toxicology 1,2 cDNA<br>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><br>(2011)            |
| Human airway epithelial<br>cells                                   | Coarse, fine, and<br>ultrafine PM from<br>Chapel Hill, North<br>Carolina, US | Affymetrix HGU133A<br>microarray  | Changes in expression of NRF2-mediated oxidative-<br>stress response genes, cell-cycle genes, DNA damage<br>checkpoint genes, and polo kinase genes  | +      | <u>Huang et al.</u><br>(2011)                |
| Human lung cell line<br>HEL12469                                   | EOM from PM <sub>2.5</sub><br>from 4 cities in Czech<br>Republic             | Illumina Human-<br>HT12v3 Expression<br>Bead Chips                            | Increased <i>CYP1B1</i> ; decreased <i>ABC Transporters</i> ,<br><i>Wnt</i> , <i>TGF-β</i> , steroid biosynthesis, glycerolipid<br>metabolism  | +      | <u>Líbalová et al.</u><br>(2012)             |
| Normal human<br>tracheobronchial epithelium<br>3-D cell constructs | PM from Swansea,<br>United Kingdom   | RT-PCR and Biorad<br>Human Stress and<br>Toxicity Pathway Finder<br>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><br><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  $\mu$ m;  $PM_{2.5}$ , particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ m; STM, standard reference mixture.

| Supplemental Tabl | e S20 Lung inflammation in a | nimals after exposure to a | air pollution particles |
|-------------------|------------------------------|----------------------------|-------------------------|
|                   |                              |                            |                         |

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

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

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

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

BALF, bronchoalveolar lavage fluid; CAPs, concentrated ambient particles; d, day or days; DCFH, 2',7'-dichlorodihydrofluorescein; DMTU, dimethylthiourea; h, hour or hours; IFN- $\gamma$ , interferon-gamma; IL-6, interleukin-6; NA, not applicable; NAC, *N*-acetylcysteine; PBS, phosphate-buffered saline; PM, particulate matter; PM<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10 µm; PM<sub>2.5</sub>, 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<br>conditions                  | Effect  | Reference                                |
|---|---|---|---|---|--|
| PM <sub>2.5</sub> from Porte d'Auteuil,<br>France   | Human nasal<br>epithelial cells                     | Water<br>(sonication)                   | 10-80 μg/cm² for<br>24 h                | Concentration-dependent increase in release of GM-CSF,<br>IL-6, and IL-8. Weak and inconsistent increase in TNF<br>(protein)  | <u>Auger et al.</u><br>(2006)            |
| PM <sub>2.5</sub> from a school<br>playground in Vitry-sur-<br>Seine, France, or SRM 1648 | Human bronchial<br>epithelial cells (16-<br>HBE)    | Sonication                              | 10–30 μg/cm² for<br>4 h                 | Concentration-dependent increase in release of GM-CSF (protein)   | <u>Baulig et al.</u><br>(2003)           |
| SRM 1648  | Alveolar type<br>2 cells or<br>macrophages          | NA                                      | 20 μg/cm² (or<br>200 μg/mL) for<br>20 h | Increased release of IL-6 and MIP2 (protein)  | <u>Becher et al.</u><br>(2007)           |
| PM <sub>10</sub> from a street in<br>London, United Kingdom                               | J774 macrophages                                    | PBS<br>(vortexing)                      | 5–100 μg/mL for<br>4 h                  | Concentration-dependent increase in TNF secretion (protein)   | <u>Brown et al.</u><br>(2004)            |
| PM <sub>10</sub> from a street in<br>London, United Kingdom                               | Mononuclear<br>blood cells                          | PBS<br>(vortexing)                      | 10 $\mu$ g/mL for 4 h                   | Increased TNF (protein) secretion   | <u>Brown et al.</u><br>(2007)            |
| EHC93   | Human alveolar<br>and bronchial<br>epithelial cells | NA                                      | 100 μg/mL for<br>24 h                   | Increased secretion of GM-CSF, IL-6, IL-8, IL-1 $\beta$ , and TNF (protein and mRNA) in macrophages, whereas there was less effect in bronchial epithelial cells  | <u>Fujii et al.</u><br>(2001, 2002)      |
| PM from Dunkirk, France   | L132  | Mechanical<br>collection<br>from plates | 19 μg/mL and<br>75 μg/mL for<br>24–72 h | Increased TNF (protein), iNOS activity, and NO production (Griess)  | <u>Garçon et al.</u><br>(2006)           |
| PM <sub>2.5</sub> from Helsinki,<br>Finland, EHC93, or SRM<br>1649                        | RAW 264.7   | Methanol<br>or water<br>(sonication)    | 150 μg/mL for<br>12 h or 24 h           | EHC93 was more inflammogenic than SRM 1649.<br>PM <sub>2.5</sub> was the least inflammogenic based on TNF, IL-1,<br>IL-6 (protein), and NO (Griess). Water and methanol<br>extraction generated the same effect             | <u>Jalava et al.</u><br>(2005)           |
| PM <sub>10</sub> from a busy street in Stockholm, Sweden.                                 | A549  | Water<br>(sonication)                   | $40 \ \mu g/cm^2$ for $4 \ h$           | Increased levels of IL-6, IL-8, and TNF (protein)   | <u>Karlsson et al.</u><br>(2006)         |
| SRM 1648  | RAW 264.7   | NA                                      | 62.5 μg/cm² for<br>4 h                  | Increased PGE <sub>2</sub> release (protein)  | <u>Schneider</u><br><u>et al. (2005)</u> |
| SRM 1649  | Mouse<br>macrophages and<br>Kupffer cells           | NA                                      | 100 μg/mL or<br>200 μg/mL for<br>24 h   | Concentration-dependent increases in IL-6 (protein). Also increased mRNA of IL-6, TNF, and IL-12  | <u>Tan et al.</u><br>(2009)              |
| EHC93 or ROFA   | AMs   | NA                                      | 0.01–0.1 mg/mL<br>for 24 h              | Concentration-dependent TNF response by EHC93.<br>ROFA less potent than EHC93. Also increased cytokine<br>profile by EHC93 (increased IL-8, IL-6, GM-CSF, MIP-1a,<br>MIP-1b, IL-1 $\alpha$ , IL-1 $\beta$ , IL-10, and TNF) | <u>van Eeden</u><br><u>et al. (2001)</u> |
| PM <sub>2.5</sub> from Cache Valley,<br>Utah, USA   | BEAS-2B   | Water<br>(sonication)                   | 0.8–3.9 μg/mL for<br>24 h               | Increased IL-6 excretion (protein). Excretion of TNF was unaltered  | <u>Watterson</u><br><u>et al. (2007)</u> |

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

# Supplemental Table S21 (continued)

| Particles   | Cells                               | Extraction                              | Exposure<br>conditions           | Effect   | Reference  |
|---|-------------------------------------|---|----------------------------------|--|--|
| SRM 1649 or ROFA  | RLE, AMs, or co-<br>culture         | NA                                      | 1–50 μg/mL for<br>24 h           | Unaltered secretion of MIP2 and TNF in mono-cultures,<br>whereas there were concentration-dependent increases in<br>co-cultures  | <u>Tao &amp; Kobzik</u><br>(2002)                                  |
| PM <sub>10</sub> , PM <sub>2.5</sub> , PM <sub>1</sub> , and PM <sub>0.4</sub><br>from Milan, Italy, collected<br>during summer or winter | THP-1 or THP-1/<br>A549 co-cultures | Water<br>(sonication)                   | $10 \ \mu g/cm^2$ for 24 h       | Increased IL-6 and IL-1 $\beta$ secretion (protein) in THP-1 or co-cultures (and IL-8 in co-cultures) exposed to PM <sub>10</sub> collected during summer (correlated with endotoxin level)  | <u>Longhin et al.</u><br>(2013)                                    |
| Fine and coarse fraction of PM from 4 cities in Europe  | Rat lung type 2<br>cells and AMs    | Methanol                                | 10–50 mg/mL for<br>24 h          | Increased MIP2, IL-6, and TNF (protein) with both<br>spatial and temporal variability. Coarse fraction more<br>potent than fine fraction   | <u>Dybing</u><br>et al. (2004),<br><u>Hetland et al.</u><br>(2005) |
| Coarse, fine, and ultrafine<br>particles from 6 cities in<br>Europe   | RAW 264.7                           | Methanol<br>(sonication)                | 15–300 μg/mL for<br>24 h         | Concentration-dependent increases in NO (Griess), TNF,<br>IL-6, and MIP2 (protein). Coarse particles more potent<br>than fine and ultrafine particles. Inter-city variability in<br>inflammation potency                             | <u>Jalava et al.</u><br>(2007)                                     |
| PM <sub>2.5</sub> from rural and urban<br>(Dakar city) sites in Senegal   | BEAS-2B                             | Mechanically<br>recovered               | 3–12 μg/mL for<br>24–48 h        | Increased TNF, IL-8, and IL-6, and unaltered IL-1 $\beta$ secretion. Increased IL-1 $\beta$ , IL-6, and IL-8 mRNA levels, and unaltered TNF. Urban sites more potent than rural site   | <u>Dieme</u><br>et al.(2012)                                       |
| PM <sub>10</sub> from different<br>locations in Mexico City,<br>Mexico  | J774A.1 and RLE                     | Mechanically<br>recovered               | 40-80 μg/cm² for<br>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><br><u>Moreno</u><br><u>et al.(2002)</u>             |
| $PM_{2.5}$ collected at a busy<br>street or urban background<br>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><br><u>et al.(2007)</u>                               |
| PM from a town with many<br>wood stoves and a rural<br>area, Denmark  | A549 and THP-1                      | Mechanical<br>collection<br>from plates | 2.5–100 μg/mL<br>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><br>et al.(2011)                                   |
| PM <sub>10</sub> from Utah Valley,<br>Utah, USA   | BEAS-2B                             | Water<br>(agitation)                    | 125–500 μg/mL<br>for 2 h or 24 h | Concentration-dependent increase in IL-6 and IL-8<br>(protein or protein) production. Lowest potency had<br>samples that were collected during a strike at the steel mill<br>in the area   | <u>Frampton</u><br>et al.(1999)                                    |
| PM from a street tunnel in<br>Oslo, Norway  | THP-1                               | Scraping off<br>the filter              | 20–280 µg/mL for<br>12 h         | Increased TNF, IL-1 $\beta$ , and IL-8 (protein) secretion.<br>Undetectable levels of IL-4, IL-6, and IL-10. No difference<br>between particles collected in seasons with or without use<br>of studded tyres                         | <u>Kocbach</u><br>et al.(2008)                                     |

# Supplemental Table S21 (continued)

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

#### Supplemental Table S21 (continued)

| Particles  | Cells                           | Extraction                         | Exposure<br>conditions              | Effect  | Reference                             |
|--|---------------------------------|------------------------------------|-------------------------------------|---|---------------------------------------|
| PM <sub>2.5</sub> from Toronto,<br>Canada, or SRM 1648                           | A549                            | Water                              | 10–1000 μg/mL<br>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 <sub>2.5</sub> samples                                   | <u>Akhtar et al.</u><br>(2010)        |
| SRM 1649, ROFA, or<br>CAPs from Boston,<br>Massachusetts, USA                    | AMs (LPS-<br>primed)            | Saline<br>(sonication)<br>for CAPs | 25–100 μg/mL for<br>20 h            | Substantial difference in potency to promote TNF<br>secretion by CAPs collected on different days. TNF<br>secretion was attributed mainly to the particle fraction,<br>whereas the water-soluble fraction had little effect | <u>Imrich et al.</u><br>(1999)        |
| ROFA   | Bronchial<br>epithelial cells   | NA                                 | 5–200 μg/mL for<br>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><br>(1997)        |
| ROFA   | RAW 264.7 and hamster AMs       | Saline<br>(sonication)             | 100–400 μg/mL<br>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><br>et al. (1998)     |
| SRM 1649 or CAPs from<br>Boston, Massachusetts, USA                              | AMs (LPS-<br>primed)            | Water<br>(sonication)              | 100 μg/mL for<br>20 h               | Increased TNF release (protein), which was diminished by treatment with NAC, catalase, or DMTU  | <u>Imrich et al.</u><br><u>(2007)</u> |
| PM <sub>10</sub> from an urban street<br>in Stockholm, Sweden                    | RAW 264.7                       | Water<br>(sonication)              | 1–100 μg/mL for<br>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><br>et al. (2007)       |
| $\mathrm{PM}_{\mathrm{2.5}}$ from Vermont, USA                                   | Murine alveolar<br>type 2 cells | Water and sonication               | 10 μg/cm² for<br>1–8 h              | Increased NFκB binding to DNA, which was ameliorated by pre-treatment with catalase, but not DFO  | <u>Shukla et al.</u><br>(2000)        |
| PM from Düsseldorf, SRM<br>1648, EHC93, volcanic and<br>oil fly ash              | Human or rat<br>alveolar cells  | Not specified                      | 18.5–500 μg/mL<br>for 18–20 h       | Increased TNP and IL-6 secretion (protein) by EHC93,<br>PM from Düsseldorf, and SRM 1648. Unaltered secretion<br>by volcanic and oil fly ash. DFO pre-treatment had no<br>effect on SRM 1648-mediated IL-6 secretion        | <u>Becker et al.</u><br>(1996)        |
| EOM of coarse and fine<br>particles from Paso del<br>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><br>(2009)         |
| EOM of road tunnel<br>particles from Shanghai,<br>China                          | A549                            | DCM<br>(sonication)                | 1–400 μg/mL for<br>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><br>(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; PGE<sub>2</sub>, prostaglandin E2; PM, particulate matter; PM<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10  $\mu$ m; PM<sub>2.5</sub>, particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ 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 <sub>2.5</sub> from Porte<br>d'Auteuil, France                    | Human nasal<br>epithelial cells                              | Water (sonication) | 10-80 μg/cm <sup>2</sup><br>for 3 h         | Increased ROS production (DCFH)  | <u>Auger et al. (2006)</u>        |
| PM <sub>2.5</sub> from Vitry-sur-<br>Seine, France, or SRM<br>1648   | Human bronchial<br>epithelial cells<br>(16-HBE)              | Sonication         | 10 μg/cm² for<br>4 h                        | Increased ROS production (DCFH)  | <u>Baulig et al. (2003)</u>       |
| PM from Düsseldorf,<br>SRM 1648, EHC93,<br>volcanic and oil fly ash  | Human or rat<br>alveolar cells                               | Not specified      | 18.5–500 μg/mL<br>for 0.5 h                 | Increased ROS production (luminol and<br>chemiluminescence) by all particles. Oil fly ash<br>particles more potent than other particles  | <u>Becker et al. (1996)</u>       |
| PM from Chapel Hill,<br>North Carolina, USA,<br>and ROFA             | Bronchial<br>epithelial cells<br>and AMs                     | Water (sonication) | 11 or 50 $\mu$ g/m <sup>3</sup> for 1 h     | Increased ROS production (DCFH and DHR). Coarse,<br>fine, and ultrafine particles generated the same level of<br>ROS on mass basis. High temporal variability in ROS<br>production | <u>Becker et al. (2005)</u>       |
| PM from a town with<br>many wood stoves and<br>a rural area, Denmark | A549 and THP-1   | Water              | 2.5–100 μg/mL<br>for 3 h                    | Concentration-dependent increase in ROS production (DCFH)  | <u>Danielsen et al.</u><br>(2010) |
| ROFA and CAPs from<br>Boston, Massachusetts,<br>USA                  | Hamster AMs  | Water (sonication) | 25–200 μg/mL<br>and 4–20 μg/mL<br>for 0.5 h | Concentration-dependent increase in ROS production (DCFH). ROFA had a stronger effect than CAPs  | <u>Goldsmith et al.</u><br>(1997) |
| Airborne PM from<br>Düsseldorf, Germany                              | Human bronchial<br>epithelial cells<br>(IB3–1 and S-9<br>CF) | Baghouse           | 25 μg/cm² for<br>1 h                        | Increased ROS production (DCFH)  | <u>Kamdar et al.</u><br>(2008)    |
| PM <sub>10</sub> from a busy street in Stockholm, Sweden             | A549   | Water (sonication) | 20 µg/cm² for<br>2 h                        | Increased ROS production (DCFH)  | <u>Karlsson et al.</u><br>(2008)  |
| PM from Osaka, Japan   | Human<br>mononuclear<br>blood cells                          | Aqueous solution   | 50–650 μg/mL<br>for 200 minutes             | Bell-shaped ROS production (lucigenin chemiluminescence assay)   | <u>Ohyama et al.</u><br>(2007)    |
| PM <sub>2.5</sub> from Vermont,<br>USA                               | Murine alveolar<br>type 2 cells                              | Water (sonication) | 10 μg/cm² for<br>1–8 h                      | Increased ROS (DCFH)   | <u>Shukla et al. (2000)</u>       |
| PM <sub>10</sub> from Beijing,<br>China                              | A549   | Water (sonication) | 10 μg/mL for<br>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 <sub>2.5</sub> from Denver,<br>Colorado, USA                      | NR8383 AMs   | Water (shaking)    | 20–200 pg/cell<br>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/cm² (or<br>200 μg/mL) for<br>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<br>conditions                                 | Effect   | Reference                         |
|--|--|--|--|--|-----------------------------------|
| Oil fly ash from a<br>power plant in Sicily,<br>Italy  | A549   | Water  | 17.2–68.8 of<br>VOSO <sub>4</sub> equiv<br>for 0.5–4 h | Concentration-dependent increase in ROS production<br>(DCFH) Ameliorated by treatment with DFO   | <u>Di Pietro et al.</u><br>(2009) |
| Coal fly ash from a<br>dumping site of a<br>thermal power plant in<br>Aligarh, India   | Mononuclear<br>blood cells                     | DMSO (0.5%) or<br>water (sonication)                         | 600–2400 ppm<br>for 24 h                               | Increased generation of ROS (DCFH) by DMSO (0.5%) and ROS production by aqueous extract  | <u>Dwivedi et al.</u><br>(2012)   |
| SRM 1648   | Human<br>pulmonary artery<br>endothelial cells | NA   | 1–100 μg/mL for<br>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<br>4 h                                 | Increased ROS (DCFH)   | <u>Schneider et al.</u><br>(2005) |
| PM <sub>10</sub> , PM <sub>2.5</sub> , PM <sub>1</sub> , and<br>PM <sub>0.4</sub> from Milan,<br>Italy, collected during<br>summer or winter | A549 and THP-1                                 | Water (sonication)   | 10 µg/cm² for<br>1 h                                   | All size fractions increased ROS production. Samples<br>collected during winter generally slightly more potent<br>than samples collected during summer   | <u>Longhin et al.</u><br>(2013)   |
| PM <sub>10</sub> from Utah Valley,<br>Utah, USA, dust from 3<br>different years  | AMs  | Water (sonication)   | ≤ 1 mg/mL for<br>20 minutes or<br>24 h                 | Increased ROS production (chemiluminescence) by<br>samples from the year of collection, which was not<br>affected by DFO treatment. Unaltered (or decreased)<br>ROS production by dihydrorhodamine 123 assay | <u>Soukup et al.</u><br>(2000)    |
| ROFA and CAPs from<br>Boston, Massachusetts,<br>USA  | Hamster AMs                                    | Water (sonication)   | 25–200 μg/mL<br>for 0.5 h                              | Concentration-dependent increase in ROS production<br>(DCFH), which was inhibited by DFO. Substantial<br>variation in ROS production by CAPs collected on<br>different days                                  | <u>Goldsmith et al.</u><br>(1998) |
| EOM of PM from<br>Taiwan, China  | MCF-7  | Hexane/acetone<br>(ultrasound)                               | 0.04–0.05 m³-<br>air equiv for<br>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 <sub>10</sub> from<br>Kaifaqu district,<br>Dalian, China   | HepG2  | Sequential<br>extraction in<br>DCM, acetone, and<br>methanol | 7.5–30 mg/mL<br>for 1 h                                | Concentration-dependent increase in ROS production (DCFH)  | <u>Jiang et al. (2011)</u>        |
| EOM of road tunnel<br>particles from<br>Shanghai, China  | A549   | DCM (sonication)   | 10–400 μg/mL<br>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<sub>10</sub>, particulate matter with particles of aerodynamic diameter < 10  $\mu$ m; PM<sub>2.5</sub>, particulate matter with particles of aerodynamic diameter < 2.5  $\mu$ m; ROFA, residual oil fly ash; ROS, reactive oxygen species; SRM, standard reference mixture.

| Particles  | Extraction                           | Method  | Effect   | Reference   |
|--|--------------------------------------|---|--|---|
| Fine particles from Duisburg,<br>Germany   | Water<br>(sonication)                | ESR with $H_2O_2$ and DMPO as spin trap                 | Concentration-dependent increase in ROS production<br>(DMPO-OH signal), which was inhibited by DFO or<br>catalase  | <u>Knaapen et al. (2002)</u>  |
| TSP from the city centre of<br>Athens, the suburb of Piraeus, or<br>street dust, Greece                    | PBS (sonication)                     | ESR with H <sub>2</sub> O <sub>2</sub> and without DMPO | Little difference in potency between TSP from different<br>locations, whereas street dust generated lower levels of<br>ROS. Decreased ROS production by treatment with DFO         | <u>Valavanidis et al.</u><br>(2000)                                       |
| TSP, PM <sub>10</sub> , and PM <sub>2.5</sub> from<br>Athens, Greece                                       | Water<br>(sonication)                | ESR with H <sub>2</sub> O <sub>2</sub> and without DMPO | Detection of DMPO-OH signals. No difference between size fractions   | <u>Valavanidis et al.</u><br>(2005)                                       |
| Coal fly ash from a dumping<br>site of a thermal power plant in<br>Aligarh, India                          | DMSO (0.5%) or<br>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<br>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><br><u>(1999)</u>                                 |
| PM <sub>2.5</sub> from San Joaquin Valley,<br>California, USA  | Salt solution<br>(shaking)           | Benzoate as chemical<br>probe of hydroxyl<br>radicals   | Particles from urban area more potent than particles<br>from rural area. Treatment with DFO decreased ROS<br>production  | <u>Shen et al. (2011),</u><br><u>Shen &amp; Anastasio</u><br>(2011, 2012) |
| SRM 1648 or SRM 1649   | Water                                | Deoxyribose assay                                       | Concentration-dependent increase in ROS production.<br>SRM 1649 more potent than SRM 1648. Inhibition of<br>ROS production by DFO  | <u>Ball et al. (2000)</u>   |
| PM from Utah Valley, Utah, USA   | Water (agitation)                    | Deoxyribose assay with $\rm H_2O_2$                     | Higher ROS production by samples collected during<br>activity at a steel mill. Decreased ROS production by DFO<br>or antioxidant (DMTU or DMSO) treatment                          | <u>Frampton et al.</u><br>(1999)  |
| Aqueous extract from Utah<br>Valley, Utah, USA, before,<br>during, and after a strike at the<br>steel mill | Water (agitation)                    | Deoxyribose assay                                       | Increased ROS production in samples before and after the<br>strike compared with samples collected during the strike.<br>Decreased ROS production by treatment with DFO or<br>DMTU | <u>Ghio &amp; Devlin</u><br>(2001)  |
| Water-soluble or insoluble<br>fraction of CAPs from Boston,<br>Massachusetts, USA                          | Water<br>(sonication)                | DCFH with $H_2O_2$                                      | Only ROS production in the presence of $H_2O_2$ . Water-<br>soluble fraction more potent than insoluble fraction.<br>Diminished ROS production by treatment with DFO               | <u>Imrich et al. (2007)</u>   |
| PM <sub>10</sub> from an urban street in<br>Stockholm, Sweden  | Water<br>(sonication)                | DTT   | Concentration-dependent increase, which was slightly inhibited by DFO  | <u>Lindbom et al.</u><br>(2007)   |
| PM <sub>2.5</sub> from urban air particulates<br>at 20 different sites (19 European<br>cities)             | Water                                | ESR and $H_2O_2$ with DMPO as spin trap                 | Different ROS production potential between sites and<br>over time. Heterogeneous association between ROS<br>production and PM constituents across locations                        | <u>Künzli et al. (2006),</u><br><u>Nawrot et al. (2009)</u>               |
| $PM_{2.5}$ 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><br>(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<br>wood stoves and a rural area,<br>Denmark                                  | Mechanical<br>collection from<br>plates | ESR (with ascorbate and DMPO) and DCFH                       | Rural particles more potent than city particles by ESR.<br>Same ROS production by particles for the DCFH assay  | Danielsen et<br>al.(2011)          |
| Fine and coarse particles from<br>Duisburg (industrial area) and<br>Borken (rural area), Germany      | Water<br>(sonication)                   | ESR with $H_2O_2$ and DMPO as spin trap                      | Fine and coarse particles from Borken generated the same<br>level of ROS. Coarse particles from Duisburg generated<br>more ROS than fine particles  | <u>Schins et al. (2004</u>         |
| Fine and coarse particles from various cities in Germany  | Water<br>(sonication)                   | ESR with H <sub>2</sub> O <sub>2</sub> and DMPO as spin trap | Coarse particles generated higher level of ROS than fine<br>particles. Samples from industrial area (Dortmund and<br>Duisburg) generated higher level of ROS than samples<br>from rural site (Borken) | <u>Shi et al. (2006)</u>           |
| PM <sub>2.5</sub> from a smelter site and<br>non-industrialized site in<br>Germany                    | Water<br>(sonication)                   | ESR  | Higher levels of ROS production in samples from industrial area   | <u>Schaumann et al.</u><br>(2004)  |
| PM <sub>10</sub> from an urban or suburban<br>site in Mexico City, Mexico                             | Mechanically<br>recovered               | ESR with $H_2O_2$ and DMPO as spin trap                      | Particles from urban site more potent than particles<br>from suburban site. Temporal variation related to wind<br>direction at sampling site  | <u>Quintana et al.</u><br>(2011)   |
| PM <sub>10</sub> from a busy street in<br>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>         |
| $PM_{10}$ from Edinburgh, Scotland  | Water<br>(sonication)                   | 2.3-dihydroxybenzoic acid production                         | Increased ROS production  | <u>Donaldson et al.</u><br>(1997)  |
| Aqueous extract or insoluble<br>fraction of TSP from North<br>Provo, Utah, USA                        | Water (agitation)                       | Deoxyribose assay with $\rm H_2O_2$                          | Increased ROS production. Higher response by soluble extract compared with insoluble fraction   | <u>Ghio et al. (1999)</u>          |
| PM <sub>10</sub> from Helsinki, Finland, collected during spring or winter                            | Methanol<br>(sonication)                | ESR with $H_2O_2$  | Increased production, without clear seasonal variation  | Salonen et al. (200                |
| $PM_{2.5}$ 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><br>(2012)   |
| Fine and coarse particles from<br>Düsseldorf, Germany   | Water<br>(sonication)                   | ESR with $H_2O_2$ and DMPO as spin trap                      | Coarse PM generated more ROS (DMPO-OH signal)<br>than fine PM when compared at equal mass. Samples<br>collected during summer had stronger ROS generating<br>potency than autumn/winter samples       | <u>Shi et al. (2003)</u>           |
| PM <sub>2.5</sub> and PM <sub>10</sub> from Maastricht,<br>Netherlands                                | Not reported                            | ESR with DMPO as spin trap                                   | No difference in ROS generating ability between $PM_{2.5}$<br>and $PM_{10}$ samples. Season-dependent variation in ROS<br>generating ability of outdoor $PM_{10}$ samples                             | <u>Briedé et al. (2005</u>         |
| PM <sub>2.5</sub> ,PM <sub>10</sub> , or TSP from school<br>playgrounds in Maastricht,<br>Netherlands | No extraction                           | ESR with DMPO as spin trap                                   | Differences in ROS generation in samples collected at<br>different locations. No clear difference between different<br>size modes   | <u>Hogervorst et al.</u><br>(2006) |

#### Supplemental Table S23 (continued)

| Particles   | Extraction            | Method                                | Effect  | Reference  |
|---|-----------------------|---------------------------------------|---|--|
| PM in different size fractions<br>from urban and rural sites,<br>United Kingdom     | Water<br>(sonication) | EST with $H_2O_2$ and DMPO            | ROS production highest for sites near traffic. Little effect related to size fractions  | Wessels et al. (2010)                            |
| PM <sub>2.5</sub> from San Joaquin Valley,<br>California, USA                       | Not specified         | DTT                                   | Temporal variation in ROS production, although no clear effect of summer and winter   | <u>Charrier &amp;</u><br><u>Anastasio (2012)</u> |
| EOM of TSP from Fresno,<br>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 <sub>2.5</sub> from Los Angeles,<br>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 <sub>2.5</sub> from Atlanta, Georgia, USA  | Water or<br>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,<br>Virginia, USA                                  | Water<br>(sonication) | DTT                                   | Ultrafine particles more potent on mass basis than $PM_{2.5}$   | <u>Jeng (2010)</u>                               |
| Coarse, fine, and ultrafine<br>particles from Los Angeles basin,<br>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<br>Mexico City, Mexico                                | Water                 | DTT                                   | Fine particles more potent than coarse particles.<br>Temporal and spatial differences in potency  | <u>De Vizcaya-Ruiz et</u><br><u>al. (2006)</u>   |
| Coarse, fine, and ultrafine<br>particles from Los Angeles basin,<br>California, USA | VACES                 | DTT                                   | Ultrafine particles generated higher DDT oxidation than<br>coarse and fine particles on mass basis. Substantial spatial<br>and temporal variation in ROS production ability of<br>ultrafine particles | <u>Li et al. (2003)</u>                          |
| PM from Los Angeles and San<br>Francisco, California, USA                           | VACES                 | DTT                                   | Higher ROS production on mass basis for ultrafine<br>particles compared with fine and coarse particles. Strong<br>correlation between organic carbon content and ROS<br>production                    | <u>Ntziachristos et al.</u><br>(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)                           |
| PM <sub>2.5</sub> from Toronto, Canada, and<br>SRM 1648                             | Water                 | DTT                                   | Similar level of ROS production of PM <sub>2.5</sub> and SRM 1648.<br>Water-soluble fraction of SRM 1648 more potent than<br>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/<br>exposure   | End-point   | Results   | Call | Reference                            |
|---|-----------------------------------|--|---|---|------|--------------------------------------|
| Hamilton,<br>Canada                                 | Herring gull                      | Feral gulls collected from a<br>steel mill environment and<br>rural reference locations  | Heritable<br>minisatellite<br>DNA mutation by<br>multilocus DNA<br>fingerprinting   | Significant > 2-fold increase in mutation rate observed<br>in offspring from the industrial site compared with<br>rural locations   | +    | <u>Yauk &amp;</u><br>Quinn<br>(1996) |
| Urban and<br>industrial<br>sites, Canada<br>and USA | Herring gull                      | Feral gulls collected from<br>4 steel mill environments,<br>2 urban sites, and 3 rural<br>reference locations                                  | Heritable<br>minisatellite<br>DNA mutation by<br>multilocus DNA<br>fingerprinting   | Significant 2-fold increase in mutation rate observed<br>in offspring from the industrial sites compared with<br>rural locations. No difference between urban sites and<br>rural sites. Minisatellite mutation rates decreased with<br>increasing distance from industrial coking oven and<br>urbanization site   | +    | <u>Yauk et al.</u><br>(2000)         |
| Hamilton,<br>Canada                                 | Swiss<br>Webster<br>mice, outbred | In situ, 10 wk exposure to<br>outdoor air near steel mills<br>and a rural reference site   | Heritable mutation<br>at ESTR loci in<br>offspring conceived<br>6 wk after exposure | Significant 1.5–2-fold increase in mutation frequency<br>at several loci observed in offspring from the steel mills<br>site compared with rural locations   | +    | <u>Somers et al</u><br>(2002)        |
| Hamilton,<br>Canada                                 | Swiss<br>Webster<br>mice, outbred | In situ, 10 wk exposure to<br>outdoor air near steel mills<br>and a rural reference site   | Heritable mutation<br>at ESTR loci in<br>offspring conceived<br>9 wk after exposure | Offspring of mice from the urban/industrial site<br>showed 1.9–2.1-fold increased mutation frequency<br>in ESTR of paternal origin, compared with offspring<br>from rural locations. HEPA filtration of outdoor air<br>significantly reduced heritable mutation rates   | +    | <u>Somers et al</u><br>(2004)        |
| Hamilton,<br>Canada                                 | C57BL/CBA<br>F1 mice,<br>inbred   | In situ, mice exposed to<br>outdoor air near steel mills<br>or HEPA-filtered air for 3,<br>10, or 16 wk, followed by<br>6 wk in the laboratory | ESTR loci mutation<br>in sperm; DNA<br>adducts in testes;<br>DNA damage in<br>sperm | Significant 1.6-fold increase in germline mutation<br>frequency at Ms6-hm observed in mice exposed<br>to outdoor air at the steel mills site compared with<br>HEPA-filtered air after 16 wk, but not at 3 wk or 10 wk.<br>DNA strand breaks in mature sperm were significantly<br>elevated in the outdoor air-exposed group after 3 wk.<br>No detectable DNA adducts observed in testes samples | +    | <u>Yauk et al.</u><br>(2008)         |
| Rome, Italy   | Wild<br>mice Mus<br>domesticus    | Feral mice collected from<br>3 sites of low, medium, and<br>high air pollution from<br>traffic   | Abnormality on<br>the sperm cells of<br>cauda epididymis                            | Significant increase in frequency of abnormal sperm<br>cells from sites with medium ( $P < 0.05$ ) and heavy<br>( $P < 0.01$ ) pollution compared with low-air-pollution<br>control site. Sperm abnormality includes banana-like<br>form, narrow form without hook, with 2 tails and<br>triangular head, amorphous, and folded form   | +    | <u>Ieradi et al.</u><br>(1996)       |

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

### Supplemental Table S24 (continued)

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

DLM, dominant lethal mutation; ESTR, expanded simple tandem repeat; HEPA, high-efficiency particulate air; LC<sub>50</sub>, 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)<br>examined | Results   | Reference  |
|-----------------------------------|--|---|--------------------------|---|--|
| Rhine-Ruhr<br>region,<br>Germany  | "City smog" collected at<br>heavily industrialized<br>location (17), collected on<br>GFFs. MeOH extraction,<br>followed by CX and Pro<br>fractionation | Syrian hamster kidney<br>cells, 18 h exposure to<br>extract in DMSO and<br>subsequently infected with<br>SV40 | Cell<br>transformation   | Significant dose-related (equiv m <sup>3</sup> /assay) increase<br>in transformation frequency induced by total<br>extract (up to 20-fold vs control). MeOH and<br>CX fractions also elicited significant increase in<br>transformation frequency. Inoculation of cells<br>transformed in vitro in Syrian golden hamsters<br>elicited malignant tumours | <u>Seemayer. et.</u><br><u>al. (1981, 1984</u> )                                     |
| Duisburg,<br>Germany              | Airborne PM from<br>industrialized Rhine–Ruhr<br>region, Draeger Box Micron<br>filter. CX extraction   | Syrian hamster kidney<br>cells, 18 h exposure to<br>extract in DMSO and<br>subsequently infected with<br>SV40 | Cell<br>transformation   | Significant dose-related (m <sup>3</sup> equiv/mL) increase in<br>transformation frequency. As little as 0.25–0.5 m <sup>3</sup><br>equiv elicited a significant enhancement in<br>transformation frequency   | <u>Seemayer et</u><br><u>al. (1987a),</u><br><u>Seemayer et al.</u><br><u>(1988)</u> |
| Rhine–Ruhr<br>region,<br>Germany  | 20 samples of "city<br>smog" collected between<br>1975 and 1986 at highly<br>industrialized locations,<br>collected on GFFs. Organic<br>extraction     | Syrian hamster kidney<br>cells, 18 h exposure to<br>extract in DMSO and<br>subsequently infected with<br>SV40 | Cell<br>transformation   | Düsseldorf provided as an example. Significant<br>dose-related (equiv m <sup>3</sup> /mL) increase in<br>transformation frequency. As little as 0.5 m <sup>3</sup><br>equiv elicited a significant enhancement in<br>transformation frequency   | <u>Seemayer et al.</u><br>( <u>1987b)</u>  |
| Rhine-Ruhr<br>region,<br>Germany  | 41 samples of "city<br>smog" collected between<br>1975 and 1990 at highly<br>industrialized locations,<br>collected on GFFs. Organic<br>extraction     | Syrian hamster kidney<br>cells, 18 h exposure to<br>extract in DMSO and<br>subsequently infected with<br>SV40 | Cell<br>transformation   | Site 54 (Düsseldorf) shown as an example.<br>Significant dose-related (equiv m <sup>3</sup> /mL) increase<br>in transformation frequency. As little as 0.5 m <sup>3</sup><br>equiv elicited a significant enhancement in<br>transformation frequency  | <u>Seemayer et al.</u><br>(1990a)  |
| Rhine–Ruhr<br>region,<br>Germany. | Airborne PM from<br>Duisburg and Düsseldorf,<br>collected on GFFs with<br>high-volume sampler. DCM<br>extraction                                       | Syrian hamster kidney<br>cells, 18 h exposure to<br>extract in DMSO and<br>subsequently infected with<br>SV40 | Cell<br>transformation   | Significant dose-related (equiv m <sup>3</sup> /assay) increase<br>in transformation frequency. Less than 1 m <sup>3</sup> equiv<br>required to elicit a significant enhancement in<br>transformation frequency. Higher responses in<br>March compared with October   | <u>Seemayer et al.</u><br>(1990a)  |
| Rhine–Ruhr<br>region,<br>Germany. | Airborne PM from<br>Duisburg and Düsseldorf,<br>collected on GFFs with<br>high-volume sampler. DCM<br>extraction                                       | Syrian hamster kidney<br>cells, 18 h exposure to<br>extract in DMSO and<br>subsequently infected with<br>SV40 | Cell<br>transformation   | Significant dose-related (equiv m <sup>3</sup> /assay) increase<br>in transformation frequency. As little as 0.5 m <sup>3</sup><br>equiv elicited a significant enhancement in<br>transformation frequency. Inoculation of cells<br>transformed in vitro in Syrian golden hamsters<br>produced malignant tumours (mostly sarcomas)                      | <u>Seemayer &amp;</u><br><u>Hornberg</u><br>(1998)                                   |

### Supplemental Table S25 (continued)

| Geographical location       | Test article   | Assay/exposure system   | End-point(s)<br>examined                  | Results   | Reference                          |
|-----------------------------|--|---|---|---|------------------------------------|
| Athens, Greece<br>(1983)    | Monthly PM samples<br>collected on cellulose filters<br>with high-volume sampler.<br>Hx sonication extraction                    | BALB/c 3T3 cells, 48 h<br>treatment with extract in<br>DMSO   | Cell<br>transformation<br>(type III foci) | Significant dose-related (µg of EOM/mL) increase<br>in transformation frequency   | <u>Athanasiou et</u><br>al. (1987) |
| Shanghai,<br>China          | TSP from 4 locations<br>in industrial Baoshan<br>industrial district. DEE<br>Soxhlet extraction                                  | CHO fibroblasts, treated with extracts in DMSO  | Cell<br>transformation                    | 3 of the 4 samples examined induced a significant increase in transformation frequency  | <u>Lu et al. (1997)</u>            |
| Tokyo, Japan<br>(1980–1983) | Airborne PM from single<br>urban site, collected on<br>quartz filters with high-<br>volume sampler. DCM<br>sonication extraction | Bhas42 cells (BALB/c 3T3<br>transfected with v-Ha-<br><i>ras</i> ), treated for 3, 7, and<br>10 days with extracts in<br>DMSO | Cell<br>transformation                    | Significant dose-related (µg of PM/mL) increase<br>in transformation frequency. Higher potency (per<br>mg of PM) in autumn compared with spring | <u>Ezoe et al.</u><br>(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 S26 Influence of genotype and lung cancer risk   |  |   |  |   |                                    |
|---|--|---|--|---|------------------------------------|
| Exposure group  | Country  | Genotype/phenotype<br>method  | Result   | <i>P</i> value  | Reference                          |
| GenAir is a case–<br>control study nested<br>within the EPIC cohort | EPIC, Europe   | NAT1, NAT2, GSTM1,<br>GSTM3, GSTT1, GSTPi,<br>CYP1A1, CYP1B1, MnSOD,<br>MPO, NQO1 | A polymorphism for <i>NQO1</i><br>(involved in oxidative damage<br>scavenging) was strongly associated<br>with lung cancer                           | Odds ratio of 8.06<br>(95% CI, 1.74–37.41)<br>for the homozygous<br>variant | <u>Vineis et al. (2006)</u>        |
| GenAir  | EPIC, Europe   | Several genes   | XRRC and BRCA variants in<br>gene–environment interactions in<br>lung cancer. More genes reported<br>associated with bladder cancer and<br>leukaemia |   | <u>Manuguerra et al.</u><br>(2007) |
| GenAir  | EPIC, Europe   | GSTM1, XRCC1  | Profile regression analysis of DNA<br>adducts showed increased risk from<br><i>GSTM1</i> null  |   | <u>Papathomas et al.</u><br>(2011) |
| Indoor air pollution<br>Coal exposure                               | Asia: 5 studies from<br>China and 1 from<br>Thailand | GSTM1, GSTT1, GSTP1   | <i>GSTM1</i> null genotype associated with cancer risk in 4 studies  | 0.001   | <u>Hosgood et al. (2007)</u>       |

Supplemental Table S26 Influence of genotype and lung cancer risk

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

| Supplemental Table 527 Influence of genetype and biomarkers in | humanc |
|--|--------|
| Supplemental Table S27 Influence of genotype and biomarkers in | numans |

| Exposure group  | Country              | Genotype/<br>phenotype method     | Result   | P value | Reference   |
|---|----------------------|-----------------------------------|--|---------|---|
| Bus drivers and garage<br>workers                         | Czech Republic       | Several genes                     | The carriers of at least one variant hOGG1 (Cys) allele<br>tended to higher oxidative damage to lymphocyte DNA<br>than those with the wild genotype, whereas XPD23 (Gln/<br>Gln) homozygotes were more susceptible to the induction<br>of DNA strand breaks. In contrast, <i>GSTM1</i> null variant<br>seemed to protect DNA integrity |         | <u>Bagryantseva et al.</u><br>(2010)                            |
| Police officers   | Czech Republic       | Several genes                     | The carriers of at least one variant allele, CYP1A1*2C (Ile/<br>Val), MTHFR 2656, or MS 2656, and the EPHX1-medium<br>phenotype appeared to be more susceptible specifically to<br>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,<br>GSTT1           | CYP1A1 m1 and GSTM1 null showed increased 8-OHdG   |         | <u>Prasad et al. (2013)</u>                                     |
| Outdoor air pollution<br>Bus drivers and mail<br>carriers | Denmark              | GSTM1<br>NAT2                     | DNA adducts: no significant effects<br>CAs: increased in bus drivers with <i>GSTM1</i> null and <i>NAT2</i><br>slow acetylator<br>CAs: increased in mail carriers with <i>NAT2</i> slow acetylator   | 0.0005  | <u>Nielsen et al. (1996b)</u> ,<br><u>Knudsen et al. (1999)</u> |
| Bus drivers   | Denmark              | Urinary <i>CYP1A2</i><br>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,<br>XRCC3              | DNA adducts: no significant effects  |         | <u>Palli et al. (2001)</u>                                      |
| Children exposed to urban air pollution                   | Bangkok,<br>Thailand | CYP1A1, GSTM1,<br>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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