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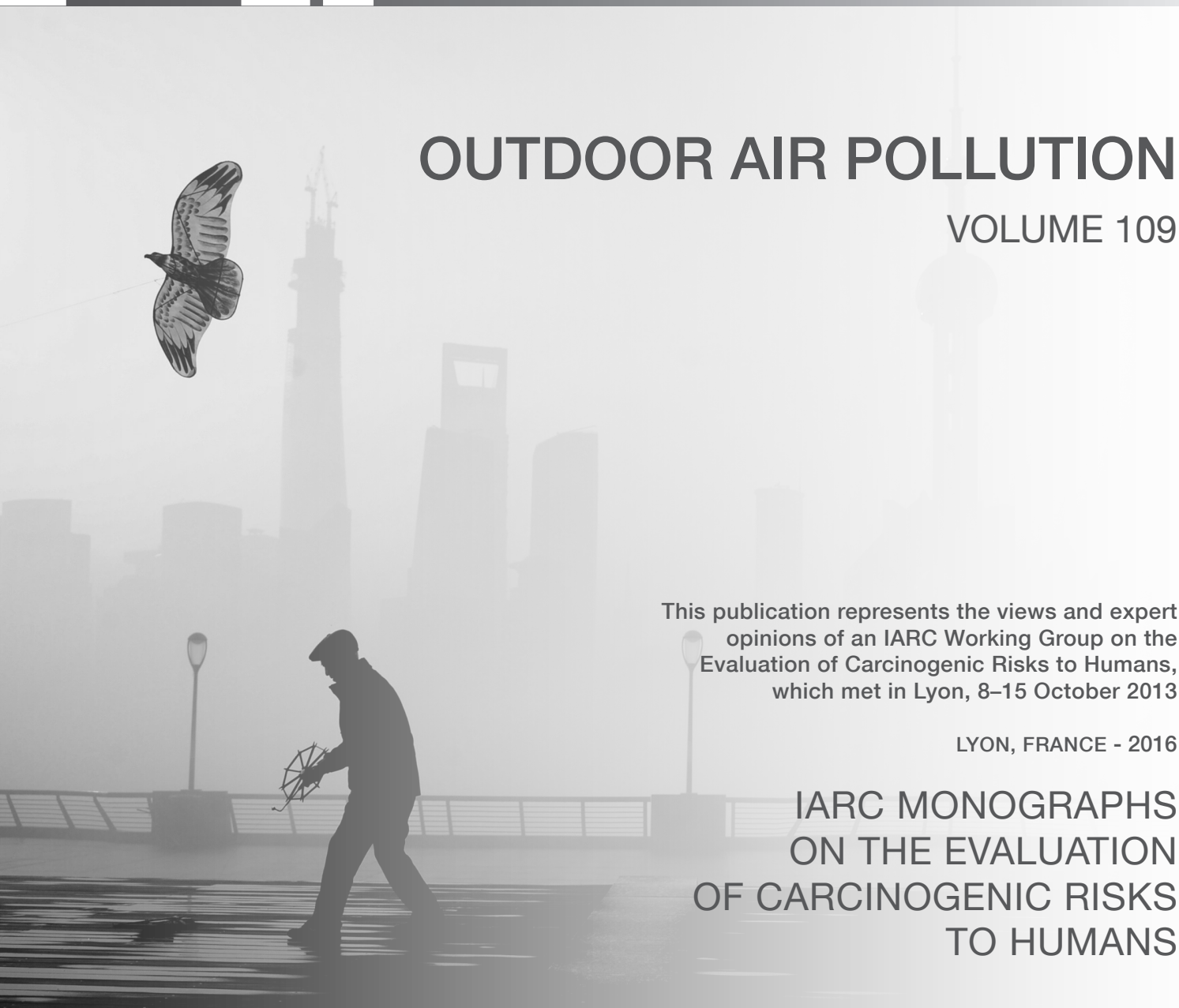

OUTDOOR AIR POLLUTION

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4. MECHANISTIC AND OTHER RELEVANT DATA

4.1 Toxicokinetic data

4.1.1 Introduction

The toxicokinetics of many of the major classes of compounds and industrial processes that contribute to outdoor air pollution have been described in previous *IARC Monograph* Volumes: carbon black, titanium dioxide, and talc (Volume 93; IARC, 2010a); household use of solid fuels and high-temperature frying (Volume 95; IARC, 2010b); non-heterocyclic polycyclic aromatic hydrocarbons (PAHs) (Volume 92; IARC, 2010c); diesel and gasoline engine exhausts (Volume 105; IARC, 2013); silica dust and asbestos (Volume 100C; IARC, 2012a); and benzene, butadiene, formaldehyde, coke production, iron and steel founding, and coal gasification (Volume 100F; IARC, 2012b).

4.1.2 Particulate matter

Some atmospheric particles are poorly soluble in water. As a consequence, after inhalation and deposition, they may remain present as particulate matter (PM) in the respiratory tract, and produce effects associated with particle toxicity.

The toxicokinetics, including deposition, clearance, and retention of poorly soluble particles and fibres, are discussed below for both humans and laboratory animals.

The deposition of *particles* within a region of the respiratory tract depends on the characteristics and physical factors that influence transport in the airways (e.g. air velocity and airway structure). The primary mechanisms for deposition of particles in the respiratory tract are sedimentation, impaction, and diffusion. Deposition by sedimentation and impaction depends on the aerodynamic diameter of the particle, whereas deposition by diffusion depends on the thermodynamic diameter of the particle (ICRP, 1994).

After inhalation, particles may either deposit in the extra-thoracic, tracheobronchial, or pulmonary/alveolar airways or remain in the airstream and be eliminated through exhalation. The deposition of particles in the respiratory tract depends primarily on the size of the inhaled particle, the route of breathing (i.e. through the nose and/or mouth), and the breathing pattern (i.e. volume and frequency). Particles of about 0.3 μm in diameter have minimal mobility in air; they are large enough that their diffusive mobility is minimal but are small enough that their sedimentation and impaction are also minimal. As a consequence, particles in this size range also have minimal deposition in the lung. In general, the deposition fraction for humans for most particle sizes smaller than 3–4 μm (aerodynamic diameter) is greater for the alveolar region than for the tracheobronchial airways. The deposition fraction decreases in the alveolar region for particles larger than 3–4 μm and smaller than 0.01 μm due to their removal in the extra-thoracic airways (particularly during nasal breathing) and tracheobronchial airways (NCRP, 1997; Maynard & Kuempel, 2005).

A few studies with human volunteers are available on the kinetics of clearance and retention of inhaled particles in the respiratory tract. Retention is determined by the balance between the rate of deposition and the rate of clearance. Particles that deposit in the tracheobronchial region are cleared by mucociliary clearance, which is relatively rapid (retention half-times of ~24–48 hours) (Oberdörster, 1988; ICRP, 1994), although some fraction of the particles that deposit in the airways is cleared more slowly than expected (Stahlhofen et al., 1995). For particles that deposit in the alveolar region, the primary mechanism of clearance is phagocytosis by alveolar macrophages, followed by migration of the macrophages to the terminal bronchioles and subsequent mucociliary clearance; the particles are eventually swallowed or expectorated. Particles that are cleared via the mucociliary escalator, whether from the tracheobronchial region or the alveolar region, and then swallowed will pass through the gastrointestinal tract and are subsequently cleared via the gut (Oberdörster, 1988; ICRP, 1994). Poorly soluble particles that deposit in the alveolar region are associated with a slow clearance phase (retention half-times of months to years in humans) (Bailey et al., 1985; Freedman & Robinson, 1988; ICRP, 1994). Translocation of particles to the interstitial region (interstitium) further increases the retention time of particles in the lungs (Oberdörster, 1988; Freedman & Robinson, 1988; ICRP, 1994). Some fractions of particles that deposit in the alveolar region may also be translocated to the lung-associated lymph nodes. Translocation may occur by transepithelial migration of alveolar macrophages after phagocytosis of the particle, or by translocation of free particles to the interstitium, where they may be phagocytosed by interstitial macrophages. Inflammation may alter mucociliary clearance, phagocytosis by alveolar macrophages, and the uptake and transport of particles to and through the respiratory epithelium (Oberdörster, 1988; ICRP, 1994).

The deposition and clearance of particles vary among individuals for several reasons, including age, sex, tobacco smoking status, and health status. Pre-existing lung diseases or conditions such as asthma or chronic obstructive pulmonary disease (COPD) can influence the efficiency and pattern of deposition within the respiratory tract. Deposition also depends on the level of activity and breathing patterns. Deposition and retention determine the initial and retained dose of particles in each region and may therefore influence the risk of developing diseases specific to those respiratory tract regions (Oberdörster, 1988; ICRP, 1994). Particles that are retained in the respiratory tract can lead to inflammation and oxidative stress (see Section 4.3.1).

4.1.3 Organic molecules

A detailed overview of the toxicokinetics of selected organic compounds is available in earlier *Monographs*: PAHs (IARC, 2010c), formaldehyde (IARC, 1982, 2006, 2012b), nitroarenes (IARC, 1984, 1989, 2013), and benzene (IARC, 1982, 2012b).

4.1.4 Inorganic gases

(a) Ozone

Ozone is a highly reactive gas, poorly soluble in water. The uptake and fate of ozone in the respiratory tract of humans and animals has been modelled by Miller (1995), Miller et al. (1985), Mercer & Crapo (1989), Pinkerton et al. (1992), and Pinkerton et al. (1995). Inhaled ozone can reach the major regions of the respiratory tract (extra-thoracic, tracheobronchial, and alveolar) and be absorbed there. Modelling based on the reactivity of ozone and its low solubility predicts that tissue doses will be low in the trachea, increase to a maximum in the terminal bronchioles, and decrease with further digital progression. Ozone can produce oxidative stress (see Section 4.3.1).

(b) Sulfur dioxide

Sulfur dioxide (SO_2) is a highly reactive, water-soluble gas and therefore is almost completely absorbed in the nasal passages of individuals at rest (IARC, 1992). With exercise, the pattern of SO_2 absorption shifts from the upper airways to the tracheobronchial airways in conjunction with a shift from nasal to oronasal breathing and increased ventilatory rates (EPA, 2006; Brain, 1970; Melville, 1970; Nodelman & Ultman, 1999). Similarly to ozone, the nasal passages remove SO_2 more efficiently than the oral pathway does.

(c) Nitrogen dioxide

Nitrogen dioxide (NO_2) is a reactive gas that is absorbed mainly in the upper respiratory tract, particularly the nasal passages, in individuals at rest. Exercise causes a shift from nasal to oronasal breathing and, because NO_2 is absorbed in the oral cavity less than in the nasal passages, more of the inhaled NO_2 reaches the pulmonary region, where the NO_2 is rapidly absorbed. Studies by Postlethwait and colleagues (Postlethwait et al., 1995, 1991; Postlethwait & Bidani, 1990) indicate that NO_2 absorption in the pulmonary region is due not to its solubility in the epithelial lining fluid of the lung but rather to interaction with constituents in this fluid, such as glutathione and ascorbic acid, in reactions mediated by free radicals. This may lead to oxidative stress (see Section 4.3.1).

4.1.5 Metals, inorganic dusts, and organic dusts

Outdoor air may contain different types of fibres and particles (e.g. asbestos and silica), dusts (e.g. wood dust), and various metals (e.g. beryllium, cadmium, chromium, nickel, and arsenic), including those reviewed in previous *Monographs* and classified as Group 1 human carcinogens (IARC, 2012a).

The presence of transition metals in outdoor air pollution may lead to the formation of reactive oxygen species (ROS), which can cause oxidative damage to DNA in the Fenton reaction. This comprises the reduction of hydrogen peroxide by a transition metal ion, resulting in the formation of the reactive hydroxyl radical and the oxidized metal ion. Transition-metal ions such as those of iron, copper, chromium, and nickel donate or accept free electrons via intracellular reactions and help in creating free radicals.

DNA is a target for metal ions due to its electron-rich structure, which offers ligands and complexation sites for positively charged metal ions. Ions such as copper(II) and iron(II) are able to interact with DNA in between the bases, and nickel(II) forms a complex with the phosphate backbone (Eichhorn & Shin, 1968), whereas chromium(III) ions are able to form stable adducts with DNA (Bridgewater et al., 1994). Cells treated with some metal ions under Fenton reaction conditions show enhanced levels of certain types of DNA damage (Imlay et al., 1988). Oxidized lesions in the form of DNA strand breaks and base modifications such as 8-oxo-7,8-dihydro-2′-deoxyguanosine (8-oxodG) have been observed after exposure of DNA to the Fenton reaction involving copper (Toyokuni & Sagripanti, 1994), iron (Toyokuni & Sagripanti, 1992), nickel (Kawanishi et al., 1989), and chromium ions (Tsou et al., 1996).

4.2 Genetic and related effects

Outdoor air is a complex aerosol composed of gases (nitrogen, oxygen, carbon dioxide [CO_2], ozone, NO_2, SO_2, etc.), water vapour, PM (including both organic and inorganic PM), volatile organic compounds (VOCs), and semivolatile organic compounds (SVOCs). The complex physical–chemical characteristics of the outdoor air matrix, combined with its spatial and temporal heterogeneity, complicate assessment of genetic and related effects in human and experimental

systems. Experimental installations (e.g. exposure chambers) have been used to expose human subjects to components of outdoor air such as fine PM (Brook et al., 2002); however, there are only few such assessments of genotoxic effects in experimentally exposed humans (Vinzents et al., 2005; Bräuner et al., 2007). Importantly, human studies have monitored genetic and related effects in individuals exposed to outdoor air under specific circumstances (e.g. outdoor occupational exposures); the results obtained are generally compared with effectively matched controls. [The Working Group noted that although examinations of exposures to outdoor air pollution in selected occupations (e.g. filling station attendants, bus drivers, airport tarmac workers) are relevant for hazard identification, the exposures may not be representative, in terms of both compositions and level, of exposures in the general population.]

Similarly, a wide range of studies have used in situ exposures, including of rodents, birds, and plants (e.g. *Tradescantia* sp.), to assess the genetic and related effects of outdoor air. For in situ studies, exposure is quantified as the duration of time spent at the location of interest. Most published research on genetic and related effects induced by outdoor air used in vitro systems, primary human and animal cells, established human and animal cell lines, yeast, and bacteria, as well as naked DNA in solution. These in vitro assessments involved exposures of cultured cells, DNA, or 2′-deoxynucleotides to PM suspensions, extracts of PM, or concentrates of SVOCs.

4.2.1 Mutagenicity

(a) Humans – in vivo studies

In a cross-sectional study of 67 mothers and 64 newborns from the Cracow region of Poland, Perera et al. (2002) found that the frequency of aromatic DNA adducts measured by ^{32}P-postlabelling was positively associated with the hypoxanthine–guanine phosphoribosyltransferase (*HPRT*) mutant frequency in the cord blood of newborns (P = 0.03) after controlling for exposure to smoking, diet, and socioeconomic status. There was no significant association between mutation and DNA damage in the peripheral blood lymphocytes of the mothers. This study demonstrated a molecular linkage between somatic-cell mutation in the newborn and transplacental exposure to air pollutants.

In Denmark, a strategic environmental health programme, including studies of exposures and biomarkers related to traffic-generated air pollution, was carried out in the late 1990s. Mutagenic activity in urine was measured as biomarker of exposure in non-smoking bus drivers in city and rural areas on a work day and a day off and in non-smoking mail carriers working outdoors (on the streets) and indoors (in the office). Urinary mutagenic activity was assessed by the Ames assay with *Salmonella* tester strain YG1021 and with the addition of S9 mix (exogenous metabolic activation system). Bus drivers had higher mutagenic activity in urine than mail carriers did; the individual levels of urinary mutagenic activity were not correlated with excretion of the biomarker of exposure, 1-hydroxypyrene (1-OHP). Among bus drivers, *N*-acetyltransferase 2 (*NAT2*) fast acetylators had higher mutagenic activity in urine than *NAT2* slow acetylators did, and female bus drivers had higher mutagenic activity than male bus drivers did (Hansen et al., 2004).

(b) Experimental systems

(i) In vivo studies

Animals

The Working Group did not identify any publications that assessed the induction of mutations in experimental animals (e.g. rodents) experimentally exposed to outdoor air or samples derived from outdoor air. However, as summarized in Supplemental Table S24 (available online), Yauk & Quinn (1996) and Yauk

et al. (2000) used multilocus DNA fingerprinting and pedigree analyses to assess the frequency of heritable genetic minisatellite mutations in herring gulls (*Larus argentatus*) collected from several locations affected by urban and/or industrial activities. The results obtained showed a significant > 2-fold increase in mutation rate at industrial sites compared with rural controls and, moreover, decreasing mutation rates with increasing distance from the highlighted industrial sources.

The follow-up studies used modified animal enclosures to expose mice to outdoor air in situ at selected urban/industrial locations (Somers et al., 2002, 2004; Yauk et al., 2008). Results were compared with matched exposures at rural control sites. This strategy offers the distinct advantage of real outdoor air exposures under semi-controlled conditions (e.g. animal housing and food); however, it is generally not possible to reliably estimate the actual dose (i.e. milligrams of PM or cubic metres of air per kilogram body weight [bw] per day). These studies have shown an increase in induced heritable mutations at expanded simple tandem repeat (ESTR) loci. High-efficiency particulate air (HEPA) filtration of outdoor air reduced heritable mutation rates (Somers et al., 2004; Yauk et al., 2008) (see also Section 4.3.3b for genotoxic effects on germ cells).

Plants

Studies in plants (see Supplemental Table S1, available online) assessed the ability of outdoor air or samples derived from outdoor air to induce genetic mutations or chromosomal damage. For instance, studies by Ferreira et al. (2000, 2003, 2007) reported the mutagenic activity of outdoor air at selected locations in or near the São Paulo (Brazil) metropolitan area.

(ii) In vitro studies

Table 4.1 provides a summary of studies that have used in vitro assays to assess the ability of outdoor air or samples derived from outdoor air to induce genetic mutations or gene conversion.

Human cells

Eight studies used human h1A1v2 cells to assess the induction of mutations at the thymidine kinase $TK^{+/-}$ locus (Hannigan et al., 1997, 1998, 2005; Durant et al., 1998; Pedersen et al., 1999, 2004, 2005; Adonis & Gil, 2000).

The results showed that the mutagenic potency of PM extracts for urban sites expressed per unit of equivalent organic carbon (EOC) are generally less than 2-fold greater than values obtained for rural (control) sites (Hannigan et al., 1997, 2005; Pedersen et al., 1999, 2004, 2005). However, when expressed per equivalent cubic metre of outdoor air, the potency values for urban sites were 3–10-fold higher than those for rural sites.

Detailed source apportionment revealed that diesel and natural gas combustion emissions make a large contribution to the mutagenic activity of outdoor air PM (Hannigan et al., 1997, 2005). In addition, detailed extract fractionation revealed that polar, semipolar (e.g. nitro-PAHs, ketones, and quinones), and non-polar (e.g. PAHs) extract fractions can each account for a substantial portion of the observed mutagenic activity per unit of EOC. For example, in their analysis of organic extracts of standard reference mixture (SRM) 1649, Durant et al. (1998) noted that semipolar and non-polar extract fractions accounted for 70% of the mutagenic activity. In their analyses of samples collected in southern California, Hannigan et al. (1998) noted that aromatic substances can account for more than 50% of extract mutagenic activity. In their analyses of sites in the north-eastern USA, Pedersen et al. (2004) noted that moderate-molecular-weight PAHs can account for 4–38% of observed mutagenic activity and that polar compounds (e.g. organic acids and hydroxy-polycyclic aromatic

Table 4.1 Genetic mutations associated with outdoor air pollution in human or animal cells in vitro

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Human cells					
Southern California, USA (1993)	Airborne PM from central Los Angeles, Azusa, Rubidoux, Long Beach, and a control site (San Nicolas Island). Collected on quartz-fibre filters using a virtual impactor. DCM Soxhlet extraction	Human lymphoblastoid h1A1v2 cell line (expressing human CYP1A1), 72 h exposure to extract in DMSO	Point mutations at *TK* locus in h1A1v2 cells	PM extracts induced a significant increase in *TK* mutations. Urban samples showed similar mutagenic potency [IMF × 10^6/µg of EOC (0.137–0.176)], slightly higher than for control site (0.095). Urban sites an order of magnitude more potent when expressed as IMF × 10^6/m^3 (0.670–0.900 vs 0.077). Detailed source analyses revealed that natural gas and diesel combustion made the largest contributions to outdoor air mutagenicity. 2-Nitrofluoranthene accounted for ~1% of observed mutagenic activity	Hannigan et al. (1997, 2005)
Washington, DC, USA (1976–1977)	Time-integrated (baghouse) urban PM collected in 1976 and 1977 (SRM 1649). DCM Soxhlet extraction. Bioassay-directed fractionation	Human lymphoblastoid h1A1v2 cell line (expressing human CYP1A1), 72 h exposure to extract in DMSO	Point mutations at *TK* locus in h1A1v2 cells	PM extracts (> 180 µg equiv PM per mL) induced a significant increase in *TK* mutations. 70% of activity in non-polar and semipolar fraction (A), 30% in polar fraction (B). 4% of fraction A mass accounted for 70% of mutagenic activity. Total of 13 PAHs accounted for 15% of mutagenic activity	Durant et al. (1998)
Southern California, USA (1993)	Urban PM from central Los Angeles, Azusa, Rubidoux, Long Beach, and a control site (San Nicolas Island). Composite PM collected on quartz-fibre filters using a virtual impactor. DCM Soxhlet extraction. Fractionation by normal-phase HPLC	Human lymphoblastoid h1A1v2 cell line (expressing human CYP1A1), 72 h exposure to extract in DMSO	Point mutations at *TK* locus in h1A1v2 cells	Extract of composite PM induced a significant increase in *TK* mutations (mutagenic potency, ~150 IMF × 10^6/mg of EOC). 46% of mutagenicity in non-polar fractions (1 and 2), 41% in semipolar fraction, 13.4% in polar fraction. Subfractionation of fraction 1 indicated that aromatic substances accounted for > 50% of total mutagenicity. Putative mutagens included cyclopenta[*cd*]pyrene, an important contributor to the observed response	Hannigan et al. (1998)

Table 4.1 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
North-eastern USA, 5 sites (1995)	Outdoor PM from 5 urban, suburban, and rural locations. Annual composites of PM (> 3 μm) collected on quartz-fibre filters using a virtual impactor. DCM Soxhlet extraction. Detailed chemical analyses	Human lymphoblastoid h1A1v2 cell line (expressing human CYP1A1), 72 h exposure to extract in DMSO	Point mutations at $TK^{+/-}$ locus in h1A1v2 cells	27 of 30 PM extracts induced significant increases in $TK^{+/-}$ mutations. Annual averages for IMF × 10^6/μg of EOC similar for 3 Boston sites (0.08–0.1) and similar for downtown and rural sites in Rochester, New York (0.16–0.19). Mutagenic activity IMF × 10^6/m^3 in urban areas 1.5–2-fold higher than in rural areas (e.g. 0.42–0.63). Mutagenic activity ~2-fold higher in winter than in summer for all sites. Known mutagens accounted for 16–26% of total mutagenic activity. PAHs accounted for 1–16% of mutagenic activity. 6*H*-benzo[*cd*]pyrene-6-one accounted for 3–5% of mutagenic activity	Pedersen et al. (1999, 2005)
North-eastern USA, 5 sites (1995)	Outdoor PM from 5 urban, suburban, and rural locations. Annual composites of PM (> 3 μm) collected on quartz-fibre filters using a virtual impactor. DCM Soxhlet extraction. Fractionation by normal-phase HPLC into 4 fractions of increasing polarity	Human lymphoblastoid h1A1v2 cell line (expressing human CYP1A1), 72 h exposure to extract in DMSO	Point mutations at $TK^{+/-}$ locus in h1A1v2 cells	Extracts of PM from 5 sites induced significant increases in $TK^{+/-}$ mutations (IMF × 10^6/μg of EOC, 0.05–0.3). Activity per m^3 of air in urban Boston ~3-fold higher than in rural area (0.66 vs 0.22). Bioassay-directed fractionation indicated semipolar fraction (nitro-PAHs, ketones, quinones) accounted for 35–82% of total mutagenic activity (per mg of EOC). Non-polar fraction (moderate-molecular-weight PAHs) accounted for 4–38% of mutagenic activity. Polar fraction (carboxylic acids, hydroxy-PACs) accounted for 14–32% of mutagenic activity	Pedersen et al. (2004)
Santiago, Chile (1996)	TSP from an urban, heavy-traffic site, collected on GFFs using a high-volume sampler. DCM sonication extraction	Human lymphoblastoid h1A1v2 cell line (expressing human CYP1A1), 72 h exposure to extract in DMSO	Point mutations at *TK* locus in h1A1v2 cells	Significant elevation in $TK^{+/-}$ mutation frequency for all tested concentrations. Mutagenic potency (400×10^6/m^3) 400-fold higher than reported for PM_{10} from Los Angeles in 1993. Mutation potency × 10^6/μg of EOC ~2-fold higher than for Los Angeles (0.300 vs 0.137)	Adonis & Gil (2000)
Animal cells					
Duisburg, Germany	Airborne PM from industrialized Rhine–Ruhr region. Draeger Box Micron filter. CX extraction	Chinese hamster V79 lung cells, 24 h exposure to extract in DMSO	*Hprt* mutations	Significant induction of *Hprt* mutations (i.e. 8-azaguanine-resistant colonies) for 4 m^3 and 8 m^3 equiv of air	Seemayer et al. (1987a, 1988)

Table 4.1 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Paris, France (1983–1985)	Airborne PM from urban site, collected on GFFs using a high-volume sampler. DCM or Ac sonication extraction	Chinese hamster V79 lung cells, 1–3 h exposure to extract in DMSO, with and without Aroclor 1254-induced rat liver S9	*Hprt* mutations	Significant, dose-related (per µg of EOM) increase in *Hprt* mutations (i.e. 6-thioguanine-resistant colonies); increase with exogenous S9 activation. Maximum induction, ~1.25 mutants per 10^6 survivors per m^3	Courtois et al. (1988)
Athens, Greece	Monthly PM samples collected on cellulose filters using a high-volume sampler. Hx sonication extraction	BALB/c 3T3 mouse embryonic fibroblast cells, 48 h treatment with extract in DMSO	Ouabain-resistant colony assay	Weak, non-significant induction of ouabain-resistant colonies	Athanasiou et al. (1987)

Ac, acetone; CX, cyclohexane; DCM, dichloromethane; DMSO, dimethyl sulfoxide; EOC, equivalent organic carbon; EOM, extractable organic matter; equiv, equivalent; GFFs, glass-fibre filters; h, hour or hours; HPLC, high-performance liquid chromatography; Hprt, hypoxanthine–guanine phosphoribosyl transferase; Hx, hypoxanthine; IMF, induced mutant fraction; PACs, polycyclic aromatic compounds; PAHs, polycyclic aromatic hydrocarbons; PM, particulate matter; PM_{10}, particulate matter with particles of aerodynamic diameter < 10 µm; SRM, standard reference mixture; TK, thymidine kinase; TSP, total suspended particles.

compounds [hydroxy-PACs]) can account for 14–32% of the observed mutagenic activity. Pedersen et al. (1999, 2005) noted that mutagenic activity per cubic metre is approximately 2-fold higher in the winter relative to the summer across all sites investigated.

Adonis & Gil (2000) used the *TK*$^{+/-}$ locus mutation assay in h1A1v2 cells to assess the mutagenic activity of an organic extract of PM collected from an urban, heavy-traffic site in Santiago, Chile. The reported mutagenic potency (expressed per cubic metre equivalent per assay millilitre) is more than 400-fold greater than that observed in Los Angeles (i.e. Hannigan et al., 1997, 1998, 2005). Comparisons based on potency expressed per unit of EOC revealed that extracts of PM from Santiago are approximately 2-fold more potent than extracts of PM from Los Angeles.

Animal cells

Several studies have demonstrated that organic extracts of outdoor air PM can induce significant dose-dependent increases in mutant frequency in cultured non-human mammalian cells. For instance, studies by Seemayer et al. (1987a, 1988) revealed that extracts of PM collected from the industrialized Rhine–Ruhr region in Germany induced a significant increase in *Hprt* mutant frequency. Similarly, Courtois et al. (1988) noted that organic extracts of PM collected from an urban location in Paris, France, induced a significant dose-dependent increase in *Hprt* mutant frequency. Source apportionment revealed noteworthy contributions by natural gas and diesel combustion emissions, and extract fractionation revealed substantial contributions from each of several chemical classes, including non-polar compounds (e.g. PAHs), semipolar compounds (e.g. nitro-PAHs and quinones), and polar compounds (e.g. organic acids and hydroxy-PACs).

(c) Yeast

Some extracts of outdoor air PM can induce point mutations, gene conversion, and mitochondrial mutations in yeast (see Supplemental Table S2, available online). In autumn and winter samples from 1993–1994, point mutation frequencies were increased more than 10-fold compared with untreated controls. Rossi et al. (1995) noted high variability across season and year, with no evidence of temporal decline between 1990 and 1994. Buschini et al. (2001) noted that toluene extracts were more mutagenic than acetone extracts and that smaller-sized PM (i.e. PM with particles of aerodynamic diameter < 2.5 μm [$PM_{2.5}$]) was frequently more mutagenic per microgram of PM equivalent than PM with particles of aerodynamic diameter < 10 μm (PM_{10}). A similar study by Bronzetti et al. (1997) revealed that organic extracts of PM collected from a high-traffic site in Pisa, Italy, elicited significant increases in gene conversions and point mutations per cubic metre in the presence of exogenous metabolic activation.

(d) Bacteria

More than 250 scientific publications addressed bacterial mutagenicity assays of outdoor air or samples derived from outdoor air (in North America, South America, Europe, Asia, or Oceania) (see Supplemental Tables S3–S9; Supplemental Figures S1 and S2, available online). These studies analysed organic extracts of airborne PM and/or concentrates of SVOCs collected on adsorbents. Several studies have shown that a modification of the pre-incubation assay known as the microsuspension assay provides enhanced sensitivity to combustion emissions such as those present in urban air PM (Kado et al., 1983, 1986; Agurell & Stensman, 1992).

Identification of putative mutagens

Bagley et al. (1992) and Gundel et al. (1993) noted substantial declines in mutagenic activity in TA98 strains deficient in nitroreductase (NR) (TA98NR). The results indicated a strong involvement of nitro-PAHs, in particular compounds such as the dinitropyrenes.

Many studies of organic extracts of PM from urban areas compared mutagenic potencies in the absence of S9 between TA98 and TA98NR to infer the involvement of nitroarenes in the observed responses. For example, analyses of extracts of PM collected in France, Japan, the Netherlands, and Sweden recorded a marked reduction in mutagenic activity in TA98NR without S9 relative to TA98 (Festy et al., 1984; de Raat & de Meijere, 1988; Takagi et al., 1992; Strandell et al., 1994).

A wide range of studies used the NR-deficient and *O*-acetyltransferase (OAT)-deficient versions of TA98 to examine the influence of nitroarenes in determining the mutagenic activity of organic PM extracts. These studies collectively examined extracts of PM collected from a wide variety of urban and/or industrial locations in Chile, Germany, Italy, Japan, Mexico, Norway, Spain, Sweden, and the USA (Alfheim et al., 1983; Tokiwa et al., 1983; Löfroth et al., 1985; Moriske et al., 1985; Wolff et al., 1986; Crebelli, 1989; De Flora et al., 1989; Takagi et al., 1992; Adonis & Gil, 1993, Espinosa-Aguirre et al., 1993; Gundel et al., 1993; Strandell et al., 1994; Casellas et al., 1995; Sato et al., 1995). Some of these studies noted dramatic reductions in mutagenic activity in the NR-deficient strain TA98NR relative to TA98. Some studies have noted that the relative decline in mutagenic potency of air samples in strain TA98NR relative to TA98 is seasonally variable (Erdinger et al., 2005).

Similarly, numerous studies have used metabolically enhanced *Salmonella* strains such as YG1021 (NR-enhanced) and YG1024 (OAT-enhanced) for comparative assessment of PM extracts. For example, studies of extracts of PM collected from urban and/or industrial areas in Brazil, Italy, Japan, Mexico, and Poland noted substantial increases in mutagenic activity in strains YG1021 and/or YG1024 without S9 (Espinosa-Aguirre et al., 1993; Yamaguchi et al., 1994; Jadczyk & Kucharczyk, 2005; Pereira et al., 2010; Traversi et al., 2011; Lemos et al., 2012). These studies frequently found that OAT enhancement contributed to larger relative increases in potency (without S9) compared with NR enhancement. For example, Yamaguchi et al. (1994) observed 3–4-fold increases in potency in YG1021 and 5–10-fold increases in potency in YG1024 for extracts of PM collected from a high-traffic site in Kobe, Japan.

In addition, numerous studies have used the TA98-derived frameshift strain YG1041, which overexpresses both NR and OAT, for analyses of extracts of PM from diverse regions (e.g. Brazil, the Czech Republic, Denmark, and Poland) (Binková et al., 2003; Sharma et al., 2007; Umbuzeiro et al., 2008; Piekarska et al., 2009). For example, Umbuzeiro et al. (2008) reported dramatic (> 30-fold) increases in the mutagenic activity in YG1041 relative to TA98 without S9 of organic extracts of PM collected from urban locations in São Paulo, Brazil. Extract fractionation confirmed that the highest activity in YG1041 without S9 was associated with nitro-PAHs. This study also indicated more modest, but nevertheless substantial, increases in mutagenic activity in OAT-enhanced strains in the presence of S9. This pattern of activity is thought to be associated with aromatic amines, including *N*-heterocyclics, as well as polar mutagens that, to date, have not been well characterized (e.g. oxy-PAHs) (Umbuzeiro et al., 2008).

Mutagenic potential and particle size

Studies from several geographical regions reported that the mutagenic activity of $PM_{2.5}$ extracts was greater per unit of extractable

organic matter (EOM) or per milligram of PM than that of PM_{10}.

Several studies have documented a noteworthy increase in potency with decreasing particle size. For instance, Kawanaka et al. (2004) found that approximately 90% of the mutagenic activity associated with extracts of PM collected in the Tokyo, Japan, area was associated with fine particles. In addition, studies that examined extracts of PM from Germany (Massolo et al., 2002) and Italy (Pagano et al., 1996; Monarca et al., 1997) specifically reported enhanced potency for the fine (< 1.5 μm) and/or ultrafine (< 0.5 μm) PM fractions. Nonetheless, small PM size fractions make relatively small contributions to atmospheric mutagenic activity per cubic metre due to low atmospheric levels (i.e. mass concentration) of fine and ultrafine PM per cubic metre.

Mutagenic potential of semivolatile organic compounds

In addition to detailed analyses of PM extracts, several studies also evaluated extracted SVOCs. For example, the study by Ciganek et al. (2004), which examined both PM and extracts from urban sites in Brno, Czech Republic, noted that extracts accounted for 15–40% of the total mutagenic activity.

Several studies have indicated that the similarities and differences between SVOC concentrates and PM extracts are affected by season, ambient temperature, adsorbent type, and the presence of S9. For example, in their study of samples collected in the Flanders region of Belgium, Du Four et al. (2004) observed that polyurethane foam extracts were more potent per microgram of EOM in the summer, whereas PM extracts were more potent in the winter. In contrast, Tuominen et al. (1988) noted that XAD extracts from samples collected in Helsinki, Finland, during the winter were generally more potent than PM extracts, with no appreciable difference between XAD and PM extracts from samples collected during the summer.

Spatial and temporal patterns in atmospheric mutagenic activity

Many studies reported markedly higher mutagenic potencies per cubic metre of air for extracts of PM collected during the colder months (winter and autumn) compared with those from the warmer months (spring and summer). Numerous studies conducted in Asia compared extracts of PM samples collected during different seasons and observed that winter and/or autumn samples were markedly more mutagenic than samples collected in spring and/or summer (e.g. Goto et al., 1982; Shimizu et al., 1982; Takagi et al., 1992; Qian et al., 1997; Qian & Zhang, 1997; Vinitketkumnuen et al., 2002). Similarly, studies conducted in European countries found comparable elevations in potency during colder months (Møller & Alfheim, 1980; Wullenweber et al., 1982; Alfheim et al., 1983; Athanasiou et al., 1986; Morozzi et al., 1992; Crebelli et al., 1995; Černá et al., 1999; Binková et al., 2003; Du Four et al., 2004; Piekarska et al., 2009, 2011). Finally, several studies conducted in North America, South America, and New Zealand also indicated elevations in mutagenic potency during colder months (Crebelli, 1989; Daisey et al., 1980; Brown et al., 2005; Cavanagh et al., 2009; Müller et al., 2001; Török et al., 1989).

Nevertheless, some studies from diverse geographical regions failed to detect any appreciable seasonal trend in mutagenic potency or noted that the mutagenic potency levels of extracts of PM collected in the summer were higher relative to those of winter samples (Commoner et al., 1978; Ohtani et al., 1985; Athanasiou et al., 1987; Adonis & Gil, 1993; Greenberg et al., 1993; Kuo et al., 1998).

The trend towards an increased atmospheric burden of PM-associated mutagens during colder months is quite clear and well substantiated. Several studies have found that the root causes of the observed seasonal trends may not be evident. Although some authors have pointed

towards contributions from fuel oil combustion for residential heating during winter months (Daisey et al., 1980), others have reported that the presence of atmospheric oxidants and the atmospheric transformation of nitroarenes are important determinants of seasonal fluctuation in PM mutagenic activity (Arey et al., 1988). Villalobos-Pietrini et al. (2006) noted the importance of a ground-level temperature inversion. Finally, other studies (Festy, 1980; Festy et al., 1984) support the contention that mutagens associated with winter PM are chemically different from mutagens associated with PM emitted during warmer months.

Day-to-day and diurnal variability

Marked day-to-day variability in mutagenic potency has been reported. For instance, a study in Sagamihara, Japan (Takagi et al., 1992), found lower potency on Sundays and holidays and concluded that vehicular emissions were significant contributors to the mutagenic activity of atmospheric PM. Other studies noted differences in mutagenic activity during the day compared with evenings (Møller et al., 1982; Gupta et al., 1996). A study by Kameda et al. (2004) of PM extracts collected in Osaka, Japan, reported peaks in potency in the early morning and late evening; potency corresponded with peaks in atmospheric levels of nitrogen oxide (NO), carbon monoxide (CO), and 1-nitropyrene. Some studies indicated that diurnal patterns varied with the season. For example, Shimizu et al. (1982) observed that daytime PM potency per cubic metre for samples collected in the centre of Tokyo, Japan, exceeded night-time PM potency for winter samples only. Conversely, Sakitani & Hayashi (1986) found that daytime potency exceeded nighttime potency for summer and autumn samples only.

Temporal trends

Two studies investigated temporal trends in PM-associated mutagenicity across an extended period of time. Matsumoto et al. (1998) monitored PM-associated mutagenic activity in Sapporo, Japan, between 1974 and 1992 and indicated a modest 44–50% temporal decline in mutagenic activity with exogenous metabolic activation. This corresponded with a marked 75–80% temporal decline in benzo[*a*]pyrene (B[*a*]P) adsorbed to PM (nanograms per cubic metre). Mutagenic activity without exogenous metabolic activation did not change over time. Similarly, Poli et al. (1999) examined PM-associated mutagenic activity in Parma, Italy, between 1991 and 1998 and reported a marked decline in mutagenic activity between 1992 and 1998.

Spatial variability

Many studies examined the spatial variability in the atmospheric burden of PM-associated mutagenic activity. The most common comparisons concern site-specific conditions related to urbanization, industrial activities, and/or traffic density. For example, many studies conducted in a wide range of locations (Brazil, the Czech Republic, Germany, Greece, Italy, Japan, the Netherlands, Poland, Saudi Arabia, Taiwan [China], Thailand, and the USA) have observed that the mutagenic potency per cubic metre of extracts of PM collected from urban sites near roadways and/or sites described as high-traffic sites is markedly higher relative to more rural reference sites (Preidecker, 1980; Athanasiou et al., 1986; de Raat & de Meijere, 1988; Yu et al., 1989; Wei et al., 1991; Vellosi et al., 1994; Sato et al., 1995; Černá et al., 1999; Vinitketkumnuen et al., 2002; Erdinger et al., 2005; Elassouli et al., 2007; Piekarska et al., 2011).

Elevated mutagenic activity per cubic metre was reported at residential areas located downwind of urban/industrial locations (e.g. de Raat, 1983). Some studies, including those conducted in China (Kong et al., 1994; Zhao et al., 1996), the Netherlands, (van Houdt et al., 1987), and Chile (Gil et al., 1997), observed substantial levels of PM-associated mutagenic activity per cubic metre at control sites, such as a suburban park.

Few studies have compared the mutagenic activity per cubic metre of PM extracts collected from different elevations. Comparisons of extracts of PM collected from ground level with samples collected from the same location at an elevated site, such as a rooftop, reported reduced potency at the site with higher elevation (e.g. Alfheim et al., 1983).

Effect of combustion

Viau et al. (1982) found an increase in the PM-associated mutagenic activity with S9 activation during "smoky" conditions caused by a forest fire in Kentucky, USA. de Andrade et al. (2011) reported that increased levels of PM-associated mutagenic activity were associated with cane-burning activities near São Paulo, Brazil. Similarly, al-Khodairy et al. (1998) noted that increased levels of mutagenic activity were associated with oil well fires in Kuwait. Nevertheless, several studies that examined levels of PM-associated mutagenic activity close to a suspected source (e.g. a municipal waste incinerator or an aluminium smelting operation) were unable to detect any appreciable influence of the source (e.g. Alfheim et al., 1984; Watts et al., 1989).

Several studies conducted fairly rigorous source apportionment and found that a substantial portion of PM-associated atmospheric mutagenicity is from mobile-source emissions (e.g. Israël & Busing, 1983; Lee et al., 1994; Hannigan et al., 2005). Other studies highlighted emissions from wood smoke as more important than mobile-source emissions (e.g. Claxton et al., 2001). Daisey et al. (1980) indicated that in their study in New York City, 50% of PM-associated atmospheric mutagenic activity was from fuel oil combustion for residential heating.

Effect of atmospheric pollutants

Numerous studies from diverse geographical regions have reported positive associations with atmospheric pollutants. These include lead, CO, nitrogen oxides (NO_x, NO and NO_2), SO_2, PAHs, and non-methane hydrocarbons. Associations have been found between PM-associated mutagenic activities and atmospheric lead, a pollutant associated with metal refining and founding and municipal waste incineration, or, for studies conducted before the mid-1990s (UNEP, 1999), with gasoline engine emissions (Flessel et al., 1985; Pitts et al., 1985). Significant associations between PM-associated mutagenicity and NO_x, an indicator of mobile-source emissions, have also been reported (Morris et al., 1995). Several studies have also highlighted associations with SO_2, an atmospheric pollutant associated with combustion of coal, residential fuel oil, and heavy fuel oils, such as marine fuel oil (Israël & Busing, 1983; Wolff et al., 1986; Morris et al., 1995). Finally, numerous studies have documented associations between atmospheric mutagenic activity and levels of atmospheric PAHs, including several known mutagens and/or mutagenic carcinogens (Viras et al., 1990; Černá et al., 1999).

Effect of meteorological conditions

Several studies have observed that meteorological conditions, such as wind speed and direction, temperature, precipitation, humidity, and solar penetration, can influence the levels of PM-associated atmospheric mutagenicity.

Several studies reported that PM-associated atmospheric mutagenicity per cubic metre was negatively affected by precipitation (rain or snow).

Because moving air masses can contain urban/industrial combustion emissions, it is perhaps not surprising that several studies have highlighted the role of wind speed and direction in determining levels of PM-associated atmospheric mutagenicity per cubic metre (e.g. Commoner et al., 1978; Wang et al., 1980). Detailed analyses (e.g. as conducted by Alink et al., 1983; de Raat et al., 1985; de Raat & de Meijere, 1988, and Morris et al., 1995) specifically identified wind directions that were associated with increased

levels of atmospheric mutagenicity. For example, studies conducted in the Netherlands found increased levels of PM-associated mutagenicity for easterly or southerly winds, from Germany and Belgium.

Post-emission formation of potent mutagens: atmospheric reactions

Studies that investigated the effects of meteorological conditions on atmospheric mutagenic activity are congruent with those of Arey et al. (1988, 1992) regarding the post-emission formation of potent mutagens derived from combustion emissions. Arey et al. showed that atmospheric reactions can contribute to the formation of potent nitro-PAHs. Such observations are consistent with mutagen formation during airborne movement from an urban/industrial area to a less-congested area, and with some atmospheric transformation products being mutagenic.

Conclusions

In summary, several fundamental conclusions can be drawn from the more than 250 studies that used the *Salmonella* reverse mutation assay to examine samples derived from outdoor air (e.g. PM extracts) collected from locations on five continents over the past 30 years.

1. Outdoor air PM extracts, including samples of total suspended particles (TSP), PM_{10}, and $PM_{2.5}$, evaluated for mutagenicity yielded a significant positive response; however, the mutagenic potency values, expressed per cubic metre, ranged over 5 orders of magnitude. Thus, some atmospheric samples clearly contain very little mutagenic activity, whereas others carry a high burden of activity.
2. The mutagenic potency of outdoor air is positively associated with outdoor air PM levels, reflecting an overall correspondence between declines in air quality and increased levels of PM-associated mutagenic activity. Increased PM-associated mutagenic activity is positively associated with other measures of impaired air quality, such as increased NO_x, lead, PAHs, CO, nitro-PAHs, and SO_2.
3. Samples derived from outdoor air PM collected during colder seasons (winter) are generally more mutagenic (per cubic metre) than those from PM collected during warm seasons (summer). This is likely due to a combination of factors that include changes in source contributions, meteorological changes, and seasonal land-use changes. Concomitantly, studies find that the PM-associated mutagenic potency per cubic metre of outdoor air is inversely related with air temperature.
4. Bioassay-directed fractionation studies confirm that much of the mutagenic activity associated with the particulate portion of outdoor air is found in the moderately polar and/or polar organic fractions, and includes a wide range of acids, bases, and neutral compounds, confirming a significant role for numerous classes of organic compounds. Although several noteworthy mutagens associated with outdoor air PM have been identified (e.g. nitro-PAHs), in most cases the putative mutagens have not been well characterized.
5. Samples derived from outdoor air PM are generally more mutagenic (per cubic metre) during workdays relative to non-workdays (e.g. weekends) and are generally more mutagenic during daytime than at night. The higher mutagenic potencies during workdays and daytime are associated with higher outdoor concentrations of lead, CO, and NO_x, reflective of mobile-source emissions.
6. Studies conducted over 7 years in Parma, Italy, showed a decline (63–76%) in outdoor air mutagenic activity (per cubic metre), reflecting the potential benefits of increasingly stringent emission controls for mobile combustion sources (Poli et al., 1999). A similar study conducted over 18 years in

Sapporo, Japan, noted a modest (44–50%) decline only for mutagenic activity commonly associated with PAHs (i.e. not nitro-PAHs) (Matsumoto et al., 1998).

7. Samples derived from outdoor air PM are generally more mutagenic if sampled at ground level, relative to higher elevations. Mutagenic activity is also greatly affected by wind direction and other meteorological conditions. Precipitation events reduce the levels of PM-associated mutagenic activity (per cubic metre).
8. Smaller particles ($PM_{2.5}$) are generally more mutagenic per mass of particle than larger particles (PM_{10}); maximum mutagenic activity is associated with particles of 0.1–1.2 μm.
9. The main contributors to PM-associated outdoor air mutagenicity appear to be urbanization, industrial activity, and traffic density. In some cases, wood smoke and/or emissions associated with residential heating have been highlighted as important sources of PM-associated mutagens, and these latter sources can exceed contributions from mobile-source emissions. Many studies have associated outdoor air mutagenicity with SO_2, which is associated with the combustion of coal, residential fuel oil, and heavy fuel oils, including marine fuel oil.

4.2.2 Cytogenetic effects

(a) Humans

Cytogenetic studies have directly evaluated the frequencies of chromosomal aberrations (CAs), micronuclei (MN), or sister chromatid exchanges (SCEs) among workers exposed to polluted outdoor air or heavy-traffic roads, compared with subjects exposed primarily to indoor air. In addition, a few studies have evaluated cytogenetic end-points in populations living in urban/industrial areas versus rural areas. Most such studies compared cytogenetic end-points in peripheral blood lymphocytes.

(i) Chromosomal aberrations

Table 4.2 summarizes the studies in which CAs were evaluated as a biomarker of outdoor air exposure versus subjects who worked or spent the majority of their time indoors. The studies reviewed cover several categories of workers exposed to outdoor air, and nearly all showed an association between CAs and this exposure, although this was the case for only two exposure groups when the data were stratified by genotype/phenotype (Knudsen et al., 1999). Four studies included exposure assessments, and all of them found higher levels of exposure among the subjects exposed to outdoor air than among the controls (Burgaz et al., 2002; Cavallo et al., 2006; Srám et al., 2007; Zidzik et al., 2007).

Among the studies included in Table 4.2, three studies (Anwar & Kamal, 1988; Knudsen et al., 1999; Cavallo et al., 2006) cultured the lymphocytes for 48 hours; however, three studies (Burgaz et al., 2002; Beskid et al., 2007; Sree Devi et al., 2009) cultured the cells for 72 hours, and one study for 69 hours (Chandrasekaran et al., 1996). Culturing cells for more than 48 hours can result in increased frequencies of CAs formed during the extended period of growth in culture. However, only one study with long culturing times (Sree Devi et al., 2009) appears to have an elevated frequency of CAs among the controls. Nonetheless, all of the studies in Table 4.2 reported significantly higher frequencies of CAs among the exposed relative to control populations.

Ten studies found increased frequencies of CAs among traffic police compared with their respective control populations (Table 4.2). Thus, traffic police in Cairo, Egypt, had higher frequencies of CAs compared with police trainers (Anwar & Kamal, 1988), as did those in Ankara, Turkey, compared with office workers (Burgaz et al., 2002), as did traffic police in Hyderabad,

Table 4.2 Chromosomal aberrations in peripheral blood lymphocytes of humans exposed to outdoor air pollution

Country	Control		Exposed		*P* value	Finding	Reference
	Description (*n*)	Result[a]	Description (*n*)	Result[a]			
Egypt	Police trainers (15)	0 ± 0	Traffic police (28)	0.4 ± 0.7	< 0.05	+	Anwar & Kamal (1988)
Turkey	Office workers (23)	0.26 ± 0.14	Traffic police (15)	1.29 ± 0.30	< 0.05	+	Burgaz et al. (2002)
	Office workers (23)	0.26 ± 0.73	Taxi drivers (17)	1.82 ± 0.34	< 0.01	+	Burgaz et al. (2002)
India	Non-traffic workers (115)	3.35 ± 1.21	Traffic police (136)	6.48 ± 1.67	< 0.05	+	Sree Devi et al. (2009)
China	Police working in offices (30)	0.4%	Traffic police (45)	0.98%	< 0.01	+	Chen et al. (1999)
Czech Republic	Indoor workers (49)	0.24 ± 0.18	Traffic police (50)	0.33 ± 0.25	< 0.05	+	Beskid et al. (2007)
Slovakia	Indoor workers (45)	0.21 ± 0.20	Traffic police (46)	0.30 ± 0.19	< 0.05	+	Beskid et al. (2007)
Bulgaria	Indoor workers (25)	0.13 ± 0.13	Traffic police (26)	0.25 ± 0.14	< 0.01	+	Beskid et al. (2007)
	Indoor workers (25)	0.13 ± 0.13	Bus drivers (25)	0.25 ± 0.18	< 0.05	+	Beskid et al. (2007)
Czech Republic	Indoor workers (50)	1.94 ± 1.28	Traffic police (52)	2.33 ± 1.53	> 0.05	–	Zidzik et al. (2007)
Slovakia	Indoor workers (55)	2.14 ± 1.61	Traffic police (51)	2.60 ± 2.64	> 0.05	–	Zidzik et al. (2007)
Bulgaria	Indoor workers (45)	1.79 ± 0.77	Traffic police (50)	3.04 ± 1.64	< 0.05	+	Zidzik et al. (2007)
	Indoor workers (45)	1.79 ± 0.77	Bus drivers (50)	3.60 ± 1.63	< 0.05	+	Zidzik et al. (2007)
Italy	Indoor airport workers (31)	0	Outdoor airport workers (41)	0.37	0.005	+	Cavallo et al. (2006)
Denmark	Low-exposure bus drivers (19)	Stratified by genotype	High-exposure bus drivers (55)	Stratified by genotype		+	Knudsen et al. (1999)
	Office workers (41)	Stratified by genotype	Postal workers (60) (mail carriers)	Stratified by genotype		+	Knudsen et al. (1999)
China	Chorionic villi in women in Dalian (827)	0.11	Chorionic villi in women in Shenyang (811)	1.66	< 0.0001	+	Cui et al. (1991)
			Chorionic villi in women in Zhengzhou (1060)	0.52	< 0.0001	+	
Czech Republic	Traffic police sampled in March (low pollution) (61)	0.16 ± 0.17 by FISH	Traffic police sampled in January (high pollution) (61)	0.27 ± 0.18 by FISH	< 0.001	+	Srám et al. (2007)
		1.84 ± 1.28		2.07 ± 1.48	> 0.05	–	

[a] Results are expressed as the percentage of lymphocytes with aberrations; all studies examined 100 metaphases per subject, except for Sree Devi et al. (2009), which studied 150 metaphases per subject. All studies used conventional staining, except for Beskid et al. (2007) and, where noted, Srám et al. (2007), which used fluorescence in situ hybridization (FISH) of chromosomes 1 and 4.

+, positive; –, negative.

India, compared with subjects who did not work in traffic (Sree Devi et al., 2009), as did traffic police in Hebei City, Henan, China, compared with police who worked in offices (Chen et al., 1999). The study in Turkey included an exposure assessment, which found higher concentrations of urinary 1-OHP among the traffic police relative to the control population (Burgaz et al., 2002). The study by Beskid et al. (2007) showed that police officers who worked outdoors in Prague (Czech Republic), Košice (Slovakia), or Sofia (Bulgaria) had higher frequencies of CAs as determined by fluorescence in situ hybridization (FISH) compared with subjects who were indoors at least 90% of the time. However, when traditional cytogenetic analyses were used, only the Sofia, Bulgaria, traffic police had elevated CA frequencies (Zidzik et al., 2007). An exposure assessment of these three sets of populations showed that the police who worked outdoors had higher exposures to carcinogenic PAHs compared with the controls (Zidzik et al., 2007). Using FISH, Srám et al. (2007) showed that traffic police in Prague, Czech Republic, had higher CA frequencies in January, when air pollution (PM_{10}) was high, than in March, when the PM_{10} concentration was significantly lower. However, no differences in CA frequencies were found when traditional cytogenetic methods were used.

One study performed in the Czech Republic (Rubes et al., 2005) evaluated the association between exposure of men to polluted outdoor air and CAs in their sperm, and found no association between aneuploidy in the sperm and outdoor air pollution. A study in traffic police in Prague, Czech Republic, showed that the police had higher frequencies of CAs in sperm when sampled in January, when the PM_{10} concentrations were high, than in March, when the PM_{10} concentrations were low (Srám et al., 1999) (see Section 4.3.3a).

The frequencies of CAs were higher in taxi drivers in Ankara, Turkey, compared with office workers (Burgaz et al., 2002; Table 4.2), and the taxi drivers had higher concentrations of urinary 1-OHP compared with the control subjects.

An investigation of outdoor airport workers at the international airport in Rome, Italy, found higher frequencies of CAs in this population compared with the frequencies found among airport office workers (Cavallo et al., 2006; Table 4.2). Exposure assessments found higher concentrations of PAHs in the air outdoors compared with in the offices, but there was no difference in the concentrations of urinary 1-OHP among the exposed and control groups.

In Denmark, a strategic environmental health programme, including studies of exposures and biomarkers related to traffic-generated air pollution, was carried out with bus drivers, letters carriers, and post office workers. A study of bus drivers categorized the exposure groups as high for drivers within the city of Copenhagen, medium for drivers in the suburbs, and low for drivers in the countryside (Knudsen et al., 1999; Table 4.2). When stratified by genotype/phenotype, those bus drivers who were glutathione *S*-transferase M1 (*GSTM1*) null and had the *NAT2* slow acetylator genotype exhibited an exposure-related increase in CAs compared with those with the *NAT2* fast acetylator and *GSTM1*-positive genotypes (Knudsen et al., 1999). [The role of genotype/phenotype is discussed later in this *Monograph* (see Section 4.4).] A separate study of bus drivers in Sofia, Bulgaria, found increased frequencies of CAs in that population relative to office workers when analysed either by traditional cytogenetic methods (Zidzik et al., 2007) or by FISH (Beskid et al., 2007).

A comparison of mail carriers in Copenhagen, Denmark, with office workers found that the mail carriers who were *NAT2* slow acetylators had higher frequencies of CAs compared with those who were *NAT2* fast acetylators (Knudsen et al., 1999; Table 4.2). This result suggested that the *NAT2* genotype may influence responses to other common exposures or may influence the

baseline frequencies of CAs (Knudsen et al., 1999).

Using white blood cells from 55 children attending a school in a rural area of Thailand and from 91 children attending an urban school in Bangkok, Thailand, Tuntawiroon et al. (2007) and Ruchirawat et al. (2007) exposed the cells to 100 cGy of ionizing radiation from a ^{137}Cs source at a dose rate of 5 Gy/minute and then determined the frequency of CAs. The authors found significantly higher frequencies of deletions/metaphase among the urban schoolchildren (0.45 ± 0.01) than among the rural schoolchildren (0.26 ± 0.01). Exposure analyses showed that the urban schoolchildren had higher concentrations of urinary 1-OHP and blood benzene compared with the rural schoolchildren. In addition, the air in the urban area had higher concentrations of PAHs and benzene than that from the rural area. Together, these studies indicated that the global DNA repair system of the urban schoolchildren was less effective at repairing the DNA damage after a challenge by ionizing radiation compared with that of the rural schoolchildren.

Cui et al. (1991) determined the frequency of CAs in the chorionic villi of 2698 women (aged 25–35 years) having abortions for non-medical reasons at less than 10 weeks of pregnancy from three cities with different levels of air pollution. The three cities were Shenyang, which had heavy pollution due to industry and coal combustion (811 women), Zhengzhou, which had moderate air pollution (1060 women), and Dalian, which was the least polluted, with light industry (827 women). The incidences of polyploidy, trisomy, and chromosome structural abnormalities in the women in Shenyang were 2.3, 3.4, and 16 times, respectively, those in the women in Dalian. Similarly, the cytogenetic frequencies of polyploidy, trisomy, and chromosome structural abnormalities in the women in Zhengzhou were 3.9, 1.3, and 4.9 times, respectively, those in the women in Dalian. The data for structural abnormalities are shown in Table 4.2. The results suggested that there was a positive correlation between the incidence of numerical and/or structural CAs and the severity of air pollution, especially SO_2 concentrations (Cui et al., 1991).

(ii) Micronuclei

The studies reviewed here cover a variety of exposure situations (Table 4.3), and of those that included exposure assessments, all found higher levels of exposure to air pollutants among the group exposed to outdoor air compared with the unexposed controls. Three studies evaluated MN in buccal cells (Karahalil et al., 1999; Hallare et al., 2009; Sellappa et al., 2010), and the rest evaluated MN in lymphocytes; however, one evaluated MN in both cell types (Cavallo et al., 2006), and one evaluated MN in maternal lymphocytes and cord blood (Pedersen et al., 2009). Most studies found increased frequencies of MN in the outdoor versus indoor exposure settings, or in populations living in urban/industrial areas versus rural areas. All lymphocyte studies except that of Zhao et al. (1998) used the cytokinesis-block version of the MN assay; those studies that used buccal cells did not (Karahalil et al., 1999; Cavallo et al., 2006; Hallare et al., 2009; Sellappa et al., 2010).

Six studies investigated the induction of MN in traffic police relative to controls not exposed chronically to traffic (Table 4.3). Traffic police in Ankara, Turkey, had higher frequencies of buccal cell MN compared with the controls (details of controls not specified) (Karahalil et al., 1999; Table 4.3). Likewise, higher frequencies of buccal cell MN were found in traffic police in Manila, Philippines, compared with other residents of Manila (Hallare et al., 2009; Table 4.3). Increased frequencies of MN were found in buccal cells of traffic police in Lanzhou, China, compared with the frequencies found in police who worked in offices (Zhao et al., 1998; Table 4.3).

A study of MN in lymphocytes in traffic police in Genoa, Italy, found increased frequencies in this group relative to a group of indoor workers

Table 4.3 Micronuclei in humans exposed to outdoor air pollution

Country	Tissues	No. of cells[a]	Control		Exposed		*P* value	Finding	Reference
			Description (*n*)	Result[b]	Description (*n*)	Result[b]			
Turkey	BC	3000	Workers (20)	0.03 ± 0.03	Taxi drivers (17)	0.12 ± 0.05	< 0.0001	+	Karahalil et al. (1999)
			Workers (20)	0.03 ± 0.03	Traffic police (15)	0.10 ± 0.05	< 0.05	+	Karahalil et al. (1999)
Philippines	BC	2000	City residents (18)	6.5	Filling station attendants (18)	18.9	< 0.05	+	Hallare et al. (2009)
					Traffic police (18)	17.07	< 0.05	+	Hallare et al. (2009)
China	PBL	2000	Police working in offices (49)	1.97 ± 0.21	Traffic police (65)	4.27 ± 0.68	< 0.05	+	Bai et al. (2005)
India	BC	3000	City residents (100)	2.79 ± 0.16	Filling station attendants (110)	12.61 ± 0.39	< 0.001	+	Sellappa et al. (2010)
China		1000	Police working in offices (34)	3.22 ± 1.31	Traffic police (67)	5.72 ± 2.57	< 0.05	+	Zhao et al. (1998)
Italy	PBL	2000	Indoor workers (54)	4.49 ± 2.0	Traffic police (94)	3.75 ± 1.65	0.02	+	Merlo et al. (1997)
Czech Republic	PBL	1000	Traffic police, in May (49)	4.37 ± 2.56	Traffic police, in February (49)	7.16 ± 3.50	< 0.001	+	Rossnerova et al. (2009)
Italy	BC	2000	Indoor airport workers (31)	Buccal: 0.064 ± 0.054	Outdoor airport workers (41)	Buccal: 0.064 ± 0.098	0.251	–	Cavallo et al. (2006)
	PBL	1000		Blood: 0.710 ± 0.421		Blood: 0.815 ± 0.37	0.129	–	
Italy	PBL	2000	Laboratory workers (34)	4.03	Traffic police (82)	3.73	> 0.05	–	Bolognesi et al. (1997a)
Denmark	CBL	2000	Low traffic density (23)	0.291 ± 0.178	High traffic density (23)	0.429 ± 0.206	< 0.01	+	Pedersen et al. (2009)
China	PBL	1000	Rural residents (63)	1.02	Urban residents (66)	1.56	< 0.05	+	Ishikawa et al. (2006)
Czech Republic	PBL	500	Rural children (12)	0.49 ± 0.20	Urban children (12)	0.70 ± 0.23	0.039	+	Pedersen et al. (2006)
China	PBL	1000	Shanghai Botanical Garden officers (36)	0.69 ± 0.60	Bus drivers or bus ticket officers on route through the Dapu tunnel in Shanghai (40)	1.28 ± 1.01	< 0.01	+	Peng & Ye (1995)

[a] Number of cells analysed per sample.

[b] Results are expressed as percentage of cells with micronuclei; in the Pedersen et al. (2006, 2009) studies, the results were expressed as ‰ cells.

+, positive; –, negative; BC, buccal cells; CBL, cord blood lymphocytes; PBL, peripheral blood lymphocytes.

(Merlo et al., 1997; Table 4.3). Exposure assessments found a 30-fold higher concentration of PAHs in the air outdoors compared with that in the office space. Another study in lymphocytes of traffic police in Genoa found no increase in MN frequencies in traffic police compared with a group of laboratory workers (Bolognesi et al., 1997a; Table 4.3). Nonetheless, exposure assessments showed higher levels of B[*a*]P in the air outdoors compared with that in the laboratories. Higher frequencies of MN were found among traffic police in Hebei, China, compared with police who worked in offices (Bai et al., 2005; Table 4.3), and exposure assessments found increased concentrations of inhaled particles in the air breathed by the exposed compared with the control subjects. Increased concentrations of NO_x, CO, hydrocarbons, and lead were also found among the exposed versus the control populations.

Unlike the previous studies of traffic police, the study by Rossnerova et al. (2009) evaluated traffic police in Prague, Czech Republic, but compared the MN frequencies among the police as measured in a more-polluted month (February) versus a less-polluted month (May). Exposure assessments had shown that the air in February had higher concentrations of carcinogenic PAHs, B[*a*]P, and various VOCs (benzene, ethylbenzene, and *o*-xylene). Traffic police had frequencies of MN that were higher in the more-polluted month (February) compared with those in the less-polluted month (May) (Table 4.3).

Filling station attendants in Manila, Philippines, had higher frequencies of buccal cell MN compared with residents of Manila (Hallare et al., 2009; Table 4.3). The frequencies of buccal cell MN were higher in filling station attendants in Coimbatore City, India, compared with other residents of Coimbatore City (Sellappa et al., 2010; Table 4.3). The frequencies of buccal cell MN were higher in taxi drivers in Ankara, Turkey, compared with control subjects (Karahalil et al., 1999; Table 4.3). [The Working Group noted that this human study addressed an occupational situation and might not be broadly applicable.]

No increases in either buccal or lymphocyte MN frequencies were found among outdoor airport workers at the international airport in Rome, Italy, compared with airport office workers (Cavallo et al., 2006; Table 4.3). Exposure assessments found that the concentration of PAHs was higher outdoors than in the offices, but concentrations of urinary 1-OHP were not different between the two groups of workers.

One study found higher frequencies of MN in lymphocytes of mothers and umbilical cord blood from those mothers who lived in high- versus low-traffic areas of Denmark (Pedersen et al., 2009; Table 4.3).

Several studies evaluated subjects living in urban/industrial areas versus rural areas. For example, Ishikawa et al. (2006) showed that residents of an industrial district of Shenyang, China, had higher frequencies of MN compared with residents of a rural district of the same city (Table 4.3). Likewise, Pedersen et al. (2006) found that young children living in an urban area (Teplice, Czech Republic) had higher frequencies of MN compared with young children living in a rural area (Prachatice, Czech Republic) (Table 4.3).

Another comparison of subjects working in two different environments was performed by Peng & Ye (1995), who measured the frequency of MN in bus drivers or on-site bus ticket officers a route that runs through a tunnel in Shanghai, China, and compared the results with those obtained in officers who worked in the Shanghai Botanical Garden. Daily average TSP concentrations in the tunnel were extremely high (1.86 mg/m^3) compared with the established standard of 0.15 mg/m^3. The bus drivers and on-site bus ticket officers had higher MN frequencies compared with the officers in the botanical garden (Table 4.3).

(iii) Sister chromatid exchanges

SCEs have been used extensively as a biomarker of genotoxicity; however, unlike CAs and MN, SCEs have not turned out to be predictive of cancer risk (Norppa et al., 2006). Nonetheless, SCEs are a sensitive indicator of exposure to a variety of genotoxic agents; as reviewed below, this includes exposure to outdoor air pollution.

There were 11 studies in which SCEs in lymphocytes were investigated as a biomarker associated with exposure to outdoor air pollution (Table 4.4). Of these 11 reports, all but two found increased frequencies of SCEs in the exposed population compared with the control subjects. Among the 11 reports, four exposure groups were studied; three of the 10 studies included exposure assessments, all of which found a difference between the exposure levels of the exposed and control populations. All of the studies cultured the lymphocytes for 72 hours, except for Sree Devi et al. (2009), where the cells were cultured for 70 hours, and Cavallo et al. (2006), where the cells were cultured for 48 hours.

There were eight studies of SCEs in traffic police, all but one of which found higher frequencies of SCEs in the traffic police relative to the control populations (Table 4.4); the one negative study included an exposure assessment. Traffic police in Cairo, Egypt, had higher frequencies of SCEs compared with police trainers (Anwar & Kamal, 1988); the same was true for traffic police in Madras, India, compared with subjects not working in traffic (Chandrasekaran et al., 1996). Traffic police in Lanzhou, China, had higher frequencies of SCEs compared with police who worked in offices (Zhao et al., 1998); the same was true for traffic police in Hyderabad, India (Sreedevi et al., 2006), or Chennai City, India (Anbazhagan et al., 2010), compared with office workers. Traffic police in Bangkok, Thailand, had higher frequencies of SCEs compared with university students, who were used as the control population (Soogarun et al., 2006). Although traffic police in Genoa, Italy, did not have elevated frequencies of SCEs compared with laboratory workers, used as controls (Bolognesi et al., 1997b), an exposure assessment found that the concentration of B[*a*]P and other PAHs in the outdoor air was higher than that in the laboratory spaces. Traffic police in Hebei, China, had higher frequencies of SCEs compared with police who worked in offices (Bai et al., 2005; Table 4.4), and exposure assessments showed that there were higher concentrations of particles, NO_x, CO, hydrocarbons, and lead in the air for the exposed populations compared with the controls.

Outdoor workers at the international airport in Rome, Italy, had higher frequencies of SCEs compared with indoor workers at the airport (Cavallo et al., 2006; Table 4.4), and exposure assessments found that the outdoor air had higher concentrations of PAHs than the indoor air. Nonetheless, there was no difference in the urinary concentration of 1-OHP between the outdoor and indoor workers. The frequencies of SCEs were not higher among tunnel workers in the Umbrian Apennine Mountains, Italy, compared with outdoor workers away from traffic (Villarini et al., 2008; Table 4.4).

A comparison of subjects working in two different environments was performed by Peng & Ye (1995), who measured the frequency of SCEs in bus drivers or on-site bus ticket officers on a route that runs through a tunnel in Shanghai, China, versus that of officers who worked in the Shanghai Botanical Garden. Daily average TSP concentrations in the tunnel were extremely high (1.86 mg/m^3) compared with the established standard of 0.15 mg/m^3. The bus drivers and on-site bus ticket officers had higher SCE frequencies compared with the officers in the botanical garden (Table 4.4).

In summary, two types of studies were performed to evaluate CAs, MN, and SCEs in humans exposed to outdoor air pollution. One type studied subjects whose work involved being outside (frequently in or near traffic) for

Table 4.4 Sister chromatid exchanges in lymphocytes of humans exposed to outdoor air pollution

Country	No. of metaphases[a]	Control		Exposed				
		Description (*n*)	Result[b]	Description (*n*)	Result[b]	*P* value	Finding	References
Egypt	40	Police trainers (10)	4.8	Traffic police (21)	7.5	< 0.10	+	Anwar & Kamal (1988)
India	25	Unexposed (23)	5.67 ± 0.37	Traffic police (23)	12.78 ± 0.68	< 0.001	+	Chandrasekaran et al. (1996)
Italy	50	Laboratory workers (35)	7.36 ± 1.35	Traffic police (54)	7.47 ± 1.28	> 0.05	–	Bolognesi et al. (1997b)
China	100	Police working in offices (34)	3.73 ± 1.51	Traffic police (67)	8.81 ± 1.83	< 0.05	+	Zhao et al. (1998)
Italy	50	Indoor airport workers (31)	3.84 ± 0.58	Outdoor airport workers (41)	4.61 ± 0.80	< 0.001	+	Cavallo et al. (2006)
India	50	Office workers (60)	4.18 ± 1.85	Traffic police (85)	9.31 ± 5.29	< 0.05	+	Sreedevi et al. (2006)
Thailand	NR	University students (20)	0.24 ± 0.12	Traffic police (30)	4.40 ± 0.93	< 0.05	+	Soogarun et al. (2006)
China	NR	Police working in offices (49)	2.69 ± 0.35	Traffic police (65)	4.32 ± 0.58	< 0.05	+	Bai et al. (2005)
Italy	30	Outdoor workers away from traffic (34)	4.88 ± 0.08	Tunnel workers (39)	5.07 ± 0.11	> 0.05	–	Villarini et al. (2008)
India	25	Office workers (25)	6.49 ± 0.31	Traffic police (56)	10.62 ± 0.57	< 0.001	+	Anbazhagan et al. (2010)
China	25	Shanghai Botanical Garden officers (36)	4.50 ± 0.99	Bus drivers or bus ticket officers on route through the Dapu tunnel in Shanghai (40)	5.94 ± 1.23	< 0.001	+	Peng & Ye (1995)

[a] Number of metaphases analysed per sample.

[b] Results are from blood cells and are expressed as sister chromatid exchanges per cell.

+, positive; –, negative; NR, not reported.

most of the workday (e.g. traffic police, street vendors, and toll booth operators) compared with subjects who worked primarily indoors (e.g. office workers). Another type of study compared subjects who lived or worked in more- versus less-polluted airsheds. Nearly all studies showed that polluted outdoor air induced significantly higher cytogenetic effects relative to either indoor air or less-polluted outdoor air. These studies covered 10 countries for CAs, seven for MN, and five for SCEs. Two of these end-points (CAs and MN) are associated with increased risk of cancer, highlighting the importance of these genotoxicity biomarker studies.

(b) Experimental systems

(i) In vivo

Animals

See Table 4.5.

Chromosomal aberrations

Several studies have examined the effect of outdoor air pollution or samples derived from it on cytogenetic abnormalities in experimental animals in vivo. Only one study examined the frequency of cytogenetic abnormalities in animals exposed in situ at locations highlighted for poor air quality. Rubeš et al. (1997) investigated cytogenetic effects in peripheral blood lymphocytes of dairy cattle in the Teplice district of the Czech Republic, an industrialized area with severe air pollution, and the Prachatice district, an agricultural area with lower levels of air pollution. The results revealed a significantly higher percentage of aberrant cells (chromatid aberrations or CAs) in animals in Teplice relative to Prachatice. The CAs included chromatid breaks, isochromatid breaks, acentric fragments, and translocations (Rubeš et al., 1997).

Four studies conducted in China examined the ability of extracts of airborne PM to induce various cytogenetic abnormalities in murine bone marrow. The study by Wang & Zhang (1984) was conducted in the northern Chinese city of Harbin, which experiences a marked reduction in air quality in colder months due to residential heating by coal. The authors reported that mice exposed orally by gavage to methanol extracts of TSP collected from a residential site in the winter showed a dose-dependent increase in aneuploidy and CAs. The CAs included chromosome breaks, fragments, dicentric chromosomes, and ring chromosomes (Wang & Zhang, 1984).

The other three studies were conducted in Taiyuan, an industrialized city in north-western China that contains chemical production facilities and coal-fired electricity generation facilities. Yang & Wu (1984) found that mice (strain not specified) treated with a single intraperitoneal injection of methanol extracts of TSP collected from several locations had dose-dependent increases in CAs in bone marrow cells. Marked increases were noted for samples from industrial, commercial, and residential sites, and the CAs included chromatid or chromosome breaks for the industrial or high-traffic sites and ring chromosomes for the commercial or residential sites. The authors observed that the frequency of induced CAs was positively associated with PM level (Yang & Wu, 1984).

A similar study in Taiyuan involved intraperitoneal exposures of mice to inorganic (i.e. nitric acid) PM extracts, or PM extracts prepared using simulated lung fluid. The results showed significant dose-dependent increases in CA frequency and increased responses for particles smaller than 2.5 μm. For the inorganic extract, the authors reported that the observed CA frequency was correlated with concentrations of lead, manganese, chromium, nickel, and cadmium (Lei et al., 1993). The study by Sun et al. (1995) investigated CAs in male germ cells of Kunming mice treated intraperitoneally with dichloromethane (DCM) extracts of TSP collected downwind of a coal-fired electricity generation facility. The results revealed dose-dependent increases in sperm abnormalities, CAs

Table 4.5 Cytogenetic damage associated with outdoor air pollution in experimental animals in vivo

Geographical location	Assay/exposure system	End-point(s) examined	Results	Reference
Bohemia, Czech Republic (1992, 1993)	Cytogenetic damage in bovine lymphocytes from cow herds on 5 farms in the industrialized Teplice district and 5 farms in the agricultural Prachatice district	Chromatid and chromosomal aberrations in lymphocytes, excluding gaps	Significant elevation in frequency of aberrant cells for 2 of the 3 time periods investigated	Rubeš et al. (1997)
Harbin, China (1981)	TSP, MeOH Soxhlet extraction. Five consecutive daily gavage doses in sunflower oil	Polyploidy and CAs in bone marrow cells (details not provided)	Dose-dependent increase in aneuploidy and CAs (e.g. breaks, fragments, dicentrics, rings)	Wang & Zhang (1984)
Taiyuan, China (1984)	TSP from 5 locations, MeOH extraction. Single i.p. injection in corn oil	Polyploidy and CAs in bone marrow cells (details not provided)	Dose-dependent increase in CAs; mainly breaks and rings for industrial, commercial, or residential sites. Control-site TSP extract also elicited a significant positive response. Significant correlation between CA frequency and PM level	Yang & Wu (1984)
Taiyuan, China (1990)	Size-fractionated airborne PM, extraction with nitric acid and SLF. Single i.p. injection	Polyploidy and CAs in bone marrow cells in Kunming mice 24 h after dose	Nitric acid and SLF extract induced dose-dependent increases in CAs. Greater responses for smaller size fractions. Significant induction for particles < 2 μm in 2.5 m^3	Lei et al. (1993)
Taiyuan, China (1994)	TSP from site downwind from a coal-fired power plant, DCM extraction. Five consecutive daily i.p. doses in corn oil	CAs in spermatogonia and primary spermatocytes, and MN in early spermatids in Kunming mice 24 h after final dose for CAs; 14 d for MN	Significant increase in CAs in spermatogonia and primary spermatocytes. Dose-dependent increase in MN in early spermatids. Dose-dependent increase in abnormal sperm morphology	Sun et al. (1995)
Upper Silesia, Poland (1984–1985)	Airborne PM collected on GFFs, BZ Soxhlet extraction. Two sequential daily i.p. doses	MN in bone marrow of Balb/c mice, examined 30, 48, and 72 h after final injection	Significant increase in frequency of micronucleated PCEs	Motykiewicz et al. (1996)
Sicily, Italy (1993)	Airborne PM from urban and rural locations collected on GFFs using a high-volume sampler, CX Soxhlet extraction. Daily i.t. instillations of PM extracts for 5 consecutive days	MN in Sprague-Dawley rat PAMs and epithelial cells 72 h after final instillation of extract representing 400 m^3	Significant increase in frequency of MN in PAMs and epithelial cells, relative to sham control, for PM extract from urban location only	Izzotti et al. (1996)
Rome, Italy (1986)	Airborne PM_{10} from urban locations collected on GFFs using a high-volume sampler, DCM Soxhlet extraction. Two consecutive daily i.p. injections of PM extracts	MN in bone marrow of Swiss mice 24 h and 72 h after final injection	No significant increase in MN frequency in PCEs. Significant decline in % PCE at highest dose 48 h after exposure	Crebelli et al. (1988); Crebelli (1989)

Table 4.5 (continued)

Geographical location	Assay/exposure system	End-point(s) examined	Results	Reference
Beijing, China (1990)	TSP from 4 locations, NM sonication extraction. Two consecutive daily i.p. injections of extracts in DMSO	MN in bone marrow of Kunming mice 6 h after final injection	Significant, dose-related increase in MN frequency for all sites except control (park). Greater response for samples from industrial or commercial sites, compared with residential	Wang et al. (1991)
Shanghai, China	TSP from 13 locations, DCM sonication extraction. Two consecutive daily i.p. injections in DMSO	MN in bone marrow of Kunming mice 6 h after final injection	Significant, dose-related increase in MN frequency for samples from 10 of 13 locations	Yao et al. (1993)
Shanghai, China (1992–1993)	Airborne PM from 13 urban sites, collected on GFFs using a high-volume sampler, DCM sonication extraction, 2 daily i.p. injections (3 doses)	MN in bone marrow of Kunming mice; cells examined 6 h after final treatment	Significant, dose-related increase in frequency of micronucleated PCEs. Maximum response ~5-fold increase above control	Zhao et al. (2002)
Taiyuan, China	TSP samples from residential area, DCM sonication extraction, separated into 5 fractions, 2 consecutive daily i.p. injections	MN in bone marrow of Kunming mice; cells examined 6 h after final treatment	Significant, dose-related increase in MN frequency for all fractions except aliphatic hydrocarbons. Highest response for acidic fraction, followed by polar aromatics, basic fraction, and PAH fraction	Bai et al. (1999)
Taiyuan, China	TSP samples from foundry site and control site, DCM sonication extraction, single i.p. injection in DMSO	MN in bone marrow of Kunming mice (details not provided)	Significant, dose-related increase in MN frequency for both samples; greater response for extract of TSP from foundry site relative to control site	Zhang et al. (2002)
Lanzhou, China (1997)	TSP samples from heavy–traffic site and control site, DCM sonication extraction, 4 consecutive daily gavage doses in DMSO	MN in bone marrow of Kunming mice; cells examined 24 h after final treatment	Significant, dose-related increase in MN frequency	Zhao et al. (2001)
São Paulo state, Brazil	In situ 120 d exposures of caged Balb/c mice at a high-traffic urban location and a rural reference site	MN in peripheral blood on days 15, 30, 60, 90, and 120	ANOVA showed significant effect of treatment site and treatment time, with highest MN frequency observed at urban site after 90 d. Significant rank correlation of MN frequency and CO, NO_2, and PM_{10} for urban site	Soares et al. (2003)
Tokyo, Japan (1983)	Airborne PM_{10} collected on GFFs using a high-volume sampler, MeOH extract and MeOH:water, CX and nitromethane extract fractions. Four consecutive i.p. doses	MN in PCEs of Balb/c mice, 48 h after a single injection	Significant, dose-related increase in MN frequency for crude extract. Highest activity in nitromethane fraction	Sakitani & Suzuki (1986)

Table 4.5 (continued)

Geographical location	Assay/exposure system	End-point(s) examined	Results	Reference
West Virginia, USA (1984)	Airborne PM collected on GFFs using a high-volume sampler, Ac extraction. Single i.p. or p.o. doses of PM extracts in DMSO (5 dose levels)	SCEs in bone marrow and spleen cells of CD_1 mice	No significant increase in SCE frequency in either tissue	Krishna et al. (1986)

Ac, acetone; ANOVA, analysis of variance; BZ, benzene; CAs, chromosomal aberrations; CO, carbon monoxide; CX, cyclohexane; d, day or days; DCM, dichloromethane; DMSO, dimethyl sulfoxide; GFFs, glass-fibre filters; h, hour or hours; i.p., intraperitoneal; i.t., intratracheal; MeOH, methanol; MN, micronuclei; NM, nanomaterial; NO_2, nitrogen dioxide; PAHs, polycyclic aromatic hydrocarbons; PAMs, pulmonary alveolar macrophages; PCEs, polychromatic erythrocytes; PM, particulate matter; PM_{10}, particulate matter with particles of aerodynamic diameter < 10 μm; p.o., oral gavage; SCEs, sister chromatid exchanges; SLF, simulated lung fluid; TSP, total suspended particles.

in spermatogonia and primary spermatocytes, and frequencies of meiotic MN in early spermatids (Sun et al., 1995) (see also Section 4.3.3).

Micronuclei

Eleven published studies examined the ability of outdoor air or samples derived from outdoor air to induce MN in vivo. The majority of these studies (8 of 11) investigated the frequency of MN in haematopoietic tissues (bone marrow or peripheral blood) in mice exposed intraperitoneally to a single acute dose or to repeated (2–5) consecutive daily doses. One study examined the frequency of MN in peripheral blood of Balb/c mice exposed in situ for up to 120 days to urban air in São Paulo (Soares et al., 2003). The results revealed a significant increase in MN frequency relative to a rural control location, and MN frequency was positively correlated with atmospheric levels of CO, NO_2, and PM_{10} (Soares et al., 2003).

The study by Izzotti et al. (1996) examined the ability of cyclohexane extracts of TSP collected in Sicily, Italy, from urban and rural locations to induce MN in rat alveolar macrophages and pulmonary epithelial cells after five consecutive daily intratracheal instillations. The results showed significant 3.4-fold and 4.5-fold increases in MN frequency in pulmonary alveolar macrophages and epithelial cells, respectively, of rats treated with extracts from urban locations relative to the control (Izzotti et al., 1996). Zhao et al. (2001) noted that four consecutive gavage doses of DCM extracts of TSP collected at a heavy-traffic location in Lanzhou, an industrialized city in north-western China, induced a significant dose-dependent increase in MN frequency in bone marrow of Kunming mice (Zhao et al., 2001).

Two studies in Europe investigated the ability of PM extracts to induce increases in MN frequency in mouse bone marrow after intraperitoneal injection. Motykiewicz et al. (1990, 1996) showed that benzene extracts of PM collected from the heavily industrialized region of Upper Silesia, Poland (which has coke production, metal refining, steel foundries, etc.), induced a significant increase in micronucleated polychromatic erythrocytes (PCEs) in Balb/c mice after two consecutive intraperitoneal injections. Crebelli (1989) reported that two consecutive intraperitoneal doses of DCM extract from PM collected in Rome, Italy, failed to elicit significant increases in micronucleated PCEs in Swiss mice (Crebelli et al., 1988).

Six studies investigated the ability of organic extracts of PM collected in urban centres in China (Beijing, Shanghai, and Taiyuan) to induce significant increases in MN frequency in bone marrow of Kunming mice exposed via intraperitoneal injection. For example, Wang et al. (1991) noted that nanomaterial extracts of TSP from several sites in Beijing induced dose-dependent increases in MN frequencies and, moreover, that MN frequencies were markedly higher for samples from industrial or commercial areas relative to those from residential areas (Wang et al., 1991). Similarly, studies by Yao et al. (1993) and Zhao et al. (2002) revealed that DCM extracts of PM collected from a variety of locations in Shanghai induced significant dose-dependent increases in MN frequency for 10 of the 13 locations examined, with a maximum response approximately 5-fold above the control (Yao et al., 1993).

Studies by Bai et al. (1999) and Zhang et al. (2002) investigated the ability of DCM extracts of TSP collected in Taiyuan to induce a significant increase in MN frequency. Bai et al. (1999) found dose-dependent increases in MN frequency, and extract fractionation showed no induction of MN by the aliphatic hydrocarbon fraction but induction of high MN frequencies by fractions containing organic acids, polar aromatics, basic organics, and PAHs (Bai et al., 1999).

Zhang et al. (2002) studied a site near the Taiyuan steel foundry and compared results with those obtained from samples from a less-contaminated site at Yangqu. They found a significant

induction of MN by air sample extracts from both sites, but there was a marked increase for extracts of TSP from the foundry area. The authors observed that the results correspond to a higher incidence of lung cancer in the Taiyuan foundry area relative to the control site (Zhang et al., 2002).

A single study in Japan reported a significant dose-related increase in micronucleated PCEs in Balb/c mice exposed to a methanol extract of PM_{10} collected in Tokyo (Sakitani & Suzuki, 1986).

Sister chromatid exchanges

A single study that investigated the ability of organic extracts of PM collected in West Virginia, USA, to induce SCEs in bone marrow and spleen cells of CD1 mice exposed via single intraperitoneal or oral administration failed to show a significant increase relative to control (Krishna et al., 1986).

In summary, polluted outdoor air, outdoor air PM, or samples derived from outdoor air PM are capable of inducing significant increases in cytogenetic damage in animals exposed in situ or exposed experimentally via a variety of routes of administration. Exposures of experimental animals via oral, intraperitoneal, or intratracheal administration show a clear dose-dependent induction of cytogenetic damage recorded as CAs or MN. [The Working Group expressed concerns about intraperitoneal injections and their relevance to human cancer risk.]

Plants

Plant assays (see Supplemental Table S10, available online) have also been used to assess the ability of outdoor air pollution or samples derived from it to induce cytogenetic damage (Ma et al., 1994). In particular, many studies have examined the induction of MN in meiotic pollen mother cells (i.e. tetrads formed after the second meiotic division) of the sterile *Tradescantia* clone 4430 or isolates of *T. paludosa* or *T. pallida* exposed to outdoor air in situ for extended periods (e.g. several months) or in the laboratory to extracts of airborne PM.

In their review of the mutagenicity and carcinogenicity of outdoor air pollution, Claxton & Woodall (2007) observed that although the dynamic range of plant genotoxicity assays varies across plants and end-points, the dynamic range of the induced responses, relative to the control, for the popular *Tradescantia* assays ranges between 2-fold for the stamen-hair mutation assay and 20-fold for the MN assay. In their review of mutagens in contaminated soils, White & Claxton (2004) also critically examined the utility of the *Tradescantia* genotoxicity assays and noted the limited dynamic range and lack of sensitivity, particularly for short-term exposures.

(ii) In vitro

Cytogenetic effects induced by extracts of airborne particulates were assessed in cultured human lymphocytes, human cell lines, cultured animal primary cells, and animal cell lines. The cytogenetic effects included CAs, aneuploidy, MN, and SCEs. The results of these in vitro cytogenetic studies are summarized in Table 4.6.

Chromosomal aberrations

Human cells

A series of studies showed significant dose-related increases in the frequency of chromosome and chromatid breaks in cultured human lymphocytes exposed to organic extracts of airborne PM from the Rhine–Ruhr region of Germany (Hadnagy et al., 1986, 1989; Hadnagy & Seemayer, 1987; Seemayer et al., 1989). Acetone extracts of outdoor air PM collected in West Virginia, USA, also induced CAs in human lymphocytes in a dose-dependent manner (Krishna et al., 1984). Three studies in China also showed that extracts of outdoor air particles induced CAs in human lymphocytes. Organic extracts of airborne TSP samples collected at five sites in Lanzhou, a city heavily contaminated by

Table 4.6 Cytogenetic damage associated with outdoor air pollution in human and animal cells in vitro

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Human cells					
Rhine–Ruhr region, Germany	Airborne PM from Düsseldorf, collected on GFFs using a high-volume sampler. DCM extraction	Cultured human lymphocytes, 48 h or 72 h exposure to extract in DMSO	CAs with and without gaps	Significant, dose-related (equiv m^3/mL) increase in frequency of chromosome breaks. Chromatid breaks observed at low concentration only	Hadnagy et al. (1986), Hadnagy & Seemayer (1987)
West Virginia, USA	Airborne PM collected on GFFs using a high-volume sampler. Ac extraction	Cultured human lymphocytes, 10 h exposure to extract in DMSO	CAs with and without gaps	Significant, dose-related (mg of PM equiv/mL) increase in CA frequency (both with and without gaps)	Krishna et al. (1984)
Lanzhou, China	Airborne TSP from 5 sites with varying levels of air pollution. DCM sonication extraction	Cultured human lymphocytes, 50 h exposure to extract in DMSO	CAs with and without gaps	Significant, dose-related increase in CA frequencies for all TSP extracts. Highest response for suburban locations downwind from urban site and industrial site	Ding et al. (1999)
Shanghai, China	Airborne TSP from the Dapu tunnel. NaCl (saline) shaker extraction	Cultured human lymphocytes, exposure to saline extract	CAs	Significant, exposure-related increase in CA frequency. Effect similar to Japanese vehicle exhaust PM standard (NIES-8)	Tan et al. (2002)
Baotou and Wuwei, China (Mongolia)	$PM_{2.5}$ samples collected during sandstorm and control (non-storm) days. $PM_{2.5}$ suspension in saline	Cultured human lymphocytes, exposure to saline PM suspension	CAs with and without gaps	Significant, dose-related increase in CA frequency for samples from both cities during storm and non-storm conditions. For non-storm conditions, suspensions of $PM_{2.5}$ from industrial Baotou elicited higher CA frequencies	Wei & Meng (2006a)
Suwon, Republic of Korea	$PM_{2.5}$ collected in high-traffic area on Teflon-coated filter using a cascade impactor. DCM sonication extraction. Acid–base–neutral fractionation and subfractionation on silica	BEAS-2B human lung bronchial epithelial cells, 24 h treatment with extract in DMSO	Cytokinesis-block MN assay	Significant, dose-related (μg of EOM/mL) increase in MN frequency for crude extract. Significant increases in MN frequency for aliphatic, aromatic (i.e. PAHs and alkyl-PAHs), and semipolar (nitro-PAHs, ketones, quinones) fractions	Oh et al. (2011)
Mexico City metropolitan area, Mexico	PM_{10} from industrial and residential locations, collected on GFFs using a high-volume sampler. Water and DCM Soxhlet extraction	A549 human alveolar adenocarcinoma cells, 48 h exposure to extract in DMSO	Cytokinesis-block MN assay	Significant, dose-related (μg of PM_{10} equiv/mL) increases in MN frequency for water and DCM extracts for PM from both residential and industrial areas	Roubicek et al. (2007)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
L'Aquila, Italy	Airborne PM of 2.5–10 µm and 0.4–2.5 µm, collected using an 8-stage cascade impactor	Hs27 human skin fibroblasts, 44 h exposure to suspended PM	Cytokinesis-block MN assay	Significant, dose-related (m^3/mL) increases in MN frequency for samples collected in 6 sequential months. MN frequency values for fine PM slightly greater than for coarse PM	Poma et al. (2002)
Lanzhou, China	Airborne TSP from 5 sites with varying levels of air pollution. DCM sonication extraction	Cultured human lymphocytes, 52 h exposure to extract in DMSO	MN in harvested cells	Dose-dependent increase in MN frequency for all particle extracts. Highest response for suburban locations downwind from urban site and industrial site	Ding et al. (1999)
Taiyuan, China	Airborne particulates from a residential area, 6 size classes. Acid Soxhlet extraction, and sequential Soxhlet extraction with MeOH, Ac, and DCM	Cultured human lymphocytes, exposure to acid extract or pooled organic extract	Cytokinesis-block MN assay	For both types of extracts, dose-dependent increase in MN frequency for all samples, with enhanced responses for extracts of smaller particles. MN frequency elicited by acid extract positively correlated with metal (e.g. nickel, cadmium, chromium) concentrations	Yuan et al. (1999a, b)
Shanghai, China	Airborne PM from industrial Taopu area. NaCl (saline) shaker extraction	Cultured human lymphocytes, exposure to saline extracts	MN in harvested cells	Dose-dependent increase in MN frequency	Tan et al. (2004)
Baotou and Wuwei, China (Mongolia)	$PM_{2.5}$ samples collected during sandstorm and control (non-storm) days. NaCl (saline) PM suspension, NaCl shaker extraction, DCM Soxhlet extraction	Cultured human lymphocytes, exposures to PM suspensions, saline extracts, organic extracts	Cytokinesis-block MN assay	Dose-dependent increase in MN frequency elicited by PM suspensions and organic extracts. Higher MN frequency elicited by samples collected on non-storm days. For storm conditions, no significant difference between industrial Baotou and agricultural Wuwei	Wei & Meng (2006a), Wei et al. (2006)
Guangzhou, China	TSP and PM_{10} from a residential area. DCM sonication extraction. Fractionation by chromatography	Cultured human lymphocytes, exposure to extract in DMSO	Cytokinesis-block MN assay	TSP extracts elicited significant increase in MN frequency. No dose–response. Aromatic hydrocarbon fraction of PM_{10} extract elicited significant increase in MN frequency	Xu & Wang (2008)
Flanders, Belgium (2000)	PM_{10} from urban, rural, and industrial sites, collected on GFFs with a low-volume sampler. ASE extraction with THF:Hx (20:80)	Cultured human lymphocytes, 72 h exposure to extract in DMSO	Cytokinesis-block MN assay	Significant, dose-related (equiv m^3/mL) increase in MN frequency for urban location only. Urban PAH concentration (ng/m^3) higher than that for rural and industrial sites	Brits et al. (2004)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Silesia, Poland (1984–1985)	Airborne PM, collected on GFFs. BZ Soxhlet extraction	Cultured human lymphocytes, 72 h exposure to extract in DMSO	SCE assay	Small, dose-related increase in SCE frequency	Motykiewicz et al. (1990)
Lexington, Kentucky, USA (1980)	Airborne PM before and during forest fires, collected on GFFs using a high-volume sampler. Sonication extraction with BZ and Ac	Cultured human lymphocytes, exposure to extract in DMSO	SCE assay	Strong, dose-related (m^3 or mg of PM equiv) increase in SCE frequency for "smoky" conditions. Weak, significant response for "non-smoky" conditions	Viau et al. (1982)
Lanzhou, China	Airborne PM from a district with high incidence of lung cancer. DCM sonication extraction	Cultured human lymphocytes, 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related increase in SCE frequency	Zhang & Li (1994)
Lanzhou, China	Airborne TSP from 5 sites with varying levels of air pollution. DCM sonication extraction	Cultured human lymphocytes, 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related increase in SCE frequency for all PM extracts. Higher responses for suburban locations downwind from urban site and industrial site	Wang & Ding (1998)
Duisburg, Germany	Airborne PM from industrialized Rhine–Ruhr region. Draeger Box Micron filter. CX extraction	Cultured human lymphocytes, 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related (m^3 equiv/mL) increase in SCE frequency per metaphase in both cell types	Seemayer et al. (1987a, 1988)
Rhine–Ruhr region, Germany	Airborne PM from Duisburg and Düsseldorf, collected on GFFs using a high-volume sampler. DCM extraction	Cultured human lymphocytes, 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/mL) increase in SCE frequency for samples from both Düsseldorf and Duisburg. Higher response for heavily industrialized area (Duisburg), relative to urban. As little as 0.3 m^3 equiv of Duisburg extract required to elicit a significant increase in SCE frequency	Hadnagy et al. (1989), Seemayer et al. (1989, 1990a)
Rhine–Ruhr region, Germany	Airborne PM from Düsseldorf, collected on GFFs using a high-volume sampler. DCM extraction	Cultured human lymphocytes, 48 h or 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/mL) increase in SCE frequency	Hadnagy et al. (1986), Hadnagy & Seemayer (1987)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Rhine–Ruhr and North Rhine-Westphalia regions, Germany	Airborne PM from Duisburg, Düsseldorf, and Borken, collected by GFFs using a low-volume sampler. DCM extraction	BEAS-2B human lung bronchial epithelial cells, 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/assay) increase in SCE frequency per metaphase for samples from all locations. Highest responses for fine fraction ($PM_{2.5}$) relative to coarse (PM_{10}). Responses for industrial sites exceeded that of less-polluted area (Borken). Significant induction of SCE in response to < 0.5 m^3 equiv	Hornberg et al. (1998)
Rhine–Ruhr region, Germany	"City smog" for heavily industrialized site, collected on GFFs using a low-volume sampler. MeOH extraction	Cultured human lymphocytes, 72 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/mL) increase in SCE frequency per metaphase	Seemayer et al. (1984)
Rhine–Ruhr region, Germany	41 samples of "city smog" collected in 1975–1990 at highly industrialized locations, collected on GFFs. Organic extraction	A549 human alveolar adenocarcinoma cells (24 h and 120 h) and cultured human lymphocytes (72 h) exposed to extract in DMSO	SCE assay	Site 54 (Düsseldorf) shown as an example. Significant, dose-related (equiv m^3/mL) increase in SCE frequency per metaphase	Seemayer et al. (1988, 1989, 1990b), Hadnagy et al. (1989)
Rhine–Ruhr region, Germany	20 samples of "city smog" collected in 1975–1986 at highly industrialized locations, collected on GFFs. MeOH extraction	Cultured human lymphocytes, exposure to extract in DMSO	SCE assay	Düsseldorf provided as an example. Significant, dose-related (equiv m^3/mL) increase in SCE frequency per metaphase	Seemayer et al. (1987b)
Mexico City metropolitan area, Mexico	Seasonal PM_{10} samples collected on GFFs using a high-volume sampler. DCM sonication extraction	Cultured human lymphocytes, 4 h exposure to extract in DMSO, with and without Aroclor-induced rat liver S9	SCE assay	Significant, dose-related (µg of EOM/assay) increase in SCE frequency both with and without S9. Higher responses with S9 in April and August. In November, similar responses with and without S9. November sample had highest concentrations of PAHs and nitro-PAHs	Calderón-Segura et al. (2004)
L'Aquila, Italy	Airborne PM of 2.5–10 µm and 0.4–2.5 µm, collected using an 8-stage cascade impactor	Hs27 human skin fibroblasts, 24 h exposure to suspended PM	SCE assay	Significant increases in SCE frequency for all fine PM samples collected in 6 sequential months; 3 of 6 monthly coarse PM samples induced significant increases in SCE frequency	Poma et al. (2002)
West Virginia, USA (1982)	Airborne PM collected on GFFs using a high-volume sampler. Ac extraction	Cultured human lymphocytes, 48 h exposure to extract in DMSO	SCE assay	Significant, dose-related (mg of PM equiv/mL) increase in SCE frequency	Krishna et al. (1984)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Animal cells					
Sicily, Italy	Airborne PM from 2 locations in the centre of Catania, collected on GFFs using a high-volume sampler. DCM Soxhlet extraction	CHEL Chinese hamster epithelial liver cells and CHO cells, 24 h exposure to extract in DMSO	CAs (excluding gaps)	Significant, dose-related increase in frequency of CHEL cells with CAs for both sites. No significant increase for CHO cells without exogenous activation	Motta et al. (2004)
Baja California, Mexico	Atmospheric dust from the city of Mexicali	Balb 3T3 mouse embryonic fibroblast cells, 12 h exposure to dust suspension	Anaphase aberrations (lagging chromosomes, bridges)	Dose-related increase in anaphase aberrations, including multipolar anaphases, lagging chromosomes, and bridges	Alfaro Moreno et al. (1997)
Patagonia, Argentina	Airborne PM from two towns, inspirable dust collected on GFFs. DCM extraction	Primary F344 rat hepatocytes, 3 h exposure to extract in DMSO	CAs	No significant increase in CA frequency	Ares et al. (2000)
Basel, Switzerland	PM collected on GFFs in air conditioner units. Samples from several sites during and after a large industrial fire (Schweizerhalle). MeOH Soxhlet extraction	Chinese hamster V79 lung cells, 3 h treatment with and without Aroclor-induced rat liver S9	CAs with and without gaps	Significant induction of CAs in the presence of S9. Some urban PM collected 4–5 months after the fire also elicited positive responses, but highly variable. No evidence to support hypothesis that industrial fire released clastogenic material adsorbed to airborne PM	Zwanenburg (1988)
Silesia, Poland (1984–1985)	Airborne PM collected on GFFs at 8 "high-pollution" locations. BZ Soxhlet extraction. Extract fractionation on silica	Chinese hamster V79 lung cells, 5 h, 14 h, and 24 h treatments with each of 8 fractions	CAs with and without gaps	Significant increases in CA frequency for fractions containing polar aromatics (e.g. N-, S-, O-heterocyclics), monophenols, and basic N-heterocyclics. Higher responses for longer exposures. In some instances, dose-related increases (μg of EOM/mL)	Motykiewicz et al. (1988)
Silesia, Poland	Airborne PM collected on GFFs at 18 "high-pollution" locations in Katowice district, collected on GFFs using a high-volume sampler. BZ Soxhlet extraction. Extract fractionation on silica	Chinese hamster V79 lung cells, 16 h and 24 h treatments with each of 7 fractions	Aneuploidy, hyperdiploidy, and polyploidy	Significant increases in hyperdiploidy and polyploidy for crude extract and fraction containing monophenols	Motykiewicz et al. (1991)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Rhine–Ruhr region, Germany	Airborne PM from Duisburg and Düsseldorf, collected on GFFs using a high-volume sampler. DCM extraction	Chinese hamster V79 lung cells, 16 h treatment with extract in DMSO	Aneuploidy and C-metaphases	Dose-related (equiv air m^3/mL) increases in aberrant metaphases, polyploidy, and hyperdiploidy	Hadnagy & Seemayer, (1991)
L'Aquila, Italy	Airborne PM of 0.43–2.1 µm, collected using an 8-stage cascade impactor	RAW 264.7 mouse macrophages, 48 h exposure to suspended PM	Cytokinesis-block MN assay	Significant, dose-related (µg/cm^2) increases in MN frequency for 3 samples collected in sequential months	Poma et al. (2006)
Beijing, China (2001)	$PM_{2.5}$ collected at Beijing University. Organic and inorganic extractions (details not provided)	Balb/c 3T3 mouse embryonic fibroblast cells, 8 h exposure	Cytokinesis-block MN assay	Organic PM extract induced significant, dose-related increase in MN frequency. Inorganic extract failed to induce a significant response	Zhang et al. (2003)
Taiyuan, China	Size-fractionated airborne PM from urban, residential, and suburban locations. Soxhlet extraction with MeOH, DCM, and Ac	CHL Chinese hamster lung cells, 2 h exposure to combined extract in DMSO, with and without exogenous S9	SCE assay	All extracts elicited significant, dose-related increase in SCE frequency. Greater response for extracts of smaller PM fractions. Positive association with PAH concentrations	Yang et al. (1994)
Silesia, Poland (1984–1985)	Airborne PM collected on GFFs. BZ Soxhlet extraction	Chinese hamster V79 lung cells, 26 h exposure to extract in DMSO	SCE assay	Significant, dose-related (m^3 equiv) increase in SCE frequency	Motykiewicz et al. (1990)
Paris, France (1983–1985)	Airborne PM from urban site, collected on GFFs using a high-volume sampler. DCM or Ac sonication extraction	Chinese hamster V79 lung cells, 1–3 h exposure to extract in DMSO, with and without Aroclor 1254-induced rat liver S9	SCE assay	Significant, dose-related (per µg of EOM) increase in SCE frequency; higher with exogenous metabolic activation system	Courtois et al. (1988)
Duisburg, Germany	Airborne PM from industrialized Rhine–Ruhr region. Draeger Box Micron filter. CX extraction	Chinese hamster V79 lung cells, 24 h exposure to extract in DMSO	SCE assay	Significant, dose-related (m^3 equiv/mL) increase in SCE frequency per metaphase	Seemayer et al. (1987a, 1988)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Rhine–Ruhr region, Germany	Airborne PM from Duisburg, collected on GFFs using a high-volume sampler. DCM extraction	Primary tracheal epithelial cells from Syrian golden hamsters, 48 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/mL) increase in SCE frequency per metaphase for both cell types	Seemayer et al. (1994)
Rhine–Ruhr region, Germany	Airborne PM from Duisburg and Düsseldorf, collected on GFFs using a high-volume sampler. DCM extraction	Primary tracheal epithelial cells from Wistar rats, 48 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/assay) increase in SCE frequency per metaphase for samples from both locations	Hornberg & Seemayer, (1995)
Rhine–Ruhr region, Germany	Airborne PM from Duisburg and Düsseldorf, industrial and urban area with high traffic density, respectively, collected on GFFs using a high-volume sampler. DCM extraction	Primary tracheal epithelial cells from rat and Syrian golden hamster, 48 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/assay) increase in SCE frequency per metaphase for both samples in both cell types. Significant induction of SCE in response to as little as 1 m^3 equiv	Hornberg et al. (1996)
Rhine–Ruhr region, Germany	Airborne PM from Duisburg, collected on GFFs using a high-volume sampler. DCM extraction	Primary tracheal epithelial cells from rat and Syrian golden hamster, 48 h exposure to extract in DMSO	SCE assay	Significant, dose-related (equiv m^3/assay) increase in SCE frequency per metaphase for both cell types. Significant induction of SCE in response to as little as 0.5 m^3 equiv	Hornberg et al. (1997)
Rijnmond area, Netherlands	Airborne PM collected on GFFs using a high-volume sampler. Soxhlet extractions with MeOH, CX, BZ, or Ac. Liquid–liquid fractionation	CHO cells, 1 h treatment with extract in DMSO, with and without Aroclor-induced rat liver S9	SCE assay	Significant, dose-related (m^3/assay) increase in SCE frequency both with and without S9. Potency (SCE/m^3/assay) highest for aromatic fraction with S9, followed by oxygenated compound fraction with and without S9. Aerosol gradient downwind from urban/industrial area	de Raat, (1983)
Coastal area in central Finland (1985)	Airborne PM and semivolatiles, collected in Kokkola using a high-volume sampler. PM collected on filter, semivolatiles on XAD-2 resin. Ac extraction. Fractionation on silica	CHO cells, 4 h treatment with extract in DMSO, with and without Clophen A50-induced rat liver S9	SCE assay	4 of the 5 tested PM extracts induced significant increases in SCE frequency; slight increase with S9. Winter PM extracts more genotoxic relative to spring samples. XAD-2 extracts consistently more genotoxic. Consistent activity in PAH fraction with S9, as well as in most polar fraction with and without S9	Pyysalo et al. (1987)

Table 4.6 (continued)

Geographical location	Test article	Assay/exposure system	End-point(s) examined	Results	Reference
Athens, Greece	Monthly PM samples, collected on cellulose filters using a high-volume sampler. Hx sonication extraction	CHO cells, 26 h treatment with extract in DMSO	SCE assay	All tested samples induced significant, dose-related (μg of EOM/mL) increases in SCE frequency	Athanasiou et al. (1987)
Wageningen and Terschelling, Netherlands (1979–1980)	Airborne PM from 2 rural sites, collected on GFFs using a high-volume sampler. MeOH Soxhlet extraction	Chinese hamster V79 lung cells, 2 h treatment with extract in DMSO	SCE assay	Significant, dose-related (per m^3) increases in SCE frequency when winds from east (i.e. Germany). Negligible response when winds from north	Alink et al. (1983)
West Virginia, USA	Airborne PM, collected on GFFs using a high-volume sampler. Ac extraction.	Mice primary bone marrow and spleen cells, 34 h and 44 h exposures, respectively, to extract in DMSO	SCE assay	Significant, dose-related (mg of PM equiv/mL) increase in SCE frequency in both cell types	Krishna et al. (1986)

Ac, acetone; ASE, accelerated solvent extraction; BZ, benzene; CAs, chromosomal aberrations; CHO, Chinese hamster ovary; CX, cyclohexane; DCM, dichloromethane; DMSO, dimethyl sulfoxide; EOM, extractable organic matter; equiv, equivalent; GFFs, glass-fibre filters; h, hour or hours; Hx, hexane; MeOH, methanol; MN, micronuclei; PAHs, polycyclic aromatic hydrocarbons; 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; SCEs, sister chromatid exchanges; THF, tetrahydrofuran; TSP; total suspended particles.

coal combustion and automobile exhausts, all induced CAs in cultured human lymphocytes obtained from umbilical cord blood. The major CAs included chromatid gaps, chromosome gaps, chromatid breaks, chromosome breaks, and fragments. Potency was correlated with the degree of air pollution (Ding et al., 1999). Tan et al. (2002) examined water extracts of airborne TSP collected in a tunnel in Shanghai and noted significant induction of CAs, including fragments and dicentric chromosomes, in cultured human lymphocytes compared with controls. The authors reported that metals such as lead, zinc, manganese, and iron likely contribute to the observed increase in the frequency of CAs. Wei & Meng (2006a) examined water extracts of $PM_{2.5}$ samples collected from Baotou (an industrial city in Inner Mongolia) and Wuwei (an agricultural city in Gansu province) and noted dose-dependent increases in the frequencies of CAs in cultured human lymphocytes. Samples were collected during sandstorms as well as on non-storm control days, and CA frequencies were higher in Baotou compared with Wuwei, for non-storm conditions only. CAs included chromatid breaks, chromosome breaks, acentric fragments, dicentric chromosomes, and gaps (Wei & Meng, 2006b).

Animal cells

A study by Motta et al. (2004) showed significant increases in CA frequencies in Chinese hamster epithelial liver cells, which maintained metabolic competence, exposed to extracts of airborne PM from Catania, Italy. However, negative results were seen in Chinese hamster ovary cells that required exogenous metabolic activation (Motta et al., 2004). Alfaro Moreno et al. (1997) reported a dose-related increase in anaphase aberrations in murine Balb/c 3T3 cells exposed to a suspension of atmospheric dust collected in Mexico. A study by Zwanenburg (1988) reported significant induction of CAs in Chinese hamster V79 lung cells exposed to extracts of particles collected from several sites after a large industrial fire in Switzerland in the presence of S9 (Zwanenburg, 1988). Extracts of PM from some urban sites elicited positive responses 4–5 months after the fire, and the authors could not provide convincing evidence that the fire resulted in the release of clastogenic substances. A study conducted in Poland, which examined fractions of organic extracts of airborne PM from high-pollution locations, showed significant increases in CA frequencies in Chinese hamster V79 lung cells by fractions containing polar aromatics, monophenols, and basic *N*-heterocyclics (Motykiewicz et al., 1988). A study by Ares et al. (2000) failed to show significant increases in CA frequency in primary F344 rat hepatocytes treated with extracts of airborne PM from Patagonia (Ares et al., 2000). Two studies reported significant increases in aneuploidy in Chinese hamster V79 lung cells exposed to extracts of airborne PM from Poland (Motykiewicz et al., 1991) and Germany (Hadnagy & Seemayer, 1991).

Micronuclei

Human cells

Water or organic solvent extracts of airborne PM from five cities in China were tested for induction of MN in cultured human lymphocytes. In Lanzhou, DCM extracts of TSP from five sites with varying degrees of air pollution all caused dose-dependent increases in MN frequency in cultured human lymphocytes. The samples from sites with heavy traffic or close to petroleum industries showed more potent induction of MN compared with samples from moderately contaminated sites or relatively clean sites (Ding et al., 1999). In Shanghai, saline extracts of airborne PM from Taopu, an industrial region, caused a dose-dependent increase in MN in cultured human lymphocytes (Tan et al., 2004). Wei & Meng (2006a) and Wei et al. (2006) compared MN induction by organic and inorganic extracts of $PM_{2.5}$ collected during a sandstorm or in

non-storm conditions from the industrial city of Baotou (in Inner Mongolia) and the agricultural city of Wuwei (in Gansu province). The results indicated that organic and saline PM suspensions, collected during storm and non-storm conditions, showed dose-dependent increases in MN frequency in cultured human lymphocytes (Wei et al., 2006). DCM extracts of TSP and PM_{10} samples from Guangzhou also significantly increased MN frequency in cultured human lymphocytes, and extract fractionation showed significant MN induction by the aromatic hydrocarbon fraction of PM_{10} (Xu & Wang, 2008). A study by Yuan et al. (1999a) examined acid and organic solvent extracts of airborne PM of different sizes (< 1.1 μm, 1.1–2.0 μm, 2.0–3.3 μm, 3.3–7.0 μm, and > 7.0 μm) that were collected from a residential area in Taiyuan, and they observed dose-dependent increases in frequencies of MN in cultured human lymphocytes (Yuan et al., 1999a). Acid extract studies showed that the smaller the particulate size, the higher the MN frequency; the MN frequencies were also positively correlated with the concentrations of metals in the PM extracts (Yuan et al., 1999b). In addition, a study conducted in Flanders, Belgium, also reported a significant dose-related (i.e. equivalent cubic metres per millilitre) increase in MN frequency in cultured human lymphocytes exposed to organic extracts of urban air PM (Brits et al., 2004).

In addition to studies in cultured human lymphocytes, the induction of MN by suspended PM or extracts of PM was also investigated in human cell lines. Oh et al. (2011) found significant induction of MN in BEAS-2B human lung bronchial epithelial cells exposed to organic PM extracts and extract fractions from a high-traffic area in the Republic of Korea (Oh et al., 2011). Fractionation showed significant increases in MN for aliphatic, aromatic (i.e. PAHs), and slightly polar (i.e. nitro-PAHs and quinones) fractions. Significant increases in MN frequency were also induced in A549 human alveolar adenocarcinoma cells by water and organic extracts of industrial and residential particles from Mexico City (Roubicek et al., 2007), and in HS 27 human skin fibroblasts (Poma et al., 2002).

Animal cells

Significant increase in MN frequency were also observed in RAW 264.7 mouse macrophages exposed to suspensions of airborne PM from L'Aquila, Italy (Poma et al., 2006). Moreover, an organic extract of PM from Beijing, China, induced a significant dose-related increase in MN frequency in Balb/c 3T3 cells (Zhang et al., 2003).

Sister chromatid exchanges

Human cells

Twenty-five studies investigated the induction of SCEs in cultured human lymphocytes and a variety of cultured animal cells exposed to organic PM extracts, and most of the studies showed significant increases in SCE frequency. Organic PM extracts assessed in cultured human lymphocytes include samples derived from PM collected in Lexington, Kentucky, USA (Viau et al., 1982), Silesia, Poland (Motykiewicz et al., 1990), Lanzhou, China (Zhang & Li 1994; Wang & Ding 1998), West Virginia, USA (Krishna et al., 1984), and many sites in the Rhine–Ruhr region of Germany (Seemayer et al., 1984, 1987a, b, 1988, 1989, 1990a, 1990b; Hadnagy et al., 1986, 1989; Hadnagy & Seemayer, 1987). Seasonal PM samples from Mexico City induced significant dose-related increases in SCE frequency, with the highest frequencies produced by samples taken in November and the lowest by samples taken in April (Calderón-Segura et al., 2004). Significant increases in SCE frequencies were induced in HS 27 human skin fibroblasts exposed to suspensions of PM from L'Aquila, Italy (Poma et al., 2002), and in A549 human alveolar adenocarcinoma cells and human BEAS-2B cells exposed to organic extracts of PM collected from the

Rhine–Ruhr region of Germany (Seemayer et al., 1989; Hornberg et al., 1998).

Animal cells

A series of in vitro cytogenetic studies of organic extracts of PM from several locations within the Rhine–Ruhr region of Germany showed significant dose-dependent increases in SCE frequencies in primary tracheal epithelial cells from Syrian golden hamsters or Wistar rats (Seemayer et al., 1994; Hornberg & Seemayer, 1995; Hornberg et al., 1996, 1997). Significant dose-related increases in SCE frequencies were also induced in primary bone marrow and spleen cells exposed to extracts of PM from West Virginia, USA (Krishna et al., 1986). Three independent studies showed significant increases in SCE frequencies in Chinese hamster ovary cells exposed to extracts of PM from the Netherlands (de Raat, 1983), a coastal area in Finland (Pyysalo et al., 1987), and Athens, Greece (Athanasiou et al., 1987). Two independent studies showed significant increases in SCE frequency in Chinese hamster V79 cells exposed to organic extracts of PM from the Netherlands (Alink et al., 1983) and Paris, France (Courtois et al., 1988). The study in Finland noted that concentrates of SVOCs collected on XAD-2 resin consistently elicited stronger responses compared with PM extracts (Pyysalo et al., 1987).

A study by Yang et al. (1994) showed that extracts of airborne PM of various sizes (< 1.1 μm, 1.1–2.0 μm, 2.0–3.3 μm, 3.3–7.0 μm, and > 7.0 μm) collected from industrial, residential, and suburban districts in Taiyuan, China, induced significant dose-related increases in SCEs in Chinese hamster lung cells (Yang et al., 1994). The authors also observed that a greater response was produced by extracts of smaller-sized PM and that SCE induction was positively correlated with PAH concentrations.

In summary, substantial evidence consistently shows that organic extracts, water extracts, or suspensions of outdoor air PM from urban or industrial areas induce significant dose-related cytogenetic effects (CAs, aneuploidy, MN, and SCEs) in cultured human lymphocytes, human cell lines, cultured animal primary cells, or animal cell lines in vitro.

4.2.3 DNA damage and protein adducts

(a) DNA adducts

(i) Humans

Studies on DNA adducts in humans after exposure to polluted outdoor air are summarized in Table 4.7.

A systematic review (Demetriou et al., 2012) evaluated DNA adducts as one of several biomarkers with the potential to contribute an intermediate end-point in the association between air pollution and lung cancer and graded DNA adducts in leukocytes as A for evidence, A for replication, and B for bias.

In an early study of the effects of outdoor air pollution, male residents of an industrial and highly polluted city in Poland (Gliwice) were compared with men from a rural part of the country (Biała Podlaska) (Perera et al., 1992). Both summer and winter samples were analysed by enzyme-linked immunosorbent assay (ELISA) and by ^{32}P-postlabelling for PAH–DNA adducts and aromatic DNA adducts. The exposed residents of Gliwice had significantly increased PAH–DNA and aromatic adducts compared with rural residents. For the winter samples, the Gliwice values were significantly greater than the rural values by ELISA only, and the same was found for the summer samples. The Gliwice winter values were also significantly greater than the Gliwice summer values by both methods of analysis. Other comparisons that were statistically significant were by ^{32}P-postlabelling: control winter values were greater than exposed summer values, and exposed winter values were greater than control summer values (see Table 4.7).

Table 4.7 DNA adducts in humans exposed to outdoor air pollution

Location	Study populations	Method of analysis; source of DNA	Results	Reference
Poland	39 men in Gliwice (high pollution, exposed); 49 men in Biała Podlaska (low pollution, control)	^{32}P-postlabelling and immunoassay (ELISA) for PAH–DNA adducts; white blood cells	Exposed winter levels significantly greater than exposed summer levels; exposed winter levels significantly greater than control winter levels (ELISA only); exposed summer levels significantly greater than control summer levels (ELISA only)	Perera et al. (1992)
Poland	70 mothers and newborns in Cracow	Immunoassay (ELISA) for PAH–DNA adducts; maternal and umbilical white blood cells	Significant correlation between maternal and newborn adduct levels and outdoor air pollution levels close to place of residence	Whyatt et al. (1998)
Poland	70 mothers and newborns in Cracow (urban); 90 mothers and newborns in Limanowa (rural)	Immunoassay (ELISA) for PAH–DNA adducts; maternal and umbilical white blood cells	Intra-individual differences in Cracow cohort previously noted (Whyatt et al., 1998), but differences in adduct levels between urban and rural groups not significant. Rural district had lower outdoor pollution but heavier use of coal for residential heating. Among non-coal users, adduct levels in maternal Cracow samples 2-fold higher than those in maternal Limanowa samples ($P = 0.03$)	Perera et al. (1999)
Poland	319 non-smoking mothers and newborns in Cracow	HPLC/fluorescence for B[*a*]P–DNA adducts; white blood cells from maternal and cord blood	Significant interaction between prenatal exposure to PAHs and cord blood DNA adduct levels, especially for subjects with low levels of micronutrients (carotenoids, α-tocopherol, and retinol) in maternal blood compared with those with higher levels of the micronutrients	Kelvin et al. (2009)
Sweden	Taxi drivers (19), urban bus drivers (26), suburban bus drivers (23), and controls (22)	^{32}P-postlabelling; lymphocytes	Significantly higher adduct levels in taxi drivers ($P < 0.01$) and suburban bus drivers ($P < 0.001$) than in controls. Levels in urban bus drivers not significantly different	Hemminki et al. (1994)
Denmark	Bus drivers (90) and rural controls (60)	^{32}P-postlabelling; lymphocytes	Adduct levels were highest in drivers in central Copenhagen and intermediate in drivers in suburban Copenhagen and dormitory towns. Adduct levels in all 3 groups were significantly higher than in controls ($P < 0.001$)	Nielsen et al. (1996a)
Bangladesh	Dhaka City rickshaw drivers (46) and controls (48)	Immunoassay (ELISA) for PAH–DNA adducts; white blood cells	Adducts detectable in 19/46 rickshaw drivers vs 11/48 controls ($P = 0.06$). Mean adduct levels significantly higher in drivers than in controls ($P = 0.04$ overall; $P = 0.01$ for those with detectable adducts)	Rahman et al. (2003)
Czech Republic	30 women in Teplice (high pollution, exposed); 30 women in Prachatice (low pollution, control)	^{32}P-postlabelling; white blood cells	Significant correlation between individual personal exposures to PAHs and DNA adducts	Binková et al. (1996)

Table 4.7 (continued)

Location	Study populations	Method of analysis; source of DNA	Results	Reference
Czech Republic	51 women in Teplice (polluted area)	^{32}P-postlabelling and IHC-ACIS for PAH–DNA adducts; placenta	Bulky DNA adducts detected by ^{32}P-postlabelling in fresh frozen tissue (n = 37), with no differences between smokers, non-smokers, and ETS-exposed non-smokers. Adducts detected by IHC-ACIS in fixed (n = 14) placenta samples but not in frozen (n = 37) placenta (before fixation) samples	Pratt et al. (2011)
Czech Republic	Male police officers working outdoors in downtown Prague (n = 109)	^{32}P-postlabelling; lymphocytes	Correlation between adduct levels and levels of PAH exposure at different sampling times	Topinka et al. (2007)
Czech Republic	Male police officers (exposed; n = 53) and residents (unexposed; n = 52) in Prague	^{32}P-postlabelling; lymphocytes	No significant difference in adduct levels between exposed and control groups, but the level of a B[*a*]P-like adduct was significantly higher in the exposed group (P < 0.01)	Binková et al. (2007)
Czech Republic	Residents of Ostrava (polluted industrial region; n = 149) and Prague (relatively unpolluted city; n = 65)	^{32}P-postlabelling; lymphocytes	Levels of B[*a*]P-like adducts significantly higher in Prague than in Ostrava region, but B[*a*]P concentrations higher in Ostrava than in Prague. Levels of B[*a*]P-like adducts in Ostrava region positively affected by exposure to B[*a*]P (not found for Prague). Levels of bulky adducts negatively associated with B[*a*]P/pollution levels in both cohorts	Rossner et al. (2013a)
Czech Republic, Slovakia, and Bulgaria	Residents of Prague, Košice, and Sofia, including city police officers and bus drivers (exposed; n = 204) and controls (n = 152)	^{32}P-postlabelling; lymphocytes	Negative correlation between 8-oxodG levels and B[*a*]P-like DNA adducts (P = 0.002) and between 8-oxodG levels and bulky adducts (P = 0.04)	Singh et al. (2007a)
Denmark, Greece	Non-smoking men in Athens (n = 17) and in rural (n = 29) and urban (n = 73) areas of Denmark	^{32}P-postlabelling; white blood cells (Athens) or lymphocytes (Denmark)	Median adduct levels significantly different in the 3 groups; Athens > urban Denmark > rural Denmark	Nielsen et al. (1996b)
Denmark	50 students living in Copenhagen	^{32}P-postlabelling; lymphocytes	No significant association between DNA adduct levels and exposure markers ($PM_{2.5}$ and black smoke) measured by personal exposure monitors	Sørensen et al. (2003a)
Denmark	75 pregnant women living in Greater Copenhagen	^{32}P-postlabelling; maternal blood and cord blood cells	Adduct levels in maternal and cord blood were similar and positively correlated. Adduct levels significantly elevated in mother–newborn pairs living in medium traffic-density areas (P < 0.01) but not in high traffic-density areas	Pedersen et al. (2009)
Italy	Traffic police (n = 94) and urban residents (n = 52)	^{32}P-postlabelling; white blood cells	DNA adduct levels in police significantly higher than in controls, correlating with increased exposure to PAHs	Merlo et al. (1997)
Italy	Non-smoking police officers in Genoa (exposed; n = 34) and office workers (control)	^{32}P-postlabelling; white blood cells	Median DNA adduct level of exposed group significantly higher than median of controls	Peluso et al. (1998)

Table 4.7 (continued)

Location	Study populations	Method of analysis; source of DNA	Results	Reference
Italy	114 workers in Florence exposed to traffic pollution; 100 resident controls	^{32}P-postlabelling; white blood cells	DNA adduct levels significantly higher for traffic workers. Urban residents tended to have higher level than suburban residents (not significant), with higher levels in summer than in winter	Palli et al. (2001)
Italy	320 residents of Florence (114 traffic-exposed workers; 206 randomly sampled volunteers)	^{32}P-postlabelling; white blood cells	Significant correlation between DNA adduct levels and O_3 cumulative exposure	Palli et al. (2004)
Italy	Traffic-exposed workers (n = 62) and urban residents (n = 152) in Florence	^{32}P-postlabelling; white blood cells	DNA adduct levels in traffic-exposed workers correlated with average levels of exposure to PM_{10} over a prior time period of 1–2 weeks; levels in residents did not	Palli et al. (2008)
Italy	Residents of Pisa (urban; n = 520) and Cascina (suburban; n = 825)	ELISA for antibodies to BPDE–DNA adducts in serum	26.0% of urban subjects were positive for antibodies, compared with 17.9% of suburban residents; excess prevalence of antibody positivity for urban residents (OR, 1.49; 95% CI, 1.16–1.92)	Petruzzelli et al. (1998)
Italy	194 police officers in Rome (134 in traffic control; 60 in administrative division)	ELISA for antibodies to BPDE–DNA adducts in serum	10/134 traffic police were positive for antibodies; 1/60 office workers were positive. Difference is not significant (P = 0.095, χ^2 test)	Galati et al. (2001)
Italy	Newspaper vendors in high-traffic (n = 31) and low-traffic (n = 22) areas of Milan	^{32}P-postlabelling; lymphocytes	No difference in adduct levels between the high- and low-exposure groups	Yang et al. (1996)
Thailand	Male children aged 9–13 yr in Bangkok (high exposure; n = 107) and Chonburi (low exposure; n = 69)	^{32}P-postlabelling; lymphocytes	Mean DNA adduct level 5-fold higher in Bangkok children than in Chonburi children (P < 0.001)	Ruchirawat et al. (2007), Tuntawiroon et al. (2007)
Thailand	Police officers in Bangkok; traffic police (high exposure; n = 44) and office-based police (low exposure; n = 45)	^{32}P-postlabelling; lymphocytes	Mean DNA adduct levels significantly higher in high-exposure group than in low-exposure group (P = 0.029)	Ruchirawat et al. (2002)
Thailand	Adults living near Map Ta Phut Industrial Estate (exposed; n = 72) and residents of a control district (unexposed; n = 50)	^{32}P-postlabelling; white blood cells	Adduct levels in exposed residents, 0.85 ± 0.07 (SE) DNA adducts/10^8 nucleotides, significantly higher than in controls, 0.53 ± 0.05 (SE) (P < 0.05)	Peluso et al. (2008)
Thailand	Adults living near Map Ta Phut Industrial Estate (exposed; n = 65) and residents of a control district (unexposed; n = 45)	^{32}P-postlabelling; white blood cells	Higher levels in exposed group than in unexposed, as previously reported (Peluso et al., 2008). Increased levels of DNA adducts were correlated with marginally lower LINE-1 methylation (P = 0.06) and lower *p53* methylation (P = 0.01)	Peluso et al. (2012)

Table 4.7 (continued)

Location	Study populations	Method of analysis; source of DNA	Results	Reference
Ukraine	62 pregnant women in Kiev and Zaporizhia, Ukraine (exposed); 20 pregnant women from rural Eastern Carpathian area, Poland (unexposed)	CIA for PAH–DNA adducts; placenta	38/62 samples from exposed groups had detectable DNA adducts vs 10/20 samples from controls. Newborns with the most compromised health status had highest levels of DNA adducts	Obolenskaya et al. (2010)
Mexico	92 residents of the Mexico City metropolitan area	CIA for PAH–DNA adducts; white blood cells	Mean adduct level was significantly higher in the dry season than in the rainy season, correlating with higher airborne concentrations of PM_{10}, and $PM_{2.5}$ in the dry season	García-Suástegui et al. (2011)
Benin	Residents of Cotonou (urban, high exposure; $n = 57$), Godomey (suburban; $n = 20$), and Sohon (village; $n = 17$)	^{32}P-postlabelling; lymphocytes	DNA adduct levels in urban residents were significantly higher than those in suburban and village residents ($P < 0.001$)	Ayi-Fanou et al. (2011)
China	150 children born to non-smoking mothers in Tongliang (exposed to local coal-fired power plant)	HPLC/fluorescence for B[*a*]P–DNA adducts; white blood cells from maternal and cord blood	High cord blood adduct levels (> median) associated with decreased head circumference at birth ($P = 0.057$) and decreased weight at age 18, 24, and 30 months ($P < 0.05$). Maternal blood DNA adduct levels not associated with cord blood levels or birth outcomes	Tang et al. (2006)
China	110 non-smoking mother–infant pairs in Tongliang (exposed to local coal-fired power plant)	HPLC/fluorescence for B[*a*]P–DNA adducts; white blood cells from maternal and cord blood	Increased adduct levels in cord blood associated with DQs at age 2 yr: decreased motor area DQ, language area DQ, and average DQ	Tang et al. (2008)
China	Non-smoking mother–infant pairs in Tongliang. One cohort from 2002 ($n = 110$), before the shutdown of the local coal-fired power plant; second cohort from 2005 ($n = 107$), after shutdown of plant	HPLC/fluorescence for B[*a*]P–DNA adducts; white blood cells from maternal and cord blood	Associations between elevated adduct levels and decreased DQs at age 2 yr seen in the 2002 cohort were not observed in the 2005 cohort	Perera et al. (2008)

B[*a*]P, benzo[*a*]pyrene; BPDE, benzo[*a*]pyrene diol epoxide; CI, confidence interval; CIA, chemiluminescence immunoassay; DQ, development quotient; ETS, environmental tobacco smoke; HPLC, high-performance liquid chromatography; IHC-ACIS, immunohistochemistry with automated cellular imaging system; LINE-1, long interspersed nuclear element-1; 8-oxodG, 8-oxo-7,8-dihydro-2′-deoxyguanosine; O_3, ozone; OR, odds ratio; PM_{10}, particulate matter with particles of aerodynamic diameter < 10 µm; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 µm; SE, standard error; yr, year or years.

Other studies in Poland have focused on mother–newborn pairs. Analysis by immunoassay (ELISA) for PAH–DNA adducts of maternal and cord white blood cells of mothers and newborns from Cracow found significant correlations between adduct levels and outdoor air pollution levels (PM_{10}) close to their places of residence (Whyatt et al., 1998). In a subsequent study, the Cracow cohort was compared with a rural cohort from Limanowa, and differences in adduct levels between the urban and rural groups were not significant (Perera et al., 1999); however, it was noted that there was heavier use of coal for home heating in the rural district than in the city. Among non-coal users only, adduct levels in Cracow maternal samples were significantly higher than those in Limanowa maternal samples.

In a subsequent study of a larger group of Cracow women, PAH exposure was estimated from personal air monitors worn during pregnancy (Kelvin et al., 2009). There was a significant interaction between prenatal exposure to PAHs and the levels of cord blood B[*a*]P–DNA adducts, determined by high-performance liquid chromatography (HPLC)/fluorescence analysis. This association was stronger in babies with low blood levels of α-tocopherol and carotenoids.

An early study measured DNA adducts by ^{32}P-postlabelling in the lymphocytes of taxi drivers, urban bus drivers, suburban bus drivers, and controls (hospital workshop workers) in Stockholm, Sweden (Hemminki et al., 1994). The adduct levels in the taxi drivers and the suburban bus drivers were significantly higher than those in the controls ($P < 0.01$ and $P < 0.001$, respectively), but the adduct levels in urban bus drivers were not significantly different from those in the controls.

Another study investigated DNA adducts in bus drivers in Copenhagen, Denmark (Nielsen et al., 1996a). Significantly higher DNA adduct levels were found in the drivers in central Copenhagen compared with those driving in outer areas, and all driver groups had significantly higher levels than controls consisting of rural dwellers or the general population.

A study on rickshaw drivers in Dhaka City, Bangladesh, used ELISA to detect PAH–DNA adducts in white blood cells (Rahman et al., 2003). A higher proportion of the drivers, who were not shielded or protected from exposure to traffic pollution, had detectable DNA adducts than a control group of unexposed people (19/46 vs 11/48; $P = 0.06$), and the mean adduct level was significantly higher in the rickshaw drivers than in the controls (overall, $P = 0.04$; for those with detectable adducts, $P = 0.01$).

Studies in the Czech Republic have focused on comparisons of residents of a highly industrialized and polluted region, Northern Bohemia, with those of a relatively unpolluted rural part of the country. When women in Teplice (in the industrialized region) were compared with women in Prachatice (rural control), significant associations between bulky DNA adducts in white blood cells and individual levels of exposure to PAHs (measured by personal air monitors) were found (Binková et al., 1996). A study of the placentas of mothers in Teplice detected bulky DNA adducts by ^{32}P-postlabelling and PAH–DNA adducts by immunohistochemistry that were unrelated to the smoking status of the women (Pratt et al., 2011).

One study of male police officers working outdoors in the downtown area of Prague found a correlation between bulky DNA adducts in their lymphocytes and air levels of PAHs at the various sampling times (Topinka et al., 2007), but another study that compared male police officers in Prague with residents of the city did not find a difference in overall adduct levels between the two groups, although the level of a B[*a*]P-like adduct was significantly higher in the exposed group (Binková et al., 2007).

Comparisons of Prague residents with those of an area of higher pollution, the industrialized region of Ostrava, revealed that levels of B[*a*]P-like

adducts in lymphocytes were positively affected by B[*a*]P exposure levels for the Ostrava residents but not for Prague residents (Rossner et al., 2013a). Although B[*a*]P concentrations were higher in Ostrava, levels of B[*a*]P-like adducts were higher in Prague. For total bulky adducts, levels in both cohorts were negatively associated with B[*a*]P and pollution levels.

A study that monitored residents of Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria) compared several biomarkers of exposure, including DNA adducts detected by ^{32}P-postlabelling (Singh et al., 2007a). Levels of total bulky DNA adducts, and also of B[*a*]P-like adducts, negatively correlated with levels of the oxidatively generated lesion 8-oxodG in DNA.

In a study that compared non-smoking men in Athens, Greece, with men in urban and rural areas of Denmark, the median adduct levels in white blood cells (Athens) or lymphocytes (Denmark) were significantly different in the three groups; levels in Athens were higher than those in urban Denmark, and levels in urban Denmark were higher than those in rural Denmark (Nielsen et al., 1996b). However, in another study of students living in Copenhagen, Denmark, there was no significant association between levels of bulky DNA adducts in lymphocytes and two exposure markers – levels of $PM_{2.5}$ and black smoke – measured by personal exposure monitors (Sørensen et al., 2003a). Also, among mother–newborn pairs living in Copenhagen, adduct levels were significantly elevated in maternal and cord blood of those living in medium-traffic-density areas, but not of those living in high-traffic-density areas, relative to those living in low-traffic-density areas (Pedersen et al., 2009)

Several studies in Italy have shown a positive association between exposure to outdoor air pollution and DNA adduct levels. Traffic police had significantly higher levels of bulky DNA adducts in white blood cells than age-matched urban residents (Merlo et al., 1997). Police officers in Genoa had a significantly higher median DNA adduct level than office workers (Peluso et al., 1998). Traffic-exposed workers in Florence had significantly higher DNA adduct levels than urban residents (Palli et al., 2001), with a significant correlation between adduct levels and ozone concentrations (cumulative exposure) (Palli et al., 2004). DNA adduct levels in white blood cells in traffic-exposed workers in Florence correlated with average levels of exposure to PM_{10} (Palli et al., 2008). When urban (Pisa) and suburban (Cascina) residents were compared for antibodies to B[*a*]P diol epoxide (BPDE)–DNA adducts in serum, there was a significant excess prevalence of antibody positivity among the urban residents (Petruzzelli et al., 1998). However, the same measurement carried out among police officers in Rome found only a non-significant increase (P = 0.095) among traffic police (10/134 positive for antibodies) compared with those with office duties (1/60 positive) (Galati et al., 2001). An earlier study of newspaper vendors, in whom bulky DNA adducts were measured in lymphocytes, did not find a difference between those working in high-traffic areas and those working in low-traffic areas of Milan (Yang et al., 1996).

In Thailand, schoolchildren in Bangkok were found to be exposed to levels of airborne PAHs 3.5-fold higher than those in a rural area (Ruchirawat et al., 2007, Tuntawiroon et al., 2007); in the same study, mean levels of bulky DNA adducts in blood lymphocytes in the Bangkok schoolchildren were 5 times those in the rural schoolchildren. Bangkok traffic police had significantly higher levels of bulky DNA adducts than office-based police (Ruchirawat et al., 2002). Another Thai study, of residents near an industrial estate, found higher bulky adduct levels in white blood cells (Peluso et al., 2008) and also lower methylation of the *p53* gene, an epigenetic effect, associated with increased levels of DNA adducts (Peluso et al., 2012).

A study of placental DNA samples from Ukraine (exposed group) compared them with

samples from a rural area of Poland (control group) for DNA adducts measured by immunoassay (Obolenskaya et al., 2010). A higher proportion of the Ukrainian group had detectable levels of PAH–DNA adducts compared with the Polish group, and those newborns with the most compromised health status also had the highest adduct levels. Among residents of Mexico City, whose white blood cell DNA was monitored by the same immunoassay technique as for the Ukrainian and Polish placental samples, it was found that the seasonal variation in the mean adduct level correlated with airborne concentrations of PM_{10} and $PM_{2.5}$; all parameters were higher in the dry season than in the rainy season (García-Suástegui et al., 2011).

A single study in Africa found similar results to those of the studies in Europe and Asia; in Benin, levels of bulky DNA adducts in lymphocytes were significantly higher among urban residents than among people living in suburban or village environments (Ayi-Fanou et al., 2011).

Studies of non-smoking mother–newborn pairs in Tongliang, China, a city whose principal source of air pollution was a coal-fired power plant, have also measured the effect on birth outcomes and subsequent child development. When measured by HPLC/fluorescence, B[*a*]P–DNA adduct levels in cord blood, but not in maternal blood, were associated with decreased weight at up to 30 months and decreased head circumference at birth (Tang et al., 2006). Increased adduct levels in cord blood were also associated with several physical and cognitive scores measured at age 2 years (Tang et al., 2008). The subsequent closure of the power plant yielded the opportunity to make comparisons between infants born before the closure and those born after the closure. The associations between elevated adduct levels in cord blood and deficiencies in development seen in the earlier cohort were not seen in the post-closure cohort of infants, suggesting the benefit of reduced exposure to air pollution of children prenatally and/or postnatally (Perera et al., 2008).

Collectively, the majority of these studies demonstrate the presence of elevated levels of DNA adducts in adults occupationally exposed to outdoor air pollution, relative to comparable groups in environments with lower levels of pollution. Seasonal differences were also observed. Studies in children and newborns have found similar differences.

(ii) Experimental studies – in vivo systems

See Table 4.8.

When cyclohexane extracts of air particles from rural and urban areas of Sicily, Italy, were instilled intratracheally in rats for 5 consecutive days, they were found to lead to the formation of lung DNA adducts, detected by ^{32}P-postlabelling and synchronous fluorescence spectroscopy (Izzotti et al., 1996). Higher levels of adducts were found in the animals treated with the urban extract than in those treated with the rural sample.

Feral pigeons were caught at four different locations in the Netherlands and analysed for their DNA adduct levels in kidney, lung, and liver (Schilderman et al., 1997). Although the levels of PAHs in the particulate samples reflected the density of traffic at each location, no differences were found in the tissue adduct levels (measured by ^{32}P-postlabelling) in pigeons from the different sites.

Extracts of airborne particles from Shanghai, China, when tested on mouse skin, gave rise to DNA adducts, detected by ^{32}P-postlabelling, in the skin, liver, and kidney, but not in the lung, of the animals (Zhao et al., 2003). Most of the genotoxic activity of the fractions was attributed to the PAH component of the extracts.

When mice were exposed to diesel exhaust particles by inhalation, increased levels of bulky DNA adducts (detected by ^{32}P-postlabelling) were formed in their lungs (Dybdahl et al., 2004). In another study, oral exposure of pregnant mice

Table 4.8 DNA adducts in animals in vivo exposed to outdoor air pollution or extracts of air particles

Material or exposure	Species; route of exposure	Method of analysis	Results	Reference
Extracts of diesel exhaust	Mice; applied topically to skin	^{32}P-postlabelling	DNA adducts detected in lung > skin > liver	Gallagher et al. (1990)
Extracts of air particles from rural and urban sites in Sicily, Italy	Rats; intratracheal instillation	^{32}P-postlabelling; synchronous fluorescence spectroscopy	DNA adducts detected in lung. Urban > rural	Izzotti et al. (1996)
Feral pigeons caught at 4 locations in the Netherlands	Pigeons; environmental exposure	^{32}P-postlabelling	DNA adducts detected in kidney, liver, and lung. No association with PAH levels at each city site	Schilderman et al. (1997)
Extracts of airborne particles from Shanghai, China	Mice; applied topically to skin	^{32}P-postlabelling	DNA adducts detected in skin, liver, and kidney; not detected in lung	Zhao et al. (2003)
Diesel exhaust particles	Mice; inhalation	^{32}P-postlabelling	DNA adducts detected in lung	Dybdahl et al. (2004)
Diesel exhaust particles	Pregnant mice; oral exposure	^{32}P-postlabelling	DNA adducts detected in embryos	Reliene et al. (2005)
Vicinity of steel mills and major highway	Mice; environmental exposure	^{32}P-postlabelling	DNA adducts detected in lung (somatic tissue) but not in testis (germline tissue)	Yauk et al. (2008)

PAHs, polycyclic aromatic hydrocarbons.

to diesel exhaust particles resulted in detectable levels of bulky DNA adducts in the embryos (Reliene et al., 2005).

In a study investigating the germline mutagenicity of outdoor air pollution, a group of mice was exposed in situ in the vicinity of steel mills and a major highway (Yauk et al., 2008). When mouse samples were analysed by ^{32}P-postlabelling, bulky DNA adducts were detected in lung tissue but were below the limit of detection in testis tissue.

(iii) Experimental studies – in vitro systems

See Supplemental Table S11 (available online).

Human cells

Air samples from Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria) were compared in metabolically competent human hepatoma HepG2 cells, human diploid lung fibroblasts (HEL), and the human monocytic leukaemia cell line THP-1 (Sevastyanova et al., 2007). DNA adduct formation was highest in HepG2 cells, followed by HEL and then THP-1 cells. Winter samples were more active than summer samples; for the winter samples, the activity was highest for Prague, followed by Sofia and then Košice, but for the summer samples, the order was reversed, with the highest activity for Košice, followed by Sofia and then Prague. However, when the activities were related to the extractable content per cubic metre of air, the Sofia samples had the highest genotoxic activity regardless of the sampling period.

Animal cells

SRM 1649a was extracted with DCM and fractionated before testing for DNA adduct-forming activity in rat liver epithelial WB-F344 cells (Andrysík et al., 2011). When analysed by ^{32}P-postlabelling and HPLC, the crude extract formed only one major adduct peak, which corresponded with the (+)-*anti*-BPDE–dG

adduct. When analysed by ^{32}P-postlabelling and thin-layer chromatography, the crude extract and the non-polar fraction (containing PAHs, methylated PAHs, polychlorinated biphenyls, and polychlorinated dibenzodioxins/furans) gave rise to detectable adducts, but the polar fraction (containing oxygenated derivatives of PAHs) did not.

Air samples from the Czech Republic have also been tested in cellular assays for DNA adducts. Crude and fractionated DCM extracts of air samples from Teplice (industrialized area) and Prachatice (rural area) were incubated with cultured rat hepatocytes and Chinese hamster V79NH lung cells expressing nitroreductase activity (Topinka et al., 2000). In hepatocytes, the highest DNA adduct-forming activity was found in the fractions containing most of the PAHs and nitro-PAHs, and in V79NH cells, the highest levels were caused by the fraction containing only nitro-PAHs. Winter samples had 3–4-fold higher binding potential than summer samples.

Acellular systems

Several studies have investigated the effect of extracts of air samples from areas of the Czech Republic with different levels of air pollution on calf thymus DNA in the presence of S9. The extent of DNA adduct formation, detected by ^{32}P-postlabelling, was determined.

Overall, these studies demonstrated the potential of organic extracts of air sample particulates to form DNA adducts when incubated with DNA in the presence of an acellular metabolizing system, and when incubated with mammalian cells. Samples collected in the winter were generally more active than samples collected in the summer.

(b) Protein adducts

Several studies (see Supplemental Table S12, available online) have investigated the value of protein adducts in monitoring human exposure to environmental carcinogens, by comparing residents of cities with those living in rural environments, or by comparing occupations resulting in exposure to outdoor air pollution, such as bus and taxi drivers, with occupations with bystander exposure to air pollution, such as traffic police or street newspaper vendors, and with workers with indoor occupations. Most of these studies have used B[*a*]P as the standard pollutant and have measured adducts of its activated form, BPDE, with either haemoglobin in red blood cells or albumin in blood serum, mainly by ELISA or HPLC and GC-MS. The potential for such adducts to result from tobacco smoking or diet has generally been recognized, and most studies have attempted to control for these exposures.

In a study that also measured DNA adducts (see Section 4.2.3a(i)), Hemminki et al. (1994) measured PAH–plasma protein adduct levels in taxi drivers (n = 19), urban bus drivers (n = 26), suburban bus drivers (n = 21), and controls (n = 21) in Stockholm, Sweden. The levels were significantly elevated, relative to controls, in the taxi drivers ($P < 0.001$) but not in either group of bus drivers.

Studies comparing residents of industrialized, polluted regions of countries with residents of rural, unpolluted regions of the same countries have shown mixed results. Such a study in Denmark found that the rural residents (n = 29) had non-significantly higher levels of albumin adducts compared with urban residents (n = 73) (Nielsen et al., 1996b). A study of mothers and newborns in Denmark found that among non-smoking women resident in a rural area, adduct levels were significantly lower in a suburban group (n = 37) than in city dwellers (n = 40), but levels in rural dwellers were not significantly different from those in city dwellers (Autrup & Vestergaard, 1996). Levels of albumin adducts in cord blood were lower than those in maternal blood, and adduct levels in maternal blood were slightly higher in smokers and rural

residents than in non-smokers and suburban and city dwellers.

A study in the Czech Republic found no significant difference in serum albumin adduct levels between women in a polluted region (Teplice, n = 30) and those in a rural region (Prachatice, n = 30) (Binková et al., 1996). A study in Poland found that plasma albumin adduct levels in rural controls (n = 45) were significantly lower than those in exposed residents (n = 36) (summer samples) but were not correlated with air levels of B[*a*]P (stationary sampling) (Kure et al., 1997).

In a study of residents of Munich, Germany, that also considered diet and smoking as possible sources of B[*a*]P–protein adducts, adduct levels did not correlate with estimated dietary intake of B[*a*]P. Levels of albumin and haemoglobin adducts of B[*a*]P tended to be higher in suburban residents than in city dwellers; this was of borderline significance for B[*a*]P–albumin (Scherer et al., 2000).

A study in Germany analysed aromatic amine–haemoglobin adducts in children aged 7 years and found the highest levels of several aromatic amine adducts in children from Munich (population, 1.3 million); children from Augsburg (population, 250 000) had intermediate levels, and children from Eichstätt (population 13 000) had the lowest levels (Richter et al., 2001).

A study of traffic police (n = 44) in Bangkok, Thailand, found that they had significantly higher levels of BPDE–serum albumin adducts than police working in offices (n = 45) (Ruchirawat et al., 2002). A study of street newspaper vendors in Milan, Italy, found that those working at sites with high traffic flow (n = 30) had significantly higher levels of BPDE–haemoglobin adducts than those working at low-traffic sites (n = 23) (Pastorelli et al., 1996).

Thus, in most, but not all, of these studies of protein adducts, levels in urban dwellers were higher than those in suburban and rural dwellers, with elevated levels found in workers occupationally exposed to traffic pollution, similar to findings in studies that measured DNA adducts (see Section 4.2.3a).

(c) DNA strand breaks

(i) Humans – in vivo studies

Associations between air pollution and biomarkers of oxidative stress, including DNA strand breaks, have been assessed in a variety of biomonitoring studies, including controlled exposure, panel, and cross-sectional studies. The controlled exposure studies have typically been better than panel and cross-sectional studies because of better control for possible confounders. Table 4.9 provides an overview of studies that have assessed the association between air pollution exposure and DNA strand breaks in cells from humans.

In Belgium, a cross-sectional study showed that subjects in locations with heavy industry had increased levels of DNA strand breaks in leukocytes compared with subjects in low-pollution areas (Staessen et al., 2001). The levels of DNA strand breaks in leukocytes correlated with ozone levels as well as urinary excretion of 1-OHP and benzene metabolites (*trans*,*trans*-muconic acid [*t*,*t*-MA] and *o*-cresol) in univariate models (Koppen et al., 2007; Staessen et al., 2001). A later cross-sectional study in Belgium of subjects in areas with different types of air pollution showed that the highest levels of DNA strand breaks in leukocytes were observed in subjects living closest to air pollution sites, whereas there was no correlation between exposure markers (*t*,*t*-MA and 1-OHP) and levels of DNA strand breaks (De Coster et al., 2008; Ketelslegers et al., 2008).

A study in Benin investigated the association between four groups of exposed subjects, encompassing taxi-moto drivers in the city of Cotonou, subjects living near roads with heavy traffic or in the suburbs, and village controls. Exposure to air pollution was determined by urinary excretion of benzene metabolites and the number

Table 4.9 DNA strand breaks in blood cells from humans exposed to outdoor air pollution

Exposure	Exposure assessment	Reference
Subjects living in a rural village and 2 suburbs of Antwerp, Belgium	O_3: 15–58 μg/m³ 1-OHP (urine) *t,t*-MA (urine) *o*-Cresol (urine)	Koppen et al. (2007); Staessen et al. (2001)
Subjects living in Flanders, Belgium	1-OHP (urine) *t,t*-MA (urine)	De Coster et al. (2008); Ketelslegers et al. (2008)
Taxi-moto drivers, people living or working near busy roads, and rural controls in Benin	Outdoor (stationary) sampling of UFP (midday 1 h concentration at a busy street intersection and a town square in a rural village, 265 145 and 6961 UFP/cm³, respectively) and urinary excretion of *S*-PMA	Avogbe et al. (2005)
People living near or working at an oil refinery plant and controls from another location in Brazil	PM_{10}: 9–62 μg/m³ (in the location of the oil refinery plant)	Coronas et al. (2009)
Subjects living in towns with or without industrial areas in Brazil	TSP: higher in the towns with industry (84–154 μg/m³) than in non-industrial towns (28–104 μg/m³)	Pereira et al. (2013)
Male police officers working in traffic or indoors in Shanghai, China	Level of exposure was obtained by personal monitoring of $PM_{2.5}$ in traffic police (115.4 ± 46.2 μg/m³) and officers working indoors (74.9 ± 40.1 μg/m³)	Li et al. (2010)
Traffic police working in traffic or indoors in 8 districts in Guangzhou, China	NR	Zhu et al. (2003)
Mothers and newborn children in Teplice and Prachatice, Czech Republic	Air pollution levels were not specified, but levels of PM were typically higher in Teplice than in Prachatice	Srám et al. (1998)
Panel study of subjects in Teplice, Czech Republic	Personal PAH concentration in $PM_{2.5}$ (6.2–10 ng/m³)	Binková et al. (1996)
Police officers and controls in Prague, Czech Republic	Outdoor and personal PAH concentration (6.5 and 12.4 ng/m³ in February; 3.7 and 16.7 ng/m³ in June)	Cebulska-Wasilewska et al. (2005)
Police officers and a control group of subjects who were matched for age, sex, and length of employment in Prague, Czech Republic	$PM_{2.5}$ (stationary monitoring data: 33 ± 40 μg/m³ and 15 ± 9 μg/m³) PAHs (personal exposure: 8.5 ± 9 ng/m³ and 3.0 ± 3.4 ng/m³)	Novotna et al. (2007)
Bus drivers, garage workers, and controls in Prague, Czech Republic	Personal PAH concentration (3.9–5.7 ng/m³)	Bagryantseva et al. (2010)
Subjects living in Copenhagen, Denmark	Benzene (personal exposure and urinary excretion of *S*-PMA)	Sørensen et al. (2003c)
Panel study of students living in Copenhagen, Denmark	Personal $PM_{2.5}$: 16.1 (10–24.5) μg/m³ $PM_{2.5}$: 9.2 (5.3–14.8) μg/m³ (stationary monitoring stations)	Sørensen et al. (2005)
Subjects exposed to outdoor air while bicycling in Copenhagen, Denmark	Personal UFP: 32 400 and 13 400 UFP/cm³ PM_{10}: 23.5 μg/m³ (street) and 16.9 μg/m³ (background) NO_2: 32.1 and 24.2 μg/m³ (street) and 11.3 μg/m³ (background)	Vinzents et al. (2005)

concentration of ultrafine particles (UFP) at specific sites in Cotonou or in the village (midday 1-hour average, 6961–265 145 UFP/cm³). In addition, the personal exposure level of benzene was assessed as *S*-phenyl mercapturic acid (*S*-PMA) excretion in urine. The authors showed

Table 4.9 (continued)

Exposure	Exposure assessment	Reference
Controlled exposure to outdoor air in a chamber for 24 h in Copenhagen, Denmark	Personal UFP: 6169–15 362 UFP/cm³ (unfiltered air) and 91–542 UFP/cm³ (filtered air) NO_x: 25.3 ppb (unfiltered air), 28.3 ppb (filtered air), 11.6 ppb (background), and 59.5 ppb (busy street) O_3: 12.1 ppb (unfiltered air), 4.3 ppb (filtered air), 30.1 ppb (background), and 19.5 ppb (busy street)	Bräuner et al. (2007)
Subjects living within Athens (urban) or outside Athens (rural), Greece	None	Piperakis et al. (2000)
Subjects living in Florence (polluted area) and Sassari (non-polluted area), Italy	PM_{10}: 31–67 µg/m³ NO_x: 17–100 µg/m³ SO_2: 2.9–6.5 µg/m³ O_3: 17–75 µg/m³	Pacini et al. (2003)
Subjects in Florence, Italy	O_3 (stationary monitoring data): 15–75 µg/m³	Giovannelli et al. (2006)
Traffic police and controls in Rome, Italy	Benzene: 3.8–9.5 µg/m³	Carere et al. (2002)
Children and adults living in a low-pollution area and in Mexico City, Mexico	O_3: 269 ppb (average maximum) NO_2: usually < 53 ppb SO_2: usually < 30 ppb	Calderón-Garcidueñas et al. (1996, 1997)
Panel study of subjects in Mexico City, Mexico	NR	Fortoul et al. (2010)
Students in Mexico City, Mexico	O_3: 115–172 ppb	Rojas et al. (2000)
Children living in a low-pollution area and in Mexico City, Mexico	PM_{10}: < 14 to 53–61 µg/m³ O_3: < 10 to 261 ppb (max)	Calderón-Garcidueñas et al. (1999)
Police officers working in traffic or indoors in Bangkok, Thailand	Personal benzene (8–50 µg/m³), *t,t*-MA, *S*-PMA, and 1,3-butadiene (0.3–4.1 µg/m³) exposure	Arayasiri et al. (2010)
Children living in a rural area (Chonburi) and an urban area (Bangkok) in Thailand	Benzene (outdoor monitoring and personal exposure)	Buthbumrung et al. (2008); Ruchirawat et al. (2006, 2007)
Schoolchildren in Bangkok and a low-pollution area in Thailand	PAH: 2–26 ng/m³ 1-OHP excretion	Tuntawiroon et al. (2007)

h, hour or hours; NO_2, nitrogen oxide; NO_x, nitrogen oxides; NR, not reported; 1-OHP, 1-hydroxypyrene; O_3, ozone; PAHs, polycyclic aromatic hydrocarbons; 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; SO_2, sulfur dioxide; *S*-PMA, *S*-phenyl mercapturic acid; TSP, total suspended particles; *t,t*-MA, *trans,trans*-muconic acid; UFP, ultrafine particles.

a positive relationship between the levels of DNA strand breaks in peripheral blood mononuclear cells (PBMCs) and air pollution levels in terms of either the outdoor air concentration of UFP or the *S*-PMA concentrations in urine (Avogbe et al., 2005).

In a study in Brazil, subjects living at a location near an oil refinery plant had higher levels of DNA strand breaks in lymphocytes compared with subjects from a city that was characterized as having little traffic and industry (Coronas et al., 2009). Another study in Brazil, with the same type of study design, showed no difference in the level of DNA strand breaks between subjects from urban industrialized and non-industrialized areas (Pereira et al., 2013). It should be noted that the results might be biased because it is difficult to separate the effect of air pollution exposure from the other variables that differ between subjects from different locations.

A study of traffic police and a matched group of reference subjects (officers working mainly indoors) in Shanghai, China, assessed exposure by personal monitoring of $PM_{2.5}$ and measured levels of DNA strand breaks in lymphocytes by the comet assay. The subjects were smokers or had stopped smoking for more than 6 months (including family members) before the study. The

personal monitoring data indicated that traffic police were exposed to higher levels of $PM_{2.5}$ (115.4 ± 46.2 μg/m³) compared with officers working indoors (74.9 ± 40.1 μg/m³). Traffic police had a higher percentage of lymphocytes with a comet tail compared with officers working indoors. In addition, the levels of DNA strand breaks, assessed as the average tail moment, were reported to be higher in lymphocytes from the group of traffic police compared with the group of officers working indoors (Li et al., 2010). [The Working Group noted that the statistical analysis of the reported results was based on the total number of cells from all the subjects, 100 scored nuclei per subject times the number of subjects, giving rise to group sizes of more than 10 000 data points for approximately 100 subjects per group. This is at odds with the standard procedure of statistical analysis for the comet assay.] Another study of police officers from eight districts in Guangzhou, China, showed that traffic police had significantly higher frequencies of DNA strand breaks in lymphocytes, measured as the comet tail length (4.2 μm; 95% confidence interval [CI], 3.98–4.42 μm), compared with officers working indoors (3.23 μm; 95% CI, 2.82–3.70 μm) ($P < 0.001$, $F = 9.23$, *t*-test). Smoking was identified as a confounding factor, although the results showed that traffic exhaust exposure was the main factor for the level of DNA strand breaks in lymphocytes (Zhu et al., 2003).

Several studies in the Czech Republic have assessed DNA strand breaks in people with different occupations or living in areas characterized by high or low air pollution levels. The early studies focused on differences in air pollution exposures between the regions of Teplice (industrial site) and Prachatice (low-pollution area). The Teplice area has higher air pollution levels than the Prachatice area. For instance, the $PM_{2.5}$ levels were 122 μg/m³ in Teplice and 44 μg/m³ in Prachatice during the winter of 1993 (Srám et al., 1996). During the summer of 1993, the levels were 29 μg/m³ in Teplice and 18 μg/m³ in Prachatice (Srám et al., 1996). In the subsequent years, the levels of air pollution were higher in Teplice than in Prachatice, although the differences were less dramatic than during the winter of 1993. During the summer of 1993 and the winter of 1998, the typical PM_{10} levels were 40–60 μg/m³ in Teplice and 20–40 μg/m³ in Prachatice (Srám et al., 1999). These early studies showed that personal exposures to PAHs in respirable particles correlated with levels of DNA strand breaks in lymphocytes (Binková et al., 1996). Mothers and children from the Teplice area and the Prachatice area had the same levels of DNA strand breaks in leukocytes (Srám et al., 1998). In Prague, police officers with personal exposure to PAHs had the same level of DNA strand breaks in lymphocytes as controls, although there was a difference in exposure. The personal PAH levels for the police officers and the controls were 6.5 ng/m³ and 12.4 ng/m³, respectively, in February and 3.7 ng/m³ and 16.7 ng/m³, respectively, in June (Cebulska-Wasilewska et al., 2005). Another study of police officers showed higher levels of DNA strand breaks in lymphocytes in the season with a high level of air pollution exposure (January; $PM_{2.5}$ = 33 μg/m³), whereas there was no effect in the season with a low level of air pollution exposure (September; $PM_{2.5}$ = 15 μg/m³) (Novotna et al., 2007). A study of bus drivers, garage workers, and office workers (controls) showed increased levels of DNA strand breaks in lymphocytes of workers exposed to air pollution (Bagryantseva et al., 2010).

A panel study of students who were living in the centre of Copenhagen, Denmark, showed no association between levels of DNA strand breaks in lymphocytes and personal exposure to $PM_{2.5}$ in the range of 10–24.5 μg/m³ (Sørensen et al., 2003a, b). Another study of residents of Copenhagen used benzene as marker of urban air pollution exposure and also showed no association between urinary excretion of *S*-PMA and levels of DNA strand breaks in lymphocytes (Sørensen et al., 2003c). The effect of personal exposure to UFP in air pollution was investigated

in people bicycling for approximately 90 minutes in the laboratory or on traffic-heavy streets in Copenhagen. This study showed no association between personal exposure to UFP and levels of DNA strand breaks in PBMCs (Vinzents et al., 2005). A later study on controlled exposure to air from a busy street in Copenhagen showed a correlation between particles in the size mode with a median diameter of 57 nm (representing carbonaceous soot) and levels of DNA strand breaks in PBMCs, whereas the size mode with a median diameter of 23 nm (representing SVOCs of diesel exhaust) was not associated with elevated levels of DNA strand breaks (Bräuner et al., 2007).

Non-smoking subjects in Athens, Greece, had elevated levels of DNA strand breaks in lymphocytes compared with subjects in a rural area; there was no difference in levels of DNA strand breaks in lymphocytes between smokers in Athens and those in the rural area (Piperakis et al., 2000).

A study in Florence, Italy, showed a positive association between urban ozone concentrations (~75 μg/m^3 in June and ~17 μg/m^3 in January) and levels of DNA strand breaks in nasal epithelial cells, and the residents of Florence had higher levels of DNA strand breaks and ozone exposure compared with people living in a city with a low air pollution level (45 μg/m^3 in June) in Sardinia (Pacini et al., 2003). Another study of subjects in Florence showed a positive association between ozone concentrations and levels of DNA strand breaks in lymphocytes (Giovannelli et al., 2006). Police officers from Rome, Italy, had unaltered levels of DNA strand breaks in leukocytes compared with a control group of office workers, despite a large difference in benzene exposure (9.5 μg/m^3 vs 3.8 μg/m^3 as measured by personal air sampling during a work shift) between the groups (Carere et al., 2002).

A study among subjects in Mexico City showed an association between levels of ozone and numbers of nasal epithelial cells with DNA strand breaks among adults from different locations in the city and in a low-pollution Pacific coastal town (Calderón-Garcidueñas et al., 1996). Embedded in the same study was also an assessment of the effect in young adults who moved to Mexico City from low-pollution small towns; the number of nasal cells with DNA strand breaks in this group of subjects increased during the first 2 weeks after arrival (Calderón-Garcidueñas et al., 1996). The same group of authors also showed that children in Mexico City had more nasal cells with DNA strand breaks compared with children in a low-pollution Pacific coastal town (Calderón-Garcidueñas et al., 1996, 1997). A seasonal variation was observed; samples of nasal epithelial cells that were collected during the autumn (with high air pollution levels) from a population chronically exposed to this atmospheric pollution had higher levels of DNA strand breaks compared with samples collected during the summer (with low air pollution levels) (Fortoul et al., 2010). A study of students in Mexico City showed that subjects living at a location with high outdoor air concentrations of ozone had elevated levels of DNA strand breaks in exfoliated tear duct cells compared with subjects from a location with lower ozone concentrations (Rojas et al., 2000). [This study had some limitations as judged by the standards that are used for comet assay analysis today. These include that the samples from exposed subjects and controls might have been collected and analysed at different times, without control for period effects, and that the results were reported as percentages of cells with DNA strand breaks rather than as numbers of lesions in the cells.] A later study by the same group investigated genotoxicity in nasal biopsies from children living in areas with different levels of exposure (a Pacific coastal town vs Mexico City) and showed positive associations between ozone exposure and elevated levels of DNA strand breaks in nasal cells (Calderón-Garcidueñas et al., 1999).

Table 4.10 DNA strand breaks in lungs of animals in vivo

Particles	Animal	Extraction	Dose and duration	Effect	Reference
Endotracheal instillation of $PM_{2.5}$ from an unspecified location in China	Wistar rats	Water (sonication)	1.5 or 7.5 mg/kg bw	Increased levels of DNA strand breaks (comet) in lung tissue	Lin et al. (2009)
Intratracheal instillation of $PM_{2.5}$ collected during normal weather and dust storm in China	Wistar rats	Water (sonication)	1.5–37 mg/kg bw for 24 h	Dose-dependent increases in levels of DNA strand breaks (comet) in lung for samples from both normal weather and dust storms	Meng & Zhang (2006b, 2007)
Intratracheal instillation of $PM_{2.5}$ or PM_{10} from locations near to or far away from traffic in Beijing, China	Wistar rats	Water (sonication)	7.5 mg/kg bw once/d for 14 d, and killed at 24 h after the last instillation	Increased level of DNA strand breaks (comet) in lung tissue. $PM_{2.5}$ generated higher levels of DNA strand breaks than PM_{10}. Particles collected closest to traffic generated the highest levels of DNA strand breaks	Zhang et al. (2011)
Intratracheal instillation of TSP samples from Minqin county, Gansu province, China, where sandstorms occurred frequently	Wistar rats	Water (sonication)	1.5, 7.5, or 37.5 mg/kg bw, and killed at 12, 24, or 48 h after the instillation	Dose-dependent increase in level of DNA strand breaks (comet) in lung tissue. Highest levels of DNA strand breaks observed at 12 h, and effects reduced at 24 h after treatment	Xu et al. (2008b)
Intratracheal instillation of SRM 1649 (urban dust)	*ApoE*$^{-/-}$ mice	NA	0.5 mg/kg bw at 26 h and 2 h before being killed	Unaltered levels of DNA strand breaks (comet) in lung tissue	Vesterdal et al. (2014)

bw, body weight; d, day or days; h, hour or hours; NA, not applicable; PM_{10}, particulate matter with particles of aerodynamic diameter < 10 μm; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μm; SRM, standard reference mixture; TSP, total suspended particles.

A study in Thailand with a relatively large benzene exposure gradient (8–50 μg/m^3) among traffic police and office-based police showed no association with levels of DNA strand breaks in leukocytes, whereas there was a correlation between the levels of 1,3-butadiene and levels of DNA strand breaks (Arayasiri et al., 2010). A series of publications from studies of schoolchildren in Bangkok, compared with children in a provincial area (Chonburi), showed that the children exposed to air pollution had higher levels of DNA strand breaks in leukocytes (Buthbumrung et al., 2008; Ruchirawat et al., 2006, 2007). Schoolchildren in Bangkok had higher levels of DNA strand breaks in lymphocytes compared with children from a low-pollution area in Thailand (Tuntawiroon et al., 2007).

(ii) DNA strand breaks in the respiratory system of animals in vivo

Several studies have assessed the level of DNA strand breaks in the lungs of animals after pulmonary exposure to air pollution particles (Table 4.10). Three studies in China have observed increased levels of DNA strand breaks in lung tissue after intratracheal instillation of relatively high doses of air pollution particles (7.5 mg/kg bw and 37 mg/kg bw) (Lin et al., 2009; Meng & Zhang, 2006b, 2007; Zhang et al., 2011). Another study in China collected TSP from a residential area in Minqin county, Gansu province, where sandstorms occurred frequently. Wistar rats were exposed to these sandstorm particles in suspension by intratracheal instillation at doses of 0, 1.5, 7.5, 37.5 mg/kg bw. The TSP exposure caused a dose-dependent increase in the level of

DNA strand breaks; the highest levels of DNA strand breaks were observed at 12 hours, and the effects were reduced at 24 hours after the exposure. The lowest dose that caused significantly increased levels of DNA strand breaks was 1.5 mg/kg bw (Xu et al., 2008b). However, another study on intratracheal instillation of SRM 1649 (i.e. urban dust from Washington, DC, USA) showed that 0.5 mg/kg bw administered twice during 24 hours did not increase levels of DNA strand breaks in lung tissue in mice (Vesterdal et al., 2014). Researchers in Brazil studied native rodents (*Ctenomys minutus*) and showed a correlation between environmental exposure to automobile emission and levels of DNA strand breaks in blood leukocytes (Heuser et al., 2002). Another study showed that dogs from different locations in São Paulo, Brazil, which had similar levels of PM_{10}, also had the same levels of DNA strand breaks in cells from the olfactory or respiratory epithelium (Kimura et al., 2010).

(iii) Human and mammalian cells in vitro

Table 4.11 lists studies that have assessed levels of DNA strand breaks in cultured cells. Several studies have shown that suspensions of PM samples or EOM of PM samples generate the same levels of DNA strand breaks in cultured cells (Brits et al., 2004; Carreras et al., 2013; Gutiérrez-Castillo et al., 2006; Healey et al., 2005; Jayasekher, 2009; Perrone et al., 2013). In addition, SRM 1649 particles retained the ability to generate DNA strand breaks in human fibroblasts after extraction in different solvents, including hexane, acetone, DCM, dimethyl sulfoxide (DMSO), and water (Karlsson et al., 2004). Another study on SRM 1648 (i.e. urban dust collected from St. Louis, Missouri, USA) showed that washed particles and the DCM extract generated lower levels of DNA strand breaks in THP-1 and A549 cells compared with the pristine particles (Don Porto Carero et al., 2001). [The concentration–response relationship was unclear.] Other studies have shown that suspensions of particles from cities in China generated DNA strand breaks in cells, as did the water and DCM extract of the particles (Meng & Zhang, 2007; Yi et al., 2014). Organic extracts of airborne particles with various sizes (< 1.1 μm, 1.1–2.0 μm, 2.0–3.3 μm, 3.3–7.0 μm, and > 7.0 μm) were also collected in a residential area in Taiyuan, China. Concentration-dependent responses of levels of DNA strand breaks in human lymphocytes were observed for airborne particles; small particles generated the highest levels of DNA strand breaks. The lowest effect level for particles smaller than 3.3 μm was 25 μg/mL (Zhang et al., 2004). The air pollution in Taiyuan consisted mainly of emissions from coal combustion, whereas the air pollution in Beijing was a mixture of coal combustion emissions and automobile exhausts. In Guangzhou, China, TSP and PM_{10} samples were collected in a residential area in spring; organic extracts were separated into three fractions by chromatography and used to study the generation of DNA strand breaks. TSP or PM_{10} extracts induced DNA strand breaks in human lymphocytes in a concentration-dependent manner. The aromatic hydrocarbon fraction of the TSP or PM_{10} extract also induced a concentration-dependent increase in DNA strand breaks in human lymphocytes (Xu & Wang, 2008). In addition, the water extracts of $PM_{2.5}$ from Guangzhou on days with haze during summer and winter generated a concentration-dependent increase in DNA strand breaks (Qin et al., 2012).

Suspension solutions of $PM_{2.5}$ collected in Taiyuan, China, during the heating season caused a concentration-dependent increase in levels of DNA strand breaks in rat alveolar macrophage cells (Meng & Zhang, 2005). In another study, organic extracts and water extracts of $PM_{2.5}$ samples collected in Wuwei and Baotou, China, during normal weather or sandstorms caused a concentration-dependent increase in levels of DNA strand breaks in rat alveolar macrophage cells (Meng et al., 2006a). Zhang et al. (2003) reported that organic extracts of $PM_{2.5}$ collected

Table 4.11 DNA strand breaks in mammalian cells in vitro

Particles	Cells	Extraction	Concentration and time	Effect	Reference
PM_{10} (and EOM) from rural, industrial, and urban sites in Flanders, Belgium	Human leukocytes	Water (shaking) or THF:Hx (20:80) at 140 bar and 100 °C	5–20 m^3 air equiv/mL for 24 h	Increased DNA strand breaks for both particles and EOM, although without concentration dependency	Brits et al. (2004)
TSP from Córdoba, Argentina	Human lymphocytes	Methylene chloride (ultrasound)	20–80 µL of extract for 24 h	Concentration-dependent increase in DNA strand breaks	Carreras et al. (2013)
PM, or EOM thereof, from Mexico City, Mexico, or SRM 1649	A549 cells	Water (ultrasound) or DCM	0.05–1.6 m^3/mL equiv for 48 h	Water-soluble and EOM had similar DNA strand break generation potential. Little difference between samples obtained at different locations. $PM_{2.5}$, PM_{10}, and SRM 1649 had the same potency	Gutiérrez-Castillo et al. (2006)
Different size fractions and EOM of PM_{10} from Leeds, United Kingdom	A549 cells	Water (vortexing or brushing off particles from the filter) or DCM (vortexing)	25 µg/mL for 24 h	Increased levels of DNA strand breaks. Organic extract generated a similar level of DNA strand breaks as pristine particles, whereas washed particles generated low levels of DNA strand breaks	Healey et al. (2005)
PM_{10} sampled near a coal power plant in Tuticorin, India	Human lymphocytes	Acid (sonication)	5 µg of aerosol extract per 50 µL for 24 h	Increased levels of DNA strand breaks	Jayasekher (2009)
$PM_{2.5}$ and PM_1 from rural, urban, and remote sites in northern Italy	A549 cells	Water (ultrasound) or MeOH	6 µg/cm^2 for 24 h	Samples from urban areas, collected during spring, were the most potent inducer of DNA strand breaks. $PM_{2.5}$ had higher potency than PM_1	Perrone et al. (2013)
SRM 1649 or its particles after extraction with organic solvents	Human fibroblasts	DCM, Hx, Ac, or DMSO	0.1–100 µg/cm^2 for 24 h	Concentration-dependent increase in DNA strand breaks (comet). Particles generated DNA strand breaks after extraction of organic material	Karlsson et al. (2004)
SRM 1648 or organic extracts thereof	A549 or THP-1 cells	Water or DCM (shaking by hand)	16–1600 ng/mL for 48 h	Inconsistently increased levels of DNA strand breaks in A549 cells. Increased levels of DNA strand breaks in THP-1 cells by particles, but unaltered levels by extracts and washed particles	Don Porto Carero et al. (2001)
$PM_{2.5}$ (or EOM) collected during normal weather and dust storm in China	Human alveolar macrophages	Water or DCM (sonication)	33–300 µg/mL for 4 h	Concentration-dependent increased levels of DNA strand breaks by particles and EOM. Samples from normal weather and dust storms generated the same levels of DNA strand breaks	Meng & Zhang (2007)

Table 4.11 (continued)

Particles	Cells	Extraction	Concentration and time	Effect	Reference
PM_{10} from Beijing, China	A549 cells	Water (sonication)	10 µg/mL for 24 h	Increased DNA strand breaks by particles as well as soluble and insoluble fractions thereof	Yi et al. (2014)
Airborne particulates with various sizes (< 1.1 µm, 1.1–2.0 µm, 2.0–3.3 µm, 3.3–7.0 µm, > 7.0 µm) collected in a residential area in Taiyuan, China. TSP concentration, 0.481 mg/m³	Human peripheral blood from a healthy adult. Cultured human peripheral blood lymphocytes	Airborne particulates with various sizes extracted by Soxhlet method with MeOH, Ac, and DCM for 4 h, respectively. Combined extracts dried and dissolved in DMSO	25, 50, 100, 200 µg/mL particulate extracts (incubation time not reported)	Concentration-dependent increases in level of DNA strand breaks	Zhang et al. (2004)
TSP and PM_{10} samples collected in a residential area in Guangzhou, China, during spring in 2005	Human peripheral blood from a healthy adult	TSP and PM_{10} samples extracted by ultrasonication with DCM and then separated into 3 fractions by chromatography	4 m³/mL for 2 h	Increased generation of DNA strand breaks	Xu & Wang (2008)
PM_{10} samples collected at 4 sites in Dalian, China, during summer and winter in 2006	HepG2 cells	PM_{10} samples extracted by ultrasonication with DCM, Ac, and MeOH for 20 min; subsequently, the 3 extracts were combined, dried, and dissolved in DMSO	0, 7.5, 15, 30 µg/mL for 1 h	Increased generation of DNA strand breaks	Jiang et al. (2011)
$PM_{2.5}$ samples collected at Beijing University, China, in March 2001	Balb/c 3T3 cells	Organic or inorganic extract	0.5, 1, 2, 4 m³/mL equiv for 12 h	Concentration-dependent increases in level of DNA strand breaks	Zhang et al. (2003)
PM_{10} from different locations in Mexico City, Mexico	Balb/c 3T3 cells	Dry sonication and brushing off particles from the filter	2.5–40 µg/cm² for 72 h	Concentration-dependent increases in DNA strand breaks, without clear difference in genotoxicity between locations. At 20 µg/cm² and 40 µg/cm², comet length did not increase beyond that obtained with 10 µg/cm²	Alfaro-Moreno et al. (2002)
PM from a town with many wood stoves and a rural area, Denmark	A549 and THP-1 cells	Mechanical collection from plates	2.5–100 µg/mL for 3 h	Concentration-dependent increase in DNA strand breaks in A549 and THP-1 cells (comet). No difference between particles from different areas	Danielsen et al. (2011)

Table 4.11 (continued)

Particles	Cells	Extraction	Concentration and time	Effect	Reference
$PM_{2.5}$ or PM_{10} from background site in Milan, Italy	BEAS-2B cells	Water (ultrasound)	25 µg/cm² for 24 h	$PM_{2.5}$ exposure increased level of DNA strand breaks. No effect of PM_{10}	Gualtieri et al. (2011)
$PM_{2.5}$ from 5 cities in the USA	IB3-1 and K543 cells	PBS (vortexing)	50 µL of extracts containing 33 µg of extracted $PM_{2.5}$ for 3 h	Increased level of DNA strand breaks, without a clear difference between particles from different cities (concern about number of repeated experiments)	Dellinger et al. (2001)
Coarse or fine particles from Mexico City, Mexico	THP-1 cells	Water	10 µg/mL for 24 h	Increased level of DNA strand breaks (comet), with some difference related to size, location, and time of sampling	De Vizcaya-Ruiz et al. (2006)
$PM_{2.5}$ collected from a busy street or urban background in Copenhagen, Denmark	A549 cells	Water	25 µg/mL for 24 h	Increased level of DNA strand breaks, although not a clear difference between street and background particles	Sharma et al. (2007)
Fine particles from various cities in Germany	A549 cells	Water (sonication)	20 µg/cm² for 3 h	Increased level of DNA strand breaks, although no difference between particles from different cities	Shi et al. (2006)
$PM_{2.5}$ from Beijing and Taiyuan, China	A549 cells	Water (ultrasonic shaking)	5–200 µg/mL for 12 h or 24 h	Concentration-dependent increase in DNA strand breaks (uncertainty about number of repeats and statistics). No difference between cities	Xu & Zhang (2004)
PM_{10} from a busy street in Stockholm, Sweden	A549 cells	Water (sonication)	40 µg/cm² (70 µg/mL) for 4 h	Increased levels of DNA strand breaks in cells after exposure to particles from a busy street (PM_{10}) as well as particles collected when running a road simulator with studded tyres ($PM_{2.5}$ or PM_{10})	Karlsson et al. (2006)
EOM or aqueous extract from $PM_{2.5}$ in Piedmont, Italy	A549 cells	DCM or water (ultrasound)	1–7 m³ equiv for 24 h	Highest levels of DNA strand breaks after exposure to EOM of PM from highway site, whereas urban and industrial sites had the same DNA strand break induction potential. Higher induction of DNA strand breaks by aqueous extracts from industrial site compared with urban and highway sites	Bonetta et al. (2009)

Table 4.11 (continued)

Particles	Cells	Extraction	Concentration and time	Effect	Reference
PM_{10},$PM_{2.5}$, PM_1, and $PM_{0.4}$ from Milan, Italy, collected during summer or winter	A549 cells	Water (sonication)	10 μg/cm² for 24 h	Increased level of strand breaks (γH2AX expression) by all samples collected during winter with a similar intensity	Longhin et al. (2013)
TSP from an urban street in Copenhagen, Denmark	A549 cells	Water (ultrasonication)	2.5–100 μg/mL for 48 h	Concentration-dependent increase in DNA strand breaks (comet assay). No difference between days of sampling	Danielsen et al. (2008)
Particles from a street tunnel in Oslo, Norway, during seasons with or without use of studded tyres	A549 and THP-1 cells	Scraping off the filter	2.5–200 μg/mL for 3 h	Increased DNA strand break sites in A549 and THP-1 cells (comet). No difference between seasons with or without use of studded tyres	Danielsen et al. (2009)
Coal fly ash from a dumping site of a thermal power plant in Aligarh, India	Mononuclear blood cells	DMSO (0.5%) or water (sonication)	3–2400 ppm for 3 h	Concentration-dependent increase for DMSO (0.5%) extract, and unaltered DNA strand break generation by aqueous extract	Dwivedi et al. (2012)
PM_{10} from a busy street in Stockholm, Sweden	A549 cells	Water (vortexing or sonication)	5–40 μg/cm² (9–70 μg/mL) for 4 h	Concentration-dependent increase in DNA strand breaks (comet)	Karlsson et al. (2005)
Airborne PM from Düsseldorf, Germany	A549 cells	Baghouse	1–100 μg/mL for 24 h	Concentration-dependent increase in DNA strand breaks (FADU assay)	Upadhyay et al. (2003)
Oil fly ash collected from a power plant in Sicily, Italy	A549 cells	Water	17.2–68.8 μM $VOSO_4$ equiv for 0.5–4 h	Concentration-dependent increase in levels of DNA strand breaks. Ameliorated by treatment with DFO (results not shown)	Di Pietro et al. (2009)
Fine particles from Düsseldorf, Germany	A549 cells	Water (sonication)	5–20 μg/cm² for 3 h	Increased level of DNA strand breaks by particles, which was diminished by treatment with DFO. Filtered suspensions less potent than suspensions with particles	Knaapen et al. (2002)
EOM of $PM_{2.5}$ and PM_{10} from Paris, Rouen, and Strasbourg, France	HeLa cells	Acetonitrile and ultrasound	200 μL of organic extract for 24 h	Extracts from $PM_{2.5}$, compared with PM_{10}, generated higher levels of DNA strand breaks (comet)	Abou Chakra et al. (2007)

Table 4.11 (continued)

Particles	Cells	Extraction	Concentration and time	Effect	Reference
EOM of PM_{10} from 4 different areas in China	HepG2 cells	Sequential extraction in DCM, Ac, and MeOH	7.5–30 µg/mL for 1 h	Concentration-dependent increase in DNA strand breaks by samples from Kaifaqu district, Dalian (industrial area). Lower levels of DNA strand breaks by samples from 3 other cities. Samples collected during winter more potent than summer samples	Jiang et al. (2011)
EOM of PM_{10} from Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria)	HepG2 cells	DCM	5–150 µg/mL for 2 h	Concentration-dependent increase in DNA strand breaks. No difference between samples with regard to season or location	Gábelová et al. (2004)
EOM of PM_{10} from Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria)	HepG2 cells	DCM	10–250 µg/mL for 2–48 h	Concentration-dependent increase in DNA strand breaks. Little spatial or temporal difference between samples on mass basis	Gábelová et al. (2007)
EOM of PM_{10} from urban sites in Saudi Arabia	Human leukocytes	Ac (Soxhlet tube)	250 µg/mL (incubation time not reported)	Increased levels of DNA strand breaks, with difference between sampling locations	Elassouli et al. (2007)
TSP and PM_{10} from an urban green park in downtown Rome, Italy	Human mononuclear blood cells	Hx and MeOH	4–144 m^3/mL for 2 h	Concentration-dependent increase in levels of DNA strand breaks. No difference in potency between EOM from TSP and PM_{10} samples	Fabiani et al. (2008)
EOM of TSP, PM_{10}, and $PM_{2.5}$ from Parma, Italy	Human leukocytes	Ac and Tl	1–3 m^3 equiv for 1 h	Extracts of $PM_{2.5}$ reported to generate higher levels of DNA strand breaks (comet) than those of TSP and PM_{10}. Number of independent experiments and statistics uncertain	Buschini et al. (2001)
EOM of $PM_{2.5}$ from a low-traffic area in Hong Kong Special Administrative Region, China	Rat fibroblasts	DCM (ultrasound)	570–2321 ng/mL for 72 h	Concentration-dependent increase in levels of DNA strand breaks. Samples collected during winter more potent than summer samples	Hsiao et al. (2000)
EOM of PM_{10} from Teplice, Czech Republic	HepG2 and Caco-2 cells	DCM	1–50 µg/mL for 24 h	Concentration-dependent increase in DNA strand breaks in HepG2 and Caco-2 cells. Winter samples more potent than summer samples	Lazarová & Slamenová (2004)

Table 4.11 (continued)

Particles	Cells	Extraction	Concentration and time	Effect	Reference
EOM of $PM_{2.5}$ from Guangzhou, China	Human lymphocytes	Sequentially, DCM (ultrasound) and Hx	5–20 m^3 air equiv/mL for 2 h	Higher DNA strand break generation of samples collected on days with haze conditions compared with non-haze. Higher generation of DNA strand breaks by samples collected from roadside compared with rooftop	Xu et al. (2008a)
EOM of PM from Taiwan, China	MCF-7 cells	Hx/Ac (ultrasound)	0.04–0.05 m^3 air equiv for 72 h	No clear difference between PM from urban and rural sites with regard to DNA strand break generation	Chen et al. (2013)
EOM of $PM_{2.5}$ from high-traffic area in Suwon, Republic of Korea	BEAS-2B cells	DCM (sonication)	1–50 µg/mL for 24 h	Concentration-dependent increase in DNA strand breaks	Oh et al. (2011)
EOM of road tunnel particles from Shanghai, China	A549 cells	DCM (sonication)	1–400 µg/mL for 24 h	Concentration-dependent increase	Shang et al. (2013)
EOM of PM_{10} from an industrial site in France	HepG2 cells	DCM (Soxhlet extractor)	0.16 µM B[*a*]P and atmospheric samples for 24 h	Increased levels of DNA strand breaks (comet)	Tarantini et al. (2009)

Ac, acetone; B[*a*]P, benzo[*a*]pyrene; DCM, dichloromethane; DFO, deferoxamine; DMSO, dimethyl sulfoxide; EOM, extractable organic matter; equiv, equivalent; h, hour or hours; Hx, hexane; 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; MeOH, methanol; min, minute or minutes; PBS, phosphate-buffered saline; SRM, standard reference mixture; THF, tetrahydrofuran; Tl, toluene; TSP, total suspended particles.

at Beijing University caused a concentration-dependent increase in levels of DNA strand breaks in Balb/c 3T3 cells. The lowest concentration of the organic extract that caused a significantly increased level of DNA strand breaks was 1 m^3 equiv/mL, whereas an aqueous extract of $PM_{2.5}$ had no significant effect on generation of DNA strand breaks (Zhang et al., 2003).

Several studies have found no clear difference in the potency to generate DNA strand breaks of particles collected at different locations (Alfaro-Moreno et al., 2002; Danielsen et al., 2011; Dellinger et al., 2001; De Vizcaya-Ruiz et al., 2006; Sharma et al., 2007; Shi et al., 2006; Xu & Zhang, 2004). Similarly, PM_{10} collected from a busy street in the centre of Stockholm, Sweden, had the same potency on a mass basis as particles collected when running a road simulator (Karlsson et al., 2006). In contrast, aqueous extracts of $PM_{2.5}$ samples from industrial sites showed a higher induction of DNA strand breaks in A549 cells (Bonetta et al., 2009). One study reported that samples of $PM_{0.4}$, PM_1, $PM_{2.5}$, and PM_{10} that were collected during the winter at a background site in Milan, Italy, were more potent than the same fractions collected during the summer (Longhin et al., 2013), whereas three studies reported no temporal variation in PM samples in regard to ability to generate DNA strand breaks (Danielsen et al., 2008; Danielsen et al., 2009, 2011). Studies on differences related to particle size indicated that the urban background $PM_{2.5}$ fraction was more potent than PM_{10} on a mass basis (Gualtieri et al., 2011; Perrone et al., 2013). In general, consistency has been observed in studies showing increased levels of DNA strand breaks by aqueous suspensions of particles from various locations, times of the year, and size fractions. Other studies that have assessed the effect of particles from only a single site also indicated increased levels of DNA strand breaks (Dwivedi et al., 2012; Karlsson et al., 2005; Upadhyay et al., 2003), which could be reduced only slightly by treatment with deferoxamine (DFO) (Di Pietro et al., 2009; Knaapen et al., 2002). This suggests that the content of soluble transition metals such as iron was not the most important constituent in particles for the formation of DNA strand breaks.

It has been shown that acetonitrile-extracted material from $PM_{2.5}$ that was collected from a location close to heavy traffic had higher potency in generating DNA strand breaks in fibroblasts compared with $PM_{2.5}$ samples from other urban zones (Abou Chakra et al., 2007). A comparative investigation of organic extract of $PM_{2.5}$ from samples collected at a highway site (with high traffic intensity) showed higher levels of DNA strand breaks in A549 cells compared with extracts from an urban site (with medium traffic intensity) and an industrial site near a foundry (Bonetta et al., 2009). Also, samples collected in the industrial area of Kaifaqu district, Dalian, China, showed higher potency in generating DNA strand breaks compared with samples from three other areas in China (Jiang et al., 2011). DCM extracts of PM_{10} samples from urban air in Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria) increased the generation of DNA strand breaks in HepG2 cells, whereas there were no clear spatial or temporal differences in potency (Gábelová et al., 2004, 2007). Another study found a difference between DCM extracts of PM_{10} from different locations in Saudi Arabia (Elassouli et al., 2007). EOM (hexane and methanol) of TSP and PM_{10} from an urban background site (a park) in Rome, Italy, had similar potency in generating DNA strand breaks in mononuclear blood cells (Fabiani et al., 2008). Another study in Parma, Italy, showed that EOM of $PM_{2.5}$ was more potent than TSP and PM_{10} in generating DNA strand breaks (Buschini et al. 2001).

Organic extracts of $PM_{2.5}$ or PM_{10} from samples that were collected in three French metropolitan areas in the winter had higher potential to generate DNA strand breaks than extracts collected in the summer (Abou Chakra

et al., 2007). Organic extracts of PM_{10} samples from an industrial area in China were more potent in generating DNA strand breaks in HepG2 cells when collected during the winter (Jiang et al., 2011). The same was shown for EOM of $PM_{2.5}$ collected in Hong Kong Special Administrative Region, China; the samples from the winter were more potent than those from the summer (Hsiao et al., 2000). EOM of PM_{10} particles collected in Teplice, Czech Republic, increased the levels of DNA strand breaks in HepG2 and Caco-2 cells, and the samples collected during the summer had a stronger effect than those collected during the winter (Lazarová & Slamenová, 2004). In addition, EOM of $PM_{2.5}$ collected on days with haze had higher potential to generate DNA strand breaks compared with extracts of samples collected on days without haze (Xu et al., 2008a). The association between EOM of PM from China, France, and the Republic of Korea has also been shown in other studies, albeit without assessment of temporal, spatial, or particle size differences (Chen et al., 2013; Oh et al., 2011; Shang et al., 2013; Tarantini et al., 2009).

(iv) Acellular test systems

Studies in acellular test systems are summarized in Supplemental Table S13 (available online). The studies on the ability of PM to generate strand breaks in DNA have typically used relaxation of plasmid or bacteriophage DNA, in which the supercoil structure is relaxed by introduction of DNA strand breaks. The literature on studies of air pollution particles generally shows that PM from air pollution is associated with relaxation of supercoiled DNA. Early studies showed that PM_{10} from Edinburgh, United Kingdom, increased the relaxation of supercoiled DNA and that this was reduced by treatment with an antioxidant (mannitol) or a metal chelating agent (DFO) (Donaldson et al., 1997; Gilmour et al., 1996). The strand-breaking potential of $PM_{2.5}$ samples from Baton Rouge, Louisiana, USA, was reduced in the presence of superoxide dismutase or catalase (Dellinger et al., 2001). The role of iron mobilization was demonstrated by studies showing that SRM 1648 and SRM 1649 were associated with strand breakage in DNA only in the presence of ascorbate, which functions as reductant (Smith & Aust, 1997). One study on coal fly ash also found increased generation of DNA strand breaks (Dwivedi et al., 2012). Particles collected in London, United Kingdom, in 1958 from an unknown site generated strand breaks in a concentration-dependent manner (Whittaker et al., 2004). Studies on different particle size fractions have produced mixed results, showing both higher potency in supercoil relaxation of small particles (Healey et al., 2005; Koshy et al., 2009; Lingard et al., 2005; Reche et al., 2012; Shao et al., 2006) and higher potency of coarse particles than fine particles (Greenwell et al., 2002). In addition, it has been shown that the potency of PM samples collected from a location near a busy motorway and steelworks depended on the wind direction, with the highest potency of strand scission activity observed for PM samples when the wind came from the motorway (Moreno et al., 2004). Another study showed that $PM_{2.5}$ from an urban site in Shanghai, China, was more potent in plasmid DNA supercoil relaxation compared with samples from a suburban site and that samples collected during the winter were more potent those collected during the summer (Senlin et al., 2008). Collectively, the studies indicate that aqueous suspensions of PM and water-soluble constituents have the ability to generate DNA strand breaks in naked DNA, which is driven mainly by production of ROS by transition metals.

Airborne particles from many cities in China have been reported to induce plasmid DNA relaxation in acellular conditions. Samples collected during sandstorms were less potent than non-sandstorm samples (Shi et al., 2004). Water extracts or particle suspensions of PM_{10} samples collected in four seasons in Lanzhou showed the ability to generate strand breaks in

plasmid DNA. Average values of TD20 (the level causing 20% of DNA damage) were 17, 625, 56, and 260 μg/mL in the winter, spring, summer, and autumn, respectively, for PM_{10} suspensions. Water extracts caused slightly lower induction of DNA strand breaks, with higher TD20. Suburban PM_{10} samples showed higher TD20 values than samples from Lanzhou. Similar to the study in Beijing, particles were collected during dust storm episodes or after days with rain. The results showed lower ability to generate DNA strand breaks (TD20 > 1000 μg/mL) compared with the PM that was collected in Lanzhou, although the TD20 values correlated negatively with metal concentration (Xiao et al., 2009). In Macao Special Administrative Region, China, PM_{10} samples from three sites (Sun Yat Sen Municipal Park, Avenida de Horta e Costa, and Macao University on Taipa Island) showed that whole PM_{10} suspension samples caused formation of DNA strand breaks with values of TD30 (the level causing 30% of DNA damage) of 3, 10, and 20 μg/mL, respectively, for the three sites. Water extracts showed slightly higher TD30 values (Shen et al., 2009).

In summary, the majority of human studies have shown positive associations between exposure to particulate air pollution and elevated levels of DNA strand breaks in leukocytes as well as nasal epithelial cells. In animal studies, elevated levels of DNA strand breaks in the lung have been noted in studies on doses of PM by instillation, whereas lower doses have not increased levels of DNA strand breaks. Studies in cultured cells and acellular conditions have provided supporting mechanistic evidence for the ability of outdoor air PM to generate DNA strand breaks.

(d) Chromatin damage in sperm

Four studies performed in the Czech Republic (Selevan et al., 2000; Rubes et al., 2005, 2007, 2010) evaluated the association between exposure of men to polluted outdoor air and chromatin damage in their sperm using the sperm chromatin structure assay (SCSA). Table 4.12 shows that all of these studies found an association between chromatin damage in sperm and exposure to elevated concentrations of various pollutants of outdoor air, including CO_2, PM_{10}, SO_2, NO_x, B[*a*]P, carcinogenic PAHs, benzene, and TSP.

(e) Oxidatively damaged nucleobases

(i) Humans

The associations between air pollution and biomarkers of oxidatively damaged nucleobases in human leukocytes have been assessed in a variety of biomonitoring studies, including controlled exposure, panel, and cross-sectional studies. A previous assessment of studies measuring oxidized nucleobases highlighted that approximately half of the published studies had either suboptimal study design or measurement of 8-oxodG by unspecific methods (Møller & Loft, 2010). The discussion of the biomonitoring studies adheres to this critical assessment of the studies. The studies on associations between exposure to air pollution particles and levels of oxidatively damaged DNA in cells from humans are listed in Table 4.13.

The first of two studies in Benin recruited taxi-moto drivers in the city of Cotonou, which has high levels of outdoor air pollution, as determined by assessments including total PAHs (35–103 ng/m^3) and urinary excretion of benzene metabolites (*S*-PMA, 6.8–9.3 μmol/mol creatinine), and a control group in a village with low air pollution (PAHs, 7.3 ng/m^3; *S*-PMA, 4.2 μmol/mol creatinine). This revealed that the taxi-moto drivers had higher levels of 8-oxodG in lymphocytes (21 lesions/10^6 dG) compared with controls in the village (11 lesions/10^6 dG) (Ayi-Fanou *et al.*, 2006). [The high background levels of 8-oxodG suggest spurious oxidation of the DNA during the HPLC-electrochemical detection (ECD) measurement, and the study design with comparison of subjects in the city and

Table 4.12 DNA fragmentation and abnormal chromatin in sperm in men exposed to outdoor air pollution

Subjects (number)	End-point	Associated air pollutant	Finding	Reference
Men (18 yr) in Teplice District, Czech Republic (272)	Abnormal chromatin (SCSA)	PM_{10}, TSP, CO_2	+	Selevan et al. (2000)
Young men in Teplice District, Czech Republic (36)	SCSA	PM_{10}, SO_2, NO_x	+	Rubes et al. (2005)
Young men in Teplice District, Czech Republic (35)	SCSA among *GSTM1* null	PM_{10}, SO_2, NO_x	+	Rubes et al. (2007)
Outdoor police in Prague, Czech Republic, sampled in winter (high pollution) and spring (low pollution) (47)	DNA damage (DFI by SCSA)	B[*a*]P, carcinogenic PAHs, benzene	+	Rubes et al. (2010)

+, positive; B[*a*]P, benzo[*a*]pyrene; CO_2, carbon dioxide; DFI, DNA fragmentation index; NO_x, nitrogen oxides; PAHs, polycyclic aromatic hydrocarbons; PM_{10}, particulate matter with particles of aerodynamic diameter < 10 μm; SCSA, sperm chromatin structure assay; SO_2, sulfur dioxide; TSP, total suspended particles; yr, year or years.

village is not optimal.] A subsequent study also used taxi-moto drivers in Cotonou and village controls, as well as groups of subjects with intermediate exposure to air pollution as determined by urinary excretion of benzene metabolites and the number concentration of UFP (midday 1-hour average, 6961–265 145 UFP/cm³). There were clear gradients in both air pollution levels (assessed as *S*-PMA) and levels of formamidopyrimidine DNA glycosylase (FPG)-sensitive sites in PBMCs between the subjects living in areas with different air pollution levels (Avogbe et al., 2005).

A panel study of students who were living in the centre of Copenhagen, Denmark, showed a positive association between personal exposure to $PM_{2.5}$ (10–24.5 μg/m³) and levels of 8-oxodG in lymphocytes, whereas the exposure did not correlate with levels of FPG-sensitive sites in lymphocytes (Sørensen et al., 2003a). In addition, there was a correlation between the levels of 8-oxodG in lymphocytes and the concentration of water-soluble transition metals in $PM_{2.5}$ that was collected over a 2-day period for each subject (Sørensen et al., 2005). The same study also showed that there was no correlation between background mass concentration of $PM_{2.5}$ measured at a stationary monitoring station or personal exposure to NO_2 and levels of 8-oxodG in lymphocytes. Another study of residents of Copenhagen used benzene as a marker of urban air pollution exposure and showed an association between urinary excretion of *S*-PMA and levels of 8-oxodG in lymphocytes, whereas the levels of endonuclease III (ENDOIII)/FPG sites were unaltered in lymphocytes (Sørensen et al., 2003c). The effect of personal exposure to UFP in air pollution was investigated in people bicycling for approximately 90 minutes in the laboratory or on traffic-heavy streets in Copenhagen. This study showed a positive association between personal exposure to UFP and levels of FPG-sensitive sites in PBMCs (Vinzents et al., 2005). The same group of researchers also studied controlled exposure to air from a busy street in Copenhagen and reported correlations between particles in the size mode with a median diameter of 23 nm (representing SVOCs of diesel exhaust) and the size mode with a median diameter of 57 nm (representing carbonaceous soot) and levels of FPG-sensitive sites in PBMCs (Bräuner et al., 2007).

A study in Florence, Italy, showed no correlation between outdoor ozone concentrations

Table 4.13 Exposure to air pollution and oxidatively damaged DNA in human leukocytes

Study population	Exposure assessment	Sex, age, smoking, number	Biomarker	Effect (notes)	Reference
Taxi-moto drivers (n = 35) and rural controls (n = 6) in Cotonou, Benin	Outdoor (stationary) concentrations of PAHs and benzene. Urinary excretion of *S*-PMA and 1-OHP	M 36 ± 5 yr NS n = 41	8-oxodG (HPLC-ECD)	Highest level in leukocytes of exposed subjects (high background level of 8-oxodG: 11 lesions/10^6 dG)	Ayi-Fanou et al. (2006)
Taxi-moto drivers, people living or working near busy roads, and rural controls in Benin	Outdoor (stationary) sampling of UFP (midday 1 h concentration at a busy street intersection and a town square in a rural village, 265 145 and 6 961 UFP/cm^3, respectively) and urinary excretion of *S*-PMA	M 34 ± 10 yr NS n = 135	FPG sites (comet)	Positive association between *S*-PMA excretion and FPG sites in PBMCs	Avogbe et al. (2005)
Panel study of students living in Copenhagen, Denmark	Personal $PM_{2.5}$: 16.1 (10–24.5) μg/m^3 $PM_{2.5}$: 9.2 (5.3–14.8) μg/m^3 (stationary monitoring stations)	M, F 20–33 yr NS n = 50	8-oxodG (HPLC-ECD) FPG sites (comet)	Correlation between personal exposure to $PM_{2.5}$ and 8-oxodG in lymphocytes, whereas no correlation between $PM_{2.5}$ and FPG sites. No correlation between biomarkers and stationary (urban background) measurements of $PM_{2.5}$	Sørensen et al. (2003a, 2005)
Subjects living in Copenhagen, Denmark	Benzene (personal exposure and urinary excretion of *S*-PMA)	M, F 27–46 yr S/NS n = 40	ENDOIII/FPG sites (comet) 8-oxodG (HPLC-ECD)	Positive association between urinary excretion of *S*-PMA and 8-oxodG in lymphocytes, whereas no association with ENDOIII/FPG sites	Sørensen et al. (2003c)
Subjects exposed to outdoor air while bicycling in Copenhagen, Denmark	Personal UFP: 32 400 and 13 400 UFP/cm^3 PM_{10}: 23.5 μg/m^3 (street) and 16.9 μg/m^3 (background) NO_2: 32.1 and 24.2 μg/m^3 (street) and 11.3 μg/m^3 (background)	M, F 25 ± 3 yr NS n = 15	FPG sites (comet)	Increased levels of FPG sites in PBMCs after bicycling in the traffic compared with bicycling in the laboratory	Vinzents et al. (2005)
Controlled exposure to outdoor air in a chamber for 24 h in Copenhagen, Denmark	Personal UFP: 6169–15 362 UFP/cm^3 (unfiltered air) and 91–542 UFP/cm^3 (filtered air) NO_x: 25.8 ppb (unfiltered air), 28.3 ppb (filtered air), 11.6 ppb (background), and 59.5 ppb (busy street) O_3: 12.1 ppb (unfiltered air), 4.3 ppb (filtered air), 30.1 ppb (background), and 19.5 ppb (busy street)	M, F 20–40 yr NS n = 29	FPG sites (comet)	Decreased levels of FPG sites in PBMCs after exposure to filtered air compared with unfiltered air	Bräuner et al. (2007)

Table 4.13 (continued)

Study population	Exposure assessment	Sex, age, smoking, number	Biomarker	Effect (notes)	Reference
Healthy subjects in Florence, Italy	O_3: 15–75 μg/m³ (stationary monitoring station)	M, F 24–80 yr S/NS *n* = 79	FPG sites (comet)	No association between O_3 exposure (days 3–30 before sampling) and levels of FPG sites in leukocytes	Giovannelli et al. (2006)
Subjects exposed to traffic (*n* = 44) and controls (*n* = 27) in Florence, Italy	O_3 (levels NR)	M, F 35–64 yr S/NS *n* = 71	FPG sites (comet)	Statistically non-significant higher level of FPG sites in leukocytes of exposed subjects. Correlation between O_3 exposure (days 60–90 before sampling) and levels of FPG sites in lymphocytes	Palli et al. (2009)
Police officers working in traffic (*n* = 24) or indoors (*n* = 24) in Bangkok, Thailand	Personal benzene (8–50 μg/m³), *t,t*-MA, *S*-PMA, and 1,3-butadiene (0.3–4.1 μg/m³) exposure	M 24–58 yr NS *n* = 48	8-oxodG (HPLC-ECD)	Increased levels of 8-oxodG in leukocytes of police officers working in traffic compared with those working indoors. Correlation between personal 1,3-butadiene exposure and levels of 8-oxodG in leukocytes	Arayasiri et al. (2010)
Subjects living near Map Ta Phut Industrial Estate, in a location with steel, oil refinery, and petrochemical factories (*n* = 58), factory workers (*n* = 67), and controls (*n* = 48) in Thailand	None	M, F 32 ± 7 yr S/NS *n* = 173	M_1dG adducts (^{32}P-postlabelling)	Increased M_1dG adducts in leukocytes of factory workers and residents of the polluted area compared with controls	Peluso et al. (2010, 2012)
Children living in a rural area (Chonburi) (*n* = 109) and an urban area (Bangkok) (*n* = 62) in Thailand	Benzene (outdoor monitoring and personal exposure)	M 9–13 yr NS *n* = 171	8-oxodG (HPLC-ECD)	Increased 8-oxodG in leukocytes. Positive correlation between individual benzene exposure and 8-oxodG in leukocytes	Buthbumrung et al. (2008)
Subjects living in a traffic-congested area in Bangkok, Thailand	$PM_{2.5}$: 183 ± 37 μg/m³	M, F 18–58 yr NS *n* = 50	8-oxodG (HPLC-MS/MS)	Association between $PM_{2.5}$ exposure and 8-oxodG in leukocytes	Vattanasit et al. (2014)

Table 4.13 (continued)

Study population	Exposure assessment	Sex, age, smoking, number	Biomarker	Effect (notes)	Reference
Police officers and controls in Prague, Czech Republic	$PM_{2.5}$ (stationary monitoring data: 33 ± 40 µg/m³ in January and 15 ± 9 µg/m³ in September) PAHs (personal exposure: 8.9 ± 1.4 µg/m³ in January and 3.1 ± 0.5 ng/m³ in September)	M 31 yr and 35 yr (median) NS *n* = 65	ENDOIII/FPG sites (comet)	Highest levels in lymphocytes of exposed subjects. Positive correlation between PAH exposure and oxidative DNA damage in samples collected in January	Novotna et al. (2007)
Bus drivers (*n* = 50), garage workers (*n* = 20), and controls (*n* = 50) in Prague, Czech Republic	Personal PAH concentration	M 24–66 yr NS *n* = 120	ENDOIII/FPG (comet)	No differences in the levels of ENDOIII/FPG sites in lymphocytes between exposed groups and control group	Bagryantseva et al. (2010)
Mothers living in Teplice (*n* = 594) or Prachatice (*n* = 297), Czech Republic	Air pollution levels not specified, but levels of PM typically higher in Teplice than in Prachatice	F 25 ± 5 yr S/NS *n* = 891	8-oxodG (ELISA)	No difference in placental levels of 8-oxodG between polluted city and control city. No association between air pollution markers and 8-oxodG in multivariable-adjusted models	Rossner et al. (2011a)
Police officers, bus drivers, and controls in Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria)	PAH concentration in personal $PM_{2.5}$ samples	M 34.1 ± 9 yr S/NS *n* = 356	8-oxodG (LC-MS/MS) M_1dG adducts (immunoslot blot)	Higher levels of 8-oxodG in lymphocytes of police officers in Košice compared with controls, whereas no effect seen in police officers in Prague. Significantly higher levels of M_1dG adducts in exposed subjects in Sofia	Singh et al. (2007b)
Children living in a low-pollution area (*n* = 12) and in Mexico City (*n* = 86), Mexico	PM_{10}: < 14 to 53–61 µg/m³ O_3: < 10 to 261 ppb (max)	M, F 6–13 yr NR *n* = 98	8-oxodG (immunohistochemistry)	Higher levels of 8-oxodG in nasal biopsies from children in Mexico City compared with children in the low-pollution area	Calderón-Garcidueñas et al. (1999)

dG, deoxyguanosine; ELISA, enzyme-linked immunosorbent assay; ENDOIII, endonuclease III; F, female; FPG, formamidopyrimidine DNA glycosylase; h, hour or hours; HPLC-ECD, high-performance liquid chromatography-electrochemical detection; LC-MS/MS, liquid chromatography-tandem mass spectrometry; M, male; M_1dG, malondialdehyde–deoxyguanosine; NO_2, nitrogen dioxide; NO_x, nitrogen oxides; NR, not reported; NS, non-smokers; 1-OHP, 1-hydroxypyrene; 8-oxodG, 8-oxo-7,8-dihydro-2′-deoxyguanosine; O_3, ozone; PAHs, polycyclic aromatic hydrocarbons; PBMCs, peripheral blood mononuclear cells; 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; S, smokers; *S*-PMA, *S*-phenyl mercapturic acid; *t,t*-MA, *trans*,*trans*-muconic acid; UFP, ultrafine particles; yr, year or years.

(days 3–30 before sampling) and levels of FPG-sensitive sites in lymphocytes from healthy subjects (Giovannelli et al., 2006). In a subsequent study, a positive correlation was shown between ozone levels (days 60–90 before sampling) and levels of FPG-sensitive sites in lymphocytes of traffic-exposed workers (Palli et al., 2009).

A study in Bangkok, Thailand, with a relatively large benzene exposure gradient (8–50 μg/m^3) between traffic police and office-based police showed no association with levels of 8-oxodG, whereas there was a correlation between personal 1,3-butadiene exposure and levels of 8-oxodG in leukocytes (Arayasiri et al., 2010). Another study in Thailand on malondialdehyde–deoxyguanosine (M_1dG) adducts, a biomarker of oxidative stress and lipid peroxidation, showed that residents living in a location near steel, oil refinery, and petrochemical factories had higher levels of DNA adducts in leukocytes (3.7 ± 0.4 lesions/10^8 nucleotides) than subjects in a location with a low air pollution level (2.9 ± 0.4 lesions/10^8 nucleotides) (Peluso et al., 2010; Peluso et al., 2012). One study on schoolchildren in Bangkok, compared with children in a provincial area (Chonburi), showed that the children exposed to air pollution had higher levels of 8-oxodG in leukocytes (Buthbumrung et al., 2008). In another study on healthy subjects living in traffic-congested areas in Bangkok, levels of 8-oxodG in leukocytes were significantly correlated with concentrations of individual exposure to $PM_{2.5}$ (Vattanasit et al., 2014).

Several studies in the Czech Republic have assessed biomarkers of oxidatively damaged DNA in people with different occupations or living in areas characterized by high or low air pollution levels. A study of police officers showed higher levels of ENDOIII/FPG sites in lymphocytes in the season with a high level of air pollution exposure ($PM_{2.5}$, 33 μg/m^3), whereas there was no effect in the season with a low level of air pollution exposure ($PM_{2.5}$, 15 μg/m^3) (Novotna et al., 2007). Studies of bus drivers, garage workers, and office workers (controls) indicated no associations between air pollution measures and levels of ENDOIII/FPG sites in lymphocytes from exposed subjects and controls (Bagryantseva et al., 2010). There was no difference in placental levels of 8-oxodG between mothers living in Teplice (urban area) and those living in Prachatice (rural area), and there was a lack of association between air pollution exposure levels and levels of 8-oxodG in multivariable-adjusted models (Rossner et al., 2011a). This research group also participated in the EXPAH project on associations between air pollution exposures in Prague (Czech Republic), Košice (Slovakia), and Sofia (Bulgaria) and biomarkers of genotoxicity in samples from male police officers, bus drivers, and office workers (Taioli et al., 2007). This study showed that police officers in Košice had higher levels of 8-oxodG in lymphocytes compared with controls, whereas there was no effect in police officers in Prague (Singh et al., 2007b). [The Working Group noted that there were very high levels of 8-oxodG in the reference group (i.e. 54 lesions/10^6 nucleotides, corresponding to 244 lesions/10^6 dG), indicating spurious oxidation during the measurement of 8-oxodG.] This study also showed that PAH-exposed subjects from Sofia had higher levels of lipid peroxidation-derived M_1dG adducts in lymphocytes, measured by immunoslot blot, compared with controls from the same city (Singh et al., 2007b).

One study showed that nasal biopsies from children living in areas with a low level of air pollution (a Pacific coastal town) had lower levels of immunostaining intensity for 8-oxodG compared with biopsies from children living in Mexico City, with a high level of air pollution (Calderón-Garcidueñas et al., 1999).

In summary, a substantial number of studies from humans (11 out of 13 studies) have reported positive associations between exposure to air pollution and levels of oxidatively damaged DNA in leukocytes.

(ii) Experimental systems

Few studies have assessed the level of oxidatively damaged DNA in lungs of animals after exposure to air pollution (see Supplemental Table S14, available online). Studies in rodents have reported no effect of intratracheal instillation of PM on levels of oxidatively damaged DNA in lung tissue. Studies in cultured cells (see Supplemental Table S15, available online) and in acellular test systems (see Supplemental Table S16, available online) have examined the induction of oxidatively damaged nucleobases by PM. Aqueous suspensions of PM from urban areas, mainly in Europe, have generated increased levels of oxidatively damaged DNA in cultured cells. There is also some evidence showing that organic extracts of PM are associated with generation of oxidatively damaged DNA in cultured cells. Aqueous suspensions of PM from urban areas, mainly in Europe, have generated increased levels of 8-oxodG in acellular conditions.

(f) Other damage

Several studies used a variety of other assays to assess the genotoxic activity of outdoor air or samples derived from outdoor air. Most used bacterial reporter assays that assess induction of error-prone DNA repair (the SOS response). The results of these studies are summarized in Supplemental Table S17 (available online). In summary, extracts, including inorganic, organic, and simulated lung fluid extracts of airborne PM from a variety of urban and industrial sites, induced significant dose-related increases in DNA damage in both bacteria and mammalian cells.

4.2.4 Gene expression

(a) Humans

See Table 4.14.

As noted in several reviews (e.g. Holloway et al., 2012), exposure of humans to air pollution can result in altered expression of a variety of genes, especially those in pathways associated with DNA damage and repair, oxidative stress, immune response, and so on.

Studies in the Czech Republic (van Leeuwen et al., 2006, 2008) evaluated gene expression in children and adults living in a rural area (Prachatice) versus those living in an urban area (Teplice). Genes that showed differential expression between the two groups of children (and two levels of outdoor air pollution) were largely in the nucleosome assembly. In the same study population, more differential gene expression between the two groups of children was observed compared with the differential gene expression between two groups of adults from the same populations. There was little overlap between the genes expressed differentially between the children and the adults; in children, the pathways most affected by the outdoor air pollution were the nucleosome and immune pathways.

A separate study in the Czech Republic found higher expression of the DNA repair gene *XRCC5* in the blood of residents of Ostrava (more polluted) than in the blood of residents of Prague (less polluted). The higher gene expression was associated with higher concentrations of carcinogenic PAHs in outdoor air (Rossner et al., 2011b).

Huang et al. (2010) exposed three subjects in a chamber in a cross-over design to filtered air or ultrafine particles (50 μg/m^3 for 2 hours) from Chapel Hill, North Carolina, USA, and identified differential expression for 10 genes in a variety of pathways, including inflammation and oxidative stress response, that could discriminate between types of PM exposure.

Hebels et al. (2011) studied nitrosamine exposure and gene expression in human lymphocytes from women participating in a mother–newborn study in Denmark. Participants were non-smoking pregnant women, with no

Table 4.14 Changes in gene expression in humans exposed to outdoor air pollution

Country	Subjects	Assay	Results		Reference
Czech Republic	Blood from 23 children from an urban area (Teplice) versus 24 children from a rural area (Prachatice)	Agilent Human 22K oligo microarray	Increased expression of genes in nucleosome response pathways	+	van Leeuwen et al. (2006)
Czech Republic	Blood from 12 adults from an urban area (Teplice) versus 12 adults from a rural area (Prachatice)	Agilent Human 22K oligo microarrays	More differential gene expression between the two sets of children than the two sets of adults, and little overlap between the children and adults; nucleosome and immune pathways most affected	+	van Leeuwen et al. (2008)
USA	Blood from 3 subjects exposed to ultrafines at 50 µg/m³ for 2 h	Affymetrix HU133 plus 2 microarray	Altered expression of genes in oxidative stress response and inflammation pathways	+	Huang et al. (2011)
Czech Republic	Blood from 64 subjects from Prague (less polluted) and 75 subjects from Ostrava (more polluted)	qPCR	Increased expression of *XRCC5* DNA repair gene among subjects from Ostrava; associated with increased concentration of c-PAHs	+	Rossner et al. (2011b)
Denmark	Blood and urine from 29 women participating in a mother–newborn biomarker study	Agilent 4x44K whole human genome microarrays NOC excretion by GC-MS	Differences in levels of NOC excretion are mainly associated with modifications in amino acid metabolism, apoptosis and survival, cell adhesion, and a few other signalling and metabolism pathways		Hebels et al. (2011)

+, positive; c-PAHs, carcinogenic polycyclic aromatic hydrocarbons; GC-MS, gas chromatography-mass spectrometry; h, hour or hours; NOC, *N*-nitroso compound; PCR, polymerase chain reaction.

residential environmental tobacco smoke and who spent the majority of their time at home. Modifications in cytoskeleton remodelling, cell cycle, apoptosis and survival, signal transduction, immune response, G-protein signalling, and development pathways were observed.

(b) Experimental systems

(i) In vivo

Several studies evaluated gene expression in lung, brain, or adipose tissue after inhalation exposure of rodents to outdoor air or to concentrated air particles (CAPs) (André et al., 2006; Heidenfelder et al., 2009; Bos et al., 2012; Soberanes et al., 2012; Tablin et al., 2012; Ljubimova et al., 2013; Mendez et al., 2013; Rowan-Carroll et al., 2013) (see Supplemental Table S18, available online). Other studies involved evaluation of gene expression in lung tissue of rats after intratracheal instillation of PM from urban air (Kooter et al., 2005; Wise et al., 2006).

(ii) In vitro

In vitro studies are summarized in Supplemental Table S19 (available online).

4.2.5 Epigenetic effects

(a) Humans

(i) DNA methylation

Collectively, the available studies generally show an association between DNA methylation and outdoor air pollution (see Table 4.15). Although air pollution enhances methylation of some genes, it reduces methylation of others. It is likely that this reflects different pathways and involves different components of air pollution (metals, PAHs, VOCs, PM, etc.). Nonetheless, the relatively consistent association between methylation of DNA and exposure to air pollution, resulting in altered gene expression, indicates that this is another mechanism by which air pollution may influence risk of cancer.

(ii) Leukocyte telomere length

Three studies have evaluated leukocyte telomere length relative to exposure to outdoor air pollution (see Table 4.16).

Truck drivers in Beijing, China, had longer telomeres than office workers (Hou et al., 2012). For both the truck drivers and the office workers, an increase in telomere length was associated with personal $PM_{2.5}$ concentration, personal elemental carbon concentration, and outdoor PM_{10} concentration on the day that blood was drawn from the subjects. However, shorter telomere length was associated with the PM_{10} concentration averaged over the 2 weeks before the blood draw. These results indicate that longer telomere length is associated with short-term exposure to outdoor air PM, consistent with an effect of PM on telomeres during acute inflammatory responses. In contrast, long exposures to PM may shorten telomeres due to extended exposures to pro-oxidants.

A population living in Massachusetts, USA, was evaluated for telomere length, which was compared with modelled exposure to carbon black as a marker for traffic-related particles (McCracken et al., 2010). This study found that telomere length shortened as carbon black exposure increased. This result is consistent with the study in China, showing that prolonged exposure to airborne particles is associated with shortened telomeres.

A third study of telomere length found shorter telomeres among traffic officers in Milan, Italy, compared with office workers (Hoxha et al., 2009). Among the traffic officers, the adjusted mean telomere length was shorter in subjects working in high traffic density compared with low traffic intensity. An additional exposure assessment found that telomere length decreased with increasing concentrations of personal exposure to benzene and toluene. Collectively, these studies show that leukocyte telomere length is shortened in subjects chronically exposed to air pollution.

(b) Experimental systems

In the study by Yauk et al. (2008), male C57BL/CBA mice were exposed for 6 weeks in situ to outdoor air near two integrated steel mills and a major highway in Hamilton, Canada; control mice breathed the same air but filtered through HEPA filters. Sperm DNA was hypermethylated in the mice breathing the unaltered outdoor air compared with those breathing the HEPA-filtered air, and this persisted after removal of the mice from the polluted air.

Soberanes et al. (2012) exposed male C57BL/6 mice to $PM_{2.5}$ CAPs from an unspecified urban area for 8 hours per day for 9 weeks. This exposure produced hypermethylation of the promoter region of the *p16* gene in the lung.

Table 4.15 Changes in DNA methylation in humans exposed to outdoor air pollution

Country	Subjects	End-points	Associated air pollutant	Reference
USA	718 residents of Greater Boston, Massachusetts	1097 blood samples assayed for LINE-1 methylation	Decreased LINE-1 methylation with increased black carbon ($P = 0.002$) Decreased LINE-1 methylation with increased $PM_{2.5}$ ($P < 0.001$)	Baccarelli et al. (2009)
USA	706 elderly residents of Greater Boston, Massachusetts	1406 blood samples assayed for LINE-1 and *Alu* methylation	Decreased LINE-1 methylation associated with increased SO_4, and decreased *Alu* methylation associated with increased black carbon	Madrigano et al. (2011)
Italy	63 male steel workers in Brescia	Methylation of *APC*, *P16*, *TP53*, and *RASSF1A* tumour suppressor genes	Increased methylation of *APC* ($P = 0.005$) and *P16* ($P = 0.006$) associated with increased metal-rich air particulate. Decreased methylation of *TP53* ($P = 0.015$), decreased methylation of *RASSF1A* ($P < 0.001$), and increased methylation of *APC* associated with increased PM_{10}	Hou et al. (2011)
USA	940 children in southern California	Methylation of *NOS2* promoter	Decreased methylation of *NOS2* promoter associated with increased $PM_{2.5}$ ($P = 0.01$)	Salam et al. (2012)
USA	699 elderly residents of Greater Boston, Massachusetts	1377 blood samples assayed for methylation of *NOS2* and *GCR* genes	Decreased methylation of *NOS2* promoter associated with increased black carbon and $PM_{2.5}$. No association between methylation of the *GCR* gene and pollutants	Madrigano et al. (2012)
Czech Republic	Children (average age, 11.6 yr); 100 in the more-polluted Ostrava region and 100 in the less-polluted Prachatice region	Methylation at 27 578 loci	Decreased methylation at 53 CpG sites in children from Ostrava, which had increased B[*a*]P, benzene, NO_2, metals, and $PM_{2.5}$ in outdoor air compared with Prachatice	Rossnerova et al. (2013)
USA	Cord blood from 164 non-smoking African-American and Dominican-American mothers in the New York City area	Global DNA methylation	Decreased global methylation in cord blood associated with increased maternal exposure to outdoor air PAH concentration	Herbstman et al. (2012)
Italy	20 male steel workers in Brescia with high exposure and 20 with low exposure to metal-rich air	Methylation of *MT-TF*, *MT-RNR1*, and D-loop of mtDNA	Increased methylation of *MT-TF* and *MT-RNR1* associated with increased metal-rich PM ($P = 0.025$) No association between D-loop methylation and pollutants	Byun et al. (2013)
Italy	20 male filling station attendants in Milan with high exposure and 20 with low exposure to benzene	Methylation of *MT-TF*, *MT-RNR1*, and D-loop of mtDNA	No association between methylation of any genes and pollutants	
China	20 truck drivers in Beijing with high exposure and 20 with low exposure to elemental carbon	Methylation of *MT-TF*, *MT-RNR1*, and D-loop of mtDNA	No association between methylation of any genes and pollutants	

Table 4.15 (continued)

Country	Subjects	End-points	Associated air pollutant	Reference
USA	704 elderly male residents of Greater Boston, Massachusetts	Methylation of *Alu*, LINE-1, *F3*, *TLR-2*, and *ICAM-1*	Decreased methylation of LINE-1 associated with increased black carbon and sulfates. Increased methylation of *Alu* and *TLR-2* associated with NO_2 and particle number	Bind et al. (2012)
Thailand	67 steel and petrochemical workers in Ma Ta Phut Industrial Estate, 65 residents of Ma Ta Phut Industrial Estate, and 45 rural residents	Methylation of LINE-1, *TP53*, *HIC1*, *P16*, and *IL-6*	Exposed population had decreased methylation of LINE-1 ($P < 0.001$), *TP53* ($P < 0.001$), and *IL-6* ($P = 0.027$) but increased methylation of *HIC1* ($P < 0.001$). Decreased methylation associated with increased DNA adducts	Peluso et al. (2012)
Belgium	Placental DNA from 240 newborns	Global methylation	Decreased global placental methylation associated with increased $PM_{2.5}$	Janssen et al. (2013)

B[*a*]P, benzo[*a*]pyrene; LINE-1, long interspersed nuclear element-1; NO_2, nitrogen dioxide; PAHs, polycyclic aromatic hydrocarbons; 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; SO_4, sulfate; yr, year or years.

Table 4.16 Effects on leukocyte telomere length in humans exposed to outdoor air pollution

Country	Subjects	Results	Reference
China	120 office workers (controls) and 120 truck drivers (exposed) in Beijing	For both controls and exposed subjects, telomere length associated with personal $PM_{2.5}$ and elemental carbon, and outdoor PM_{10} on day of blood draw. However, decreased telomere length associated with PM_{10} averaged over 2 weeks before blood draw	Hou et al. (2012)
USA	165 subjects in Massachusetts	Decreased telomere length associated with carbon black in outdoor air	McCracken et al. (2010)
Italy	57 office workers (controls) and 77 traffic officers (exposed) in Milan	Decreased telomere length among traffic officers working in high vs low traffic intensity. Decreased telomere length associated with increasing concentrations of personal exposure to benzene and toluene	Hoxha et al. (2009)

PM_{10}, particulate matter with particles of aerodynamic diameter < 10 μm; $PM_{2.5}$, particulate matter with particles of aerodynamic diameter < 2.5 μm.

4.3 Other data relevant to carcinogenicity

4.3.1 Oxidative stress and inflammation

(a) Pulmonary oxidative stress and inflammation in humans

See Table 4.17.

Studies on oxidative stress in humans who have been exposed to air pollution have mainly applied biomarkers of oxidatively damaged lipids or inflammation markers in exhaled breath or bronchoalveolar lavage fluid (BALF). The publications are grouped into controlled exposure, panel, and cross-sectional studies.

Among the controlled exposure studies are some that have obtained direct measurements of pulmonary inflammation by analysis of BALF cell counts. The Working Group has included studies on bronchial instillation of PM in humans for the purpose of bridging observations in animal models on the same exposure, although it recognizes that extrapolation to real-life human exposures is challenging. Ghio & Devlin (2001) reported results from a study in which young and healthy people were exposed by bronchial instillation to aqueous extracts of PM_{10} collected before, during, and after a steel mill strike in Utah Valley, Utah, USA (Ghio & Devlin, 2001). The particles that were collected before and after the strike were associated with higher levels of neutrophils, pro-inflammatory cytokines (interleukin-1 beta [IL-1β], tumour necrosis factor [TNF], and IL-8), and protein (a marker of epithelial damage) in the BALF at 24 hours after the instillation of 500 μg of particle extract into the lungs of non-smoking volunteers. Extracts from periods when the steel mill was operating had high concentrations of metals, and the extracts generated ROS in acellular conditions, which was diminished by addition of DFO (a metal chelator) or dimethylthiourea (an antioxidant) (Ghio & Devlin, 2001). Schaumann et al. (2004) instilled into the lungs of 12 healthy volunteers $PM_{2.5}$ (100 μg, corresponding to 24 hours of inhalation of 100 μg/m^3) from two locations in Germany, characterized as being an area with mining and smelter industry and a non-polluted area. The instillation increased the total number of cells in BALF, whereas there was no difference in differential cell counts of neutrophils, lymphocytes, and monocytes. Nevertheless, it was only instillation of particles from a polluted area that was associated with increased concentrations of some pro-inflammatory cytokines in BALF (IL-6 and TNF, but not IL-1 and IL-8) and increased ex vivo ROS production in zymosan-stimulated BAL cells. There

Table 4.17 Oxidative stress and inflammation biomarkers in exhaled breath condensate from humans exposed to air pollution

Exposure	Exposure assessment	Sex, age, smoking, number	Biomarker	Effect	Reference
Instillation of aqueous extracts of CAPs from Utah Valley, Utah, USA	500 µg and lavage 24 h later	M, F 26 ± 2 yr NS *n* = 24	Cell count, IL-1β, TNF, and IL-8 in BALF	Increased number of neutrophils and BALF content of IL-1β, TNF, and IL-8	Ghio & Devlin (2001)
Intratracheal instillation of PM from an industrialized area and a non-industrialized area in Germany	100 µg	M, F 27 ± 3 yr NS *n* = 12	Cell count, IL-1, IL-6, IL-8, and TNF	Increased number of cells, IL-6, and TNF in BALF. Unaltered levels of IL-1 and IL-8	Schaumann et al. (2004)
Exposure to CAPs from Chapel Hill, North Carolina, USA	23–311 µg/m^3 for 2 h with intermittent, moderate exercise	M, F 26 ± 1 yr NS *n* = 38	Cell count, IL-6, IL-8, and PGE_2 in BALF	Increased number of neutrophils in BALF. Unaltered levels of IL-6, IL-8, and PGE_2	Ghio et al. (2000)
Controlled exposure to ultrafine CAPs from Chapel Hill, North Carolina, USA	PNC: 40 848–205 648/cm^3 Mass: 1.2–50 µg/m^3 for 2 h with intermittent, moderate exercise	M, F 18–35 yr NS *n* = 19	IL-6, IL-8, and PGE_2	Increased levels of IL-8 in BALF. Unaltered levels of IL-6 and PGE_2	Samet et al. (2009)
Controlled exposure to outdoor air while subjects exercising at locations with low and high traffic intensity in Pennsylvania, USA	7382–252 290 particles/cm^3 NO_2: < 100 ppb O_3: 41 ppb	M 21 ± 2 yr NS *n* = 12	Exhaled NO (chemiluminescence) and MDA (HPLC)	Unaltered levels of exhaled NO. Increased MDA in EBC after exercise at location with high exposure	Rundell et al. (2008)
Controlled exposure to CAPs from Edinburgh, United Kingdom	UFP: 0 or 99 400 UFP/cm^3 Mass: 190 µg/m^3 for 2 h in subjects with stable coronary heart disease and controls NO_x: 7.2 and 6.3 ppb SO_2: 0.13 and 0.13 ppb O_3: 5.0 and 6.0 ppb	M 54 ± 2 yr (controls) and 59 ± 2 yr NS *n* = 12, *n* = 12	8-isoprostane and 3-nitrotyrosine (ELISA)	Increased EBC by CAPs exposure at 6 h and 24 h. No effect on 3-nitrotyrosine	Mills et al. (2008)
Controlled exposure of people with asthma to air in a road tunnel (Stockholm, Sweden) or a laboratory	PNC: 1.3 × 10^5/mL $PM_{2.5}$: 80 µg/m^3 PM_{10}: 183 µg/m^3 NO_2: 265 µg/m^3 for 2 h	M, F 18–55 yr NS *n* = 14	Exhaled NO, cytokines in nasal lavage	Unaltered exhaled NO. Increased IL-10, TNF, and IL-12p70 and unaltered IL-1β, IL-6, and IL-8 in nasal lavage	Larsson et al. (2010)

Table 4.17 (continued)

Exposure	Exposure assessment	Sex, age, smoking, number	Biomarker	Effect	Reference
Controlled exposure of subjects in Utrecht, Netherlands, for 5 h at 2 traffic sites and 1 urban site	PNC: 66 500/cm^3 PM_{10}: 13 or 252 μg/m^3	M, F 19–26 yr NS *n* = 31	FeNO (chemiluminescence)	Association between PNC and FeNO	Strak et al. (2012)
Subjects living in Beijing, China, before, during, and after the Olympics in 2008	$PM_{2.5}$: 98.9–71.9 μg/m^3 NO_2: 25.6–14.6 ppb SO_2: 7.5–3.0 ppb O_3: 31.8–39.6 ppb	M, F 19–33 yr NS *n* = 125	MDA (HPLC), 8-isoprostane (ELISA), FeNO, and nitrate/nitrite	Lower levels of oxidative stress biomarkers and inflammation during the Olympics compared with samples before and after the event	Gong et al. (2013), Huang et al. (2012)
Panel study of children with asthma in Seattle, Washington, USA	$PM_{2.5}$: 13 μg/m^3 personal	M, F 6–13 yr NS *n* = 19	Exhaled NO (chemiluminescence)	Positive association between $PM_{2.5}$ levels and exhaled NO in children who did not take corticosteroid medication	Koenig et al. (2003, 2005), Mar et al. (2005)
Panel study of people with asthma in Ontario, Canada	$PM_{2.5}$: 2.7–14.3 μg/m^3 SO_2: 1.3–13.8 ppb NO_2: 12.3–27.0 ppb O_3: 7.5–21.0 ppm for 4 wk	M, F 9–14 yr NS *n* = 182	TBARS (fluorescence detection), 8-isoprostanes (immunoassay), and FeNO	Positive association between TBARS in EBC and SO_2, NO_2, and $PM_{2.5}$, but not with O_3. Concentration of 8-isoprostanes in EBC was only associated with SO_2 concentration. No association between air pollution and FeNO	Liu et al. (2009a)
Panel study of children with asthma in Mexico City, Mexico	$PM_{2.5}$: 4.2–89.5 μg/m^3 NO_2: 13.9–73.5 ppb O_3: 9.8–60.7 ppb	M, F 10 ± 2 yr NS *n* = 107	MDA (fluorescence detection). IL-4, IL-10, and TNF were below limit of detection	Positive associations between outdoor air pollution ($PM_{2.5}$ and O_3) levels and MDA in EBC	Romieu et al. (2008)
Panel study of elderly subjects with asthma or COPD in Seattle, Washington, USA	$PM_{2.5}$: 10.5 μg/m^3	M, F 60–86 yr NS *n* = 16	FeNO (chemiluminescence)	Association between outdoor $PM_{2.5}$ and FeNO in subjects with asthma. No association with personal PM exposure	Jansen et al. (2005)
Panel study of subjects with coronary artery disease in retirement communities in Los Angeles basin, California, USA, in warm or cold season	$PM_{2.5}$: 24 μg/m^3 $PM_{0.25}$: 10.3 μg/m^3 NO_2: 26 ppb O_3: 33 ppb	M, F 84 ± 6 yr NS *n* = 60	FeNO (chemiluminescence)	Association between FeNO and exposure markers ($PM_{2.5}$ and O_3)	Delfino et al. (2010a)
Panel study of elderly subjects in Steubenville, Ohio, USA	$PM_{2.5}$: 20 μg/m^3 NO_2: 11 ppb O_3: 15 ppb SO_2: 13 ppb	M, F 54–91 yr NS *n* = 29	FeNO (chemiluminescence)	Association between FeNO and $PM_{2.5}$ concentration, but not NO_2, O_3, or SO_2	Adamkiewicz et al. (2004)

Table 4.17 (continued)

Exposure	Exposure assessment	Sex, age, smoking, number	Biomarker	Effect	Reference
Panel study of schoolchildren in Mexico City, Mexico	$PM_{2.5}$: 29 µg/m³ NO_2: 37 ppb O_3: 32 ppm	M, F 8–12 yr NS *n* = 208	FeNO (chemiluminescence) and IL-8	Generally positive associations between exposure markers ($PM_{2.5}$, NO_2, and O_3) and biomarkers (FeNO and IL-8) in asthmatics and non-asthmatics, albeit statistically non-significant in some analyses	Barraza-Villarreal et al. (2008)
Panel study of students in Christchurch, New Zealand	PM_{10}: typically < 40 µg/m³ $PM_{2.5}$: 22% lower than PM_{10} 1-OHP (urine)	M 12–18 yr NS *n* = 93	H_2O_2 (fluorescent probe)	No association between air pollution and H_2O_2 in EBC	Epton et al. (2008)
Cross-sectional study of subjects with lung diseases in 4 European cities	$PM_{2.5}$: 9–28 µg/m³. Also measurements in or near homes for 18 mo	M, F 36–85 yr S/NS *n* = 133	NO_x (Griess), glutathione	Association between coarse particle level at central monitoring station and NO_x. No association with personal exposures (near or inside homes). Assay for detection of glutathione was not sufficiently sensitive	Manney et al. (2012)
Children in a suburban area (Bilthoven) or an urban area (Utrecht), Netherlands	Black smoke: 16–29 µg/m³ NO_2: 41–53 µg/m³ SO_2: 5–7 µg/m³ O_3: 21–28 mg/m³	M, F 8–13 yr NS *n* = 82	IL-8 (protein), NO_x in nasal lavage and exhaled NO (chemiluminescence)	Positive association between PM_{10} and exhaled NO. IL-8 and NO_x highest in nasal lavage from children in urban area	Steerenberg et al. (2001)

BALF, bronchoalveolar lavage fluid; CAPs, concentrated ambient particles; COPD, chronic obstructive pulmonary disease; EBC, exhaled breath condensate; ELISA, enzyme-linked immunosorbent assay; F, female; FeNO, fractional exhaled nitrogen oxide; h, hour or hours; H_2O_2, hydrogen peroxide; HPLC, high-performance liquid chromatography; IL, interleukin; M, male; MDA, malondialdehyde; mo, month or months; n, number; NO, nitrogen oxide; NO_2, nitrogen dioxide; NO_x, nitrogen oxides; NS, non-smokers; 1-OHP, 1-hydroxypyrene; O_3, ozone; PGE_2, prostaglandin E2; 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; PNC, particle number concentration; S, smokers; SO_2, sulfur dioxide; TBARS, thiobarbituric acid reactive substances; TNF, tumour necrosis factor; UFP, ultrafine particles; wk, week or weeks; yr, year or years.

were unaltered levels of markers of cell damage in the BALF (protein, albumin, and lactate dehydrogenase activity), and the glutathione concentration was unaltered (Schaumann et al., 2004). A study on inhalation exposure to concentrated outdoor particles (23–311 μg/m^3) from Chapel Hill, North Carolina, USA, for 2 hours with moderate exercise (exercise 15 minutes; rest 15 minutes) and analysis of pulmonary inflammation 18 hours after cessation of the exposure showed a mild increase in neutrophils in BALF, whereas there were unaltered levels of IL-6, IL-8, and prostaglandin E2 (PGE_2) (Ghio et al., 2000). Another study in 19 healthy non-smoking volunteers from Chapel Hill on outdoor UFP (40 848–205 648 UFP/cm^3; 1–50 μg/m^3) showed a moderate increase in the concentration of IL-8 in BALF, whereas there were unaltered levels of IL-6 and PGE_2 (Samet et al., 2009).

A non-invasive way to study the effect of air pollution is by analysis of markers of inflammation and oxidative stress in exhaled air. As this is a relatively easy method, it has been used in epidemiological studies as well as studies on controlled exposures to air pollution. It was shown that healthy young subjects had elevated levels of malondialdehyde (MDA) in exhaled breath condensate (EBC) after exercise at a location with high traffic-generated UFP (252 290 UFP/cm^3) compared with the same type of exercise at a location with less traffic (7382 particles/cm^3) (Rundell et al., 2008). This study also showed lower concentrations of exhaled NO and nitrate, which was hypothesized to be due to the formation of peroxynitrite (Rundell et al., 2008). Another study of controlled exposure to elderly men with stable coronary heart disease showed that inhalation of CAPs (190 μg/m^3) increased the concentration of 8-isoprostanes in EBC at 6 hours and 24 hours, whereas there was no effect on 3-nitrotyrosine levels (Mills et al., 2008). Exposure to air in a road tunnel in Stockholm, Sweden (80 μg/m^3 of $PM_{2.5}$ for 2 hours), had no effect on exhaled NO in people with asthma, and there were inconsistent associations between exposure and levels of IL-10, IL-1β, IL-6, IL-8, and TNF in nasal lavage fluid (Larsson et al., 2010). Strak et al. (2012) studied subjects who were exposed to outdoor air at a traffic site or an urban site (5 hours, with intermittent exercise); they showed associations between the particle number concentration and fractional exhaled NO (FeNO).

The Beijing Olympics in 2008 has formed the basis for studies on the association between improvements in outdoor air quality and biomarkers of oxidative stress and inflammation. A study in 125 healthy adults showed that the EBC content of NO, nitrates, nitrites, MDA, and 8-isoprostanes (using an ELISA method) was lower in young and healthy subjects during the Olympics compared with periods before and after the Olympics (Gong et al., 2013; Huang et al., 2012).

Panel studies have typically focused on subjects with lung or cardiovascular diseases. It was shown that there was a positive association between personal $PM_{2.5}$ exposure and exhaled NO in children (aged 9–13 years) with asthma from Seattle, Washington, USA, who did not take corticosteroid medication (Koenig et al., 2003, 2005; Mar et al., 2005). Another panel study in Ontario, Canada, showed no association between air pollution exposure and FeNO; there was a positive association between levels of air pollution exposure components ($PM_{2.5}$, NO_2, and SO_2) and thiobarbituric acid reactive substances (TBARS; an oxidative stress marker) in EBC, which was not a particularly reliable assay for detection of lipid peroxidation products (Liu et al., 2009a). Positive associations between levels of $PM_{2.5}$ and ozone, based on stationary monitoring data, and MDA levels in EBC were observed in children with asthma in Mexico City (Romieu et al., 2008). A study in elderly subjects with asthma or COPD in Seattle, Washington, USA, showed an association between outdoor concentrations of $PM_{2.5}$ and FeNO, whereas there was no association

between FeNO and personal exposure to $PM_{2.5}$ (Jansen et al., 2005). In addition, elderly subjects with coronary artery disease in the Los Angeles basin, California, USA had a positive association between exposure markers ($PM_{2.5}$ and ozone) and FeNO (Delfino et al., 2010a). Studies of healthy children in Steubenville, Ohio, USA, or Mexico City also indicated positive associations between air pollution exposure and FeNO (Adamkiewicz et al., 2004; Barraza-Villarreal et al., 2008), whereas there was no association between levels of hydrogen peroxide (H_2O_2) in EBC and air pollution exposure among students in Christchurch, New Zealand (Epton et al., 2008).

A cross-sectional study of subjects with lung diseases (asthma or COPD) in four European cities showed an association between levels of coarse particles at a central monitoring station and levels of NO_x in EBC, whereas there was no association with personal exposure to $PM_{2.5}$, PM_{10}, or coarse fraction as measured either near or inside the homes (Manney et al., 2012). Another cross-sectional study on children in the Netherlands showed a positive association between PM_{10} exposure and exhaled NO; children from an urban area had higher nasal lavage levels of IL-8 and NO_x compared with children from a suburban area (Steerenberg et al., 2001).

(b) Systemic effects of inflammation in humans

The studies on systemic inflammation have mainly centred on markers of cardiovascular diseases, including acute-phase proteins (fibrinogen and C-reactive protein [CRP]), platelets, von Willebrand factor, haematocrit, whole blood viscosity, and leukocyte counts (Delfino et al., 2005). The measurement of CRP especially has been popular because it is used clinically, it can increase by more than 3 orders of magnitude during an acute-phase response, and it has a relatively short half-life in plasma (~19 hours). A recent review of the association between air pollution levels and CRP levels in humans encompassed a total of 44 publications, stratified into cross-sectional, panel, and randomized cross-over trials (Li et al., 2012). The most important conclusions from that survey were that there was an association between air pollution exposure and elevated levels of CRP in children in cross-sectional studies, whereas there were inconsistent results in adults, which might be related to the inclusion of subjects with prescribed statins or anti-inflammatory drugs. It was also noted that the randomized cross-over trials mainly showed no association between air pollution exposure and CRP levels in plasma, which could be because these studies had few subjects and relatively short exposure duration (Li et al., 2012). One of the studies on controlled exposure used relatively high concentrations of CAPs (190 μg/m^3 for 2 hours) and found no change in serum levels of CRP and total leukocyte counts, although there was a transient and marginal increase in the number of monocytes in blood (Mills et al., 2008). In addition, two studies on indoor air filtration for 24–48 hours with relatively low exposure gradients of traffic-generated emissions in Copenhagen, Denmark, showed no effect on levels of CRP, IL-6, TNF, and fibrinogen (Bräuner et al., 2008a, b). A 7-day intervention period with air filtration in homes of a wood smoke-affected area in British Columbia, Canada, found an association between the indoor concentration of fine particles and CRP levels, whereas there was no association with IL-6 levels (Allen et al., 2011).

The production of CRP is regulated in response to elevated levels of IL-6, IL-1, and TNF. A study indicated that children in Mexico City, compared with children in a low-pollution city, had a systemic pro-inflammatory state as determined by elevated plasma/serum levels of TNF, IL-1β, PGE_2, and CRP (Calderón-Garcidueñas et al., 2008). A very large study of subjects in Lausanne, Switzerland, with 6183 adult participants showed associations between short-term

exposures to PM_{10} and elevated levels of IL-1β, IL-6, and TNF, whereas there was no effect on CRP levels (Tsai et al., 2012). Other studies of people with coronary artery disease also found associations between exposure to small particles ($PM_{0.25}$) and levels of CRP, IL-6, and TNF in plasma (Delfino et al., 2008, 2009, 2010b). A study in Singapore showed that subjects had elevated serum levels of TNF, IL-1β, and IL-6 during a period with haze, with high outdoor air pollution concentrations (PM_{10}, 125 μg/m³), compared with a period afterwards with a low air pollution level (PM_{10}, 14 μg/m³) (van Eeden et al., 2001). However, there are also studies showing inconsistent associations between air pollution exposure and levels of CRP, IL-6, IL-8, serum amyloid A, and fibrinogen (Huttunen et al., 2012; Rückerl et al., 2006, 2007; Strak et al., 2013; Wu et al., 2012) or no association with levels of CRP, IL-6, IL-10, TNF, and fibrinogen (Liu et al., 2007, 2009b; Zuurbier et al., 2011).

Studies on oxidative stress biomarkers in biomonitoring include oxidation products of lipids, proteins, and DNA. Several studies of these products from areas in the Czech Republic have shown positive associations between air pollution exposure and oxidative stress markers (Bagryantseva et al., 2010; Rossner et al., 2007, 2011c, 2013b).

(c) Pulmonary inflammation and ROS production in experimental systems

(i) Pulmonary inflammation in experimental animals

Numerous studies have assessed pulmonary inflammation in animals after exposure to urban air particles (see Supplemental Table S20, available online). Notably, there is a clear effect on pulmonary inflammation after both inhalation and instillation exposure. This is observed by an increased number of leukocytes in BALF or elevated concentrations of pro-inflammatory cytokines. Increased inflammation has been observed after inhalation of CAPs from Bilthoven (Netherlands), Boston (Massachusetts, USA), Tuxedo (New York, USA), and Manhattan (New York, USA), with concentration ranges of approximately 100–1200 μg/m³ (Cassee et al., 2005; Clarke et al., 1999, 2000a, b; Gordon et al., 1998; Rhoden et al., 2004; Saldiva et al., 2002; Shukla et al., 2000).

Studies in mice intratracheally instilled with EHC93 (1 mg/mouse) have shown that the soluble fraction of the total dust sample was more inflammogenic than the insoluble fraction (Adamson et al., 1999a). Especially the content of zinc was found to be an important contributor to the inflammogenicity, although redox-active transition metals also had some effect on the inflammation response after intratracheal instillation of EHC93 (1 mg/mouse) (Adamson et al., 2000). The intratracheal instillation of SRM 1648 (1.6 mg/lung) in mice has also been associated with pulmonary inflammation in terms of increased concentrations of IL-6, TNF, and MIP2 in BALF (Becher et al., 2007).

Another study of PM from Duisburg (Germany), Prague (Czech Republic), Amsterdam (Netherlands), Helsinki (Finland), Barcelona (Spain), and Athens (Greece) showed that the coarse particles were more inflammogenic than the fine particles by intratracheal instillation (1–10 mg/kg for 4, 12, or 24 hours) in mice (Happo et al., 2007). Other studies also have indicated that coarse particles instilled intratracheally (100 μg/mouse) were more inflammogenic than fine particles (Farina et al., 2011). In addition to the potency of particles with different sizes, the studies in Europe have also revealed both temporal and spatial variation in the potency of PM (Farina et al., 2011; Happo et al., 2007).

Other studies on differences between particles have shown that desert dust from Arizona, USA, or Shapotou, China, was associated with pulmonary inflammation after instillation of 0.1 mg/mouse 4 times over 8 weeks (Ichinose et al., 2008). Instillation of 32–100 μg/mouse of

residual oil fly ash (ROFA) or SRM 1649 was associated with a higher degree of pulmonary inflammation than the same dose of World Trade Center fine particles (Gavett et al., 2003).

A few studies have used intranasal instillation, showing that $PM_{2.5}$ from the air in São Paulo (Brazil), PM from Buenos Aires (Argentina), or CAPs from Boston (Massachusetts, USA) increased the level of pulmonary inflammation (Martin et al., 2007; Martin et al., 2010; Riva et al., 2011; Sigaud et al., 2007).

Experimental studies also indicate that the pulmonary inflammation depends on oxidative stress, as indicated by a study where mice with overexpression of SOD compared with wild-type counterparts had lower levels of neutrophils, TNF, and MIP2 in BALF after an intratracheal instillation of 50 µg/mouse (Ghio et al., 2002). Similarly, pre-treatment with antioxidants decreased the level of particle-mediated pulmonary inflammation after the intratracheal instillation (10–100 µg/mouse) of urban air PM (Dick et al., 2003).

A study in Porto Alegre, Brazil, with relatively high outdoor particle concentrations (110–140 µg/m^3), showed increased pulmonary inflammation in rats (Pereira et al., 2007). However, it has also been reported that exposure to CAPs from Grand Rapids, Michigan, USA (493 µg/m^3, 8 hours/day for 13 days), Chapel Hill, North Carolina, USA (475–907 µg/m^3, 6 hours/day for 2–3 days), or Bilthoven, Netherlands (399–3612 µg/m^3, 6 hours/day for 2 days) was not associated with pulmonary inflammation in rats (Heidenfelder et al., 2009; Kodavanti et al., 2000; Kooter et al., 2006). The exposure concentrations do not appear to be different between the studies with null effect and studies that have shown pulmonary inflammation. A study on short-term inhalation exposure to EHC93 (57 µg/m^3 for 4 hours) was associated with an increased number of neutrophils in the air space and tissue of rats (Adamson et al., 1999b). Likewise, there was an increased level of pulmonary inflammation after inhalation (12 mg/m^3 for 6 hours) of ROFA or the corresponding dose administered by intratracheal instillation (110 µg/rat) (Costa et al., 2006). The effect on pulmonary inflammation was highest at 24 hours after the exposure, and it decreased gradually over the next 72 hours (Costa et al., 2006).

Relatively high bolus dose instillations of EHC93 (5–10 mg/kg) have indicated a bell-shaped response in regard to the number of cells in BALF, with the highest effect at 24–48 hours, whereas time points before (4 hours) and after (days 4–7) indicated lower effects on pulmonary inflammation (Bagate et al., 2004; Gerlofs-Nijland et al., 2005; Ulrich et al., 2002). Intratracheal instillation of the water-soluble fraction of TSP from Provo, Utah, USA, was associated with higher levels of neutrophils in BALF compared with instillation of the insoluble fraction of particles (Ghio et al., 1999).

A study of particles that were collected in Amsterdam (Netherlands), Lodz (Poland), Oslo (Norway), and Rome (Italy) showed that fine particles were more inflammogenic than coarse particles on a mass basis in rats by intratracheal instillation (Halatek et al., 2011). This study also revealed seasonal variability of particles for the influx of neutrophils in BALF, especially in Oslo, whereas the levels of TNF and MIP2 in BALF depended on both the season and the location (Halatek et al., 2011). However, another study of intratracheal instillation in spontaneously hypersensitive rats of particles (3 mg/kg or 10 mg/kg) collected in Munich (Germany), Hendrik-Ido-Ambacht (Netherlands), Dordrecht (Netherlands), Rome (Italy), and Lycksele (Sweden) showed that coarse particles were more inflammogenic than fine particles (Gerlofs-Nijland et al., 2007). Coarse particles were shown to be more inflammogenic than fine particles by intratracheal instillation in rats (Schins et al., 2004). Also, both temporal and spatial variation in the potency of PM has been shown (Gerlofs-Nijland et al., 2007; Halatek et al., 2011). A study

in Beijing, China, showed higher levels of TNF, IL-6, and IL-1 in lung homogenate after intratracheal instillation of $PM_{2.5}$ compared with the same dose of PM_{10} (7.5 mg/kg), and particles collected closest to traffic generated the highest level of inflammation (Zhang et al., 2011). Intratracheal instillation of ROFA (500 µg/rat) was associated with increased levels of neutrophils in BALF as well as elevated levels of IL-6, TNF, CCL2, and IL-1β (Roberts et al., 2003).

Collectively, there is compelling evidence for an association between exposure to air pollution particles and pulmonary inflammation in experimental animals.

(ii) ROS production in experimental animals

Relatively few studies have undertaken analysis of ROS production in vivo after pulmonary exposure. Inhalation of CAPs (300 µg/m³ for 5 hours) or ROFA (1.7 mg/m³ for 30 minutes) was associated with increased production of ROS in lung tissue, assessed by chemiluminescence (Gurgueira et al., 2002). Another study exposed rats to oil fly ash (500 µg/rat) by intratracheal instillation and subsequently injected 4-POBN (a spin trap) in the peritoneum at 1 hour before the rats were killed. Lung homogenates from exposed rats showed the presence of carbon-centred alkyl radicals, which were suspected to have been derived from peroxidation of lipids (Kadiiska et al., 2004).

(iii) Markers of inflammation in cultured cells

Studies on markers of inflammatory responses in cultured cells, summarized in Supplemental Table S21 (available online), evaluated a variety of cytokines. A substantial number of studies have documented increased levels of biomarkers of inflammation, in regard to cytokines, chemokines, and production of NO in various cell lines or primary cell cultures from rodents after exposure to authentic air pollution particles or model particles such as EHC93, SRM 1648, SRM 1649, or ROFA (Auger et al., 2006; Baulig et al., 2003; Becher et al., 2007; Brown et al., 2004, 2007; Fujii et al., 2001, 2002; Garçon et al., 2006; Jalava et al., 2005; Karlsson et al., 2006; Schneider et al., 2005; van Eeden et al., 2001; Watterson et al., 2007). Exposure of lung epithelial cells and alveolar macrophages in co-cultures to ROFA or SRM 1649 increased the secretion of MIP2 and TNF. This effect was not observed in lung epithelial cells or alveolar macrophages in mono-cultures (Tao & Kobzik, 2002). Higher levels of IL-6, IL-8, and IL-1β were also observed in A549/THP-1 in co-cultures compared with THP-1 mono-cultures after exposure to PM samples from Milan, Italy (Longhin et al., 2013).

Collectively, there is compelling evidence that exposure to PM in cultured cells is associated with inflammatory reactions, assessed mainly as secretion of cytokines and chemokines, which may be elicited secondary to oxidative stress in the cells. This association between exposure to PM and secretion of cytokines is observed especially in lung epithelial cells and macrophages. The inflammation potential seems to be higher for coarse particles compared with fine particles, which is likely to be related to the content of endotoxin in the coarse fraction. There is also some experimental evidence linking the inflammation reaction in cultured cells to oxidative stress and metal-catalysed ROS production, although it should be noted that the observations are mixed and the linkage between oxidative stress and inflammation might depend on both the physical–chemical properties of PM samples and their constituents.

(iv) ROS production in cultured cells

Studies on ROS production in cultured cells are summarized in Supplemental Table S22 (available online). It has been shown that exposure to air pollution particles was associated with intracellular ROS production, detected as oxidation products of 2′,7′-dichlorodihydrofluorescein (DCFH) or dihydroethidium, or chemiluminescence in different human cells (Auger

et al., 2006; Baulig et al., 2003; Becker et al., 1996, 2005; Goldsmith et al., 1997; Kamdar et al., 2008; Karlsson et al., 2008; Ohyama et al., 2007; Shukla et al., 2000; Yi et al., 2014; Zhang et al., 2008). Model particles such as SRM 1648, SRM 1649, EHC93, and various types of fly ashes have also been associated with intracellular ROS production (Baulig et al., 2003; Becher et al., 2007, Becker et al., 1996, 2005; Di Pietro et al., 2009; Dwivedi et al., 2012; Li et al., 2006; Schneider et al., 2005).

A few studies have assessed the ROS production potential of EOM in cultured cells. This revealed increased ROS production by extracts from both urban and rural sites in MCF-7 cells (Chen et al., 2013), PM_{10} from an industrial area in HepG2 cells (Jiang et al., 2011), and road tunnel particles in A549 cells (Shang et al., 2013).

Collectively, there is compelling evidence for intracellular ROS production in cells exposed to PM.

(v) Acellular ROS production

Studies on acellular ROS production are summarized in Supplemental Table S23 (available online). ROS can be detected by electron spin resonance (ESR) signals, which are typically obtained in experimental conditions with H_2O_2 as co-oxidant and 5,5-dimethyl-1-pyrroline-*N*-oxide (DMPO) as spin trap, indicating that the assay depends mainly on the presence of transition metals in the samples (Knaapen et al., 2002; Valavanidis et al., 2000, 2005). This is supported by observations that coal fly ash produced ROS, which correlated with the release of iron (Dwivedi et al., 2012; van Maanen et al., 1999). Nevertheless, other transition metals can be the dominant source of ROS production, which has been observed for $PM_{2.5}$ samples that were collected in the San Joaquin Valley, California, USA (Shen et al., 2011; Shen & Anastasio 2011, 2012). In addition, treatment with DFO, catalase, or antioxidants (DMSO or dimethylthiourea) diminished the ROS production of air pollution particles measured by ESR or other assays, such as oxidation products of DCFH or deoxyribose (Ball et al., 2000; Frampton et al., 1999; Ghio & Devlin, 2001; Imrich et al., 2007; Knaapen et al., 2002; Lindbom et al., 2007).

Collectively, there is strong evidence that PM generates ROS in suspensions by at least two mechanisms, encompassing transition metal catalysis or redox cycling by quinones.

In summary, controlled short-term inhalation exposures to CAPs or instillation of PM from especially urban areas in Europe and the USA have been associated with increased levels of pulmonary inflammation and oxidative stress. These observations are supported by results from cross-sectional and panel studies that show signs of pulmonary inflammation, whereas there are only few studies on oxidative stress end-points. There is strong evidence for pulmonary inflammation in animals after either inhalation or intratracheal instillation (or similar ways of exposure) of PM from urban air in Argentina, Brazil, China, Europe, and the USA. Aqueous suspensions of PM from urban areas in China, Europe, Japan, and the USA have been associated with increased ROS production in cultured cells. Organic extracts of PM from cities in China have likewise increased the intracellular ROS production. Aqueous suspensions of PM from urban areas, mainly in Europe and the USA, have promoted inflammation reactions in cultured macrophages or airway epithelial cells. A few studies on PM from China, Mexico, and Senegal also indicate inflammation in cultured cells. In comparison, very few studies have assessed the effect of organic extracts of PM on inflammation, and the results have been mixed. Aqueous suspensions of PM from urban areas, mainly in Europe and the USA, have been shown to generate ROS production in acellular conditions.

4.3.2 Non-cancer effects

Exposure to current-day concentrations of outdoor air pollution has been linked with a variety of non-cancer health effects ranging in severity from subclinical physiological changes to mortality, particularly involving cardiovascular and respiratory diseases, with evidence of additional effects on immunological, reproductive, and other systems (American Thoracic Society, 2000; WHO, 2006; Samet & Krewski, 2007) (see Supplemental Figure S3, available online) Exposure to outdoor particulate air pollution has been estimated to have contributed 3.2 million premature deaths and 74.4 million lost years of healthy life worldwide in 2010, due to cardiovascular disease, COPD, and acute lower respiratory infections, in addition to lung cancer (Lim et al., 2012). Although mortality and hospitalization have been the most studied effects of air pollution and have important public health impacts, the number of people affected by less-severe effects is larger (WHO, 2006).

(a) Cardiorespiratory effects

The cardiovascular and respiratory effects of outdoor air pollution have been examined in many studies worldwide using diverse research designs and are summarized in numerous reviews and regulatory documents (e.g. Brook, 2008; Brook et al., 2010; EPA, 2006; Hoek et al., 2013; Lai et al., 2013; WHO, 2006). The effects of airborne PM have been most extensively studied. Exposure to PM is linked with increases in all-cause, cardiovascular, and respiratory mortality, as well as with other cardiovascular and respiratory effects, including hospitalization for acute respiratory events, decreased lung function, ischaemic events, stroke, arrhythmia, and reduced heart rate variability (Brook, 2008; Hoek et al., 2013; Pieters et al., 2012; Samet & Krewski, 2007; WHO, 2006). Positive associations have also been observed between exposure to NO_x and mortality from all causes and ischaemic health disease (Hoek et al., 2013; Mustafic et al., 2012). However, because NO_2 is closely correlated with other air pollutants from traffic-related sources, it is difficult to determine whether the effects are due specifically to NO_2, to other air pollutants, or to the complex mixture of pollutants (WHO, 2006).

(b) Reproductive effects

Prenatal exposure to air pollution has been hypothesized to affect the unborn child through several mechanisms, including oxidative stress, inflammatory processes, endocrine disruption, and germ-cell changes (Schwartz, 2004; Slama et al., 2008). Research on this topic is still inconclusive, but there is some evidence that prenatal exposure to outdoor air pollution increases the risk of preterm delivery, fetal growth deficit, and cardiac birth deficit (Wigle et al., 2008). A meta-analysis based on 62 studies estimated that exposure to PM, NO_2, and CO during pregnancy was associated with low birth weight and that exposure to PM and CO was associated with preterm birth (Stieb et al., 2012). Another systematic review of air pollution exposures and birth outcomes reported similar conclusions regarding low birth weight and preterm birth, as well as associations of small-for-gestational-age births with prenatal exposure to PM and of preterm birth with exposure to SO_2 (Shah & Balkhair, 2011). Transcriptomics allows for more mechanistic and holistic studies by analysing several genes that are upregulated or downregulated in exposed populations, for example as performed in the well-characterized Czech populations, including gene profiling of newborns (Šrám et al., 2013). These studies are explorative.

(c) Immunotoxic effects

Associations between exposures to outdoor air pollution and biomarkers of immunotoxicity associated with inflammatory responses, for example cytokines, have been reported in several studies.

(d) Endocrine effects

Statistically significant associations between long-term exposure to traffic-related air pollution at the residence and diabetes mortality were reported in a Danish follow-up study of 52 061 cohort participants (Raaschou-Nielsen et al., 2013a). An association of type 2 diabetes in women with traffic-related air pollution measured by NO_2 has also been reported (Brook et al., 2008). Urinary excretion of 17-ketosteroids and 17-hydroxycorticosteroids, markers of adrenal cortex functions, was reported to be significantly lower in male children living in polluted areas compared with children living in clean areas (Watanabe, 2000), but the paper lacks important details about the study population.

4.3.3 Genotoxic and other deleterious effects on germ cells

Several lines of evidence have suggested that air pollution may cause deleterious effects to germ cells in wildlife, experimental animals, and humans (reviewed in Samet et al., 2004; Somers & Cooper, 2009; Somers, 2011). The studies of outdoor air pollution-induced genetic effects in germ cells are summarized in Table 4.18 and Supplemental Table S24 (available online).

(a) DNA damage and chromosomal aberrations in human sperm

See Table 4.18.

Several studies in the Czech Republic have evaluated the association of outdoor air pollution exposure with chromatin damage in humans by SCSA. A preliminary study showed an increased percentage of sperm with abnormal chromatin structure (denatured DNA susceptibility) (expressed as $COMP\alpha_t$, cells outside the main population of cells) at periods of high air pollution in Teplice (Selevan et al., 2000). High $COMP\alpha_t$ (> 30) has been associated with infertility and spontaneous abortion (Evenson et al., 1999). A subsequent 2-year follow-up study confirmed that there was a significant association between exposure to periods of air pollution and the increased DNA damage in human sperm; however, there was no association between aneuploidy in sperm and outdoor air pollution (Rubes et al., 2005). Rubes et al. also showed that men with the *GSTM1* null genotype exhibited increased susceptibility to sperm DNA damage associated with exposure (Rubes et al., 2007). Furthermore, significantly higher sperm DNA fragmentation index (DFI) values were observed in the winter compared with in the spring for police officers working outdoors in Prague (Rubes et al., 2010). A study in male traffic police in Prague showed that they had higher frequencies of aneuploidy in their sperm when sampled in January, when the PM_{10} concentrations were high, compared with in March, when the PM_{10} concentrations were low (Rubes et al., 1996; Srám et al., 1999).

(b) Mutations in male germ cells and the germline in animals

Studies of sentinel wildlife and laboratory rodents exposed to outdoor air (summarized in Supplemental Table S24, available online) have provided evidence for elevated germline mutation induced by air pollution (Somers & Cooper, 2009; Somers, 2011). Increased rates of germline mutations at minisatellite loci (tandem repetitive DNA loci) were seen in herring gulls (*Larus argentatus*) collected from industrial areas with high levels of air pollution, and minisatellite mutation rates decreased with increasing distance from the industrial coking oven and urbanization site (Yauk & Quinn, 1996; Yauk et al., 2000). When laboratory mice were exposed to outdoor air in a polluted industrial area near steel mills, a significant 1.5–2.0-fold elevation in heritable mutation frequency in the offspring was observed, primarily through ESTR mutation events in the paternal germline (Somers et al., 2002). This heritable mutation frequency was significantly reduced when mice were exposed in situ to air from a polluted area treated by a HEPA filtration

Table 4.18 Genotoxic effects in germ cells of humans exposed to outdoor air pollution

Location	Subjects	Experimental design/exposure	End-point	Results		Reference
Teplice, Czech Republic	Young men (18 yr)	Semen samples were collected from 272 men recruited from Teplice (industrialized area) or Prachatice (rural area)	Abnormal chromatin in sperm (SCSA)	Increased percentage of sperm with abnormal chromatin was associated with seasonally elevated levels of air pollution	+	Selevan et al. (2000)
Teplice, Czech Republic	Young men (19–21 yr)	228 semen samples were collected from 36 men in different seasons over 2 years	Sperm DFI by SCSA Aneuploidy	Increased DFI in sperm was associated with high levels of air pollution. No effect was observed for aneuploidy	+	Rubes et al. (2005)
Teplice, Czech Republic	Young men (19–21 yr)	Semen samples were collected from 35 men in different seasons	Sperm DFI by SCSA among *GSTM1* null	Statistically significant association between *GSTM1* null genotype and increased SCSA-defined DFI.	+	Rubes et al. (2007)
Prague, Czech Republic	Male outdoor police officers (33 ± 5 yr)	Semen samples were collected from 47 police officers in winter (high pollution) and spring (low pollution)	DNA damage in sperm (DFI by SCSA)	Sperm DFI was significantly higher in winter samples than in spring samples for all police and for non-smokers	+	Rubes et al. (2010)
Prague, Czech Republic	Male outdoor police officers	Outdoor police officers in Prague were sampled in winter (high pollution) and spring (low pollution)	Aneuploidy for YY8 (FISH)	YY8 aneuploidy was significantly associated with the season of heaviest air pollution	+	Rubes et al. (1996); Srám et al. (1999)

+, positive; DFI, DNA fragmentation index; FISH, fluorescence in situ hybridization; SCSA, sperm chromatin structure assay; yr, year or years.

system that removed 99.97% of particles 0.3 μm in diameter (Somers et al., 2004). [The Working Group noted that airborne PM was an important factor in induction of germline ESTR mutations.] Taken together, the series of studies indicate that exposure to outdoor air pollution could cause damage to male gametes, as shown in observed germline mutations and DNA damage in sperm of mice, although their contributions to fertility and reproduction are still unknown.

(c) Sperm abnormality in animals

In a sperm morphology study of mice treated with extracts of air samples collected in Shanghai, China, an increased frequency of germ-cell deformations was observed for most sites in the winter (Mao et al., 1993). A subsequent animal experimental study in Taiyuan, China, showed that intraperitoneal treatment of male Kunming mice with particle extracts from the residential area downwind of a coal combustion power plant induced sperm abnormality, CAs of spermatogonia and primary spermatocytes, and MN in spermatids (Sun et al., 1995). Elevated frequencies of head and tail deformities in the sperm were observed in feral mice living in an area with air highly polluted by automobile traffic in Rome, Italy (Ieradi et al., 1996).

In summary, a series of studies in humans, wildlife, and experimental animals indicate that outdoor air pollution might cause heritable mutations, sperm abnormalities, and germ-cell DNA damage. The evidence for heritable mutation derives from studies examining gulls as well as inbred and outbred mice.

4.3.4 Oncogenic cell transformation

Studies that used cultured animal cells to assess the ability of outdoor air to induce malignant cell transformation are summarized in Supplemental Table S25 (available online).

Several such studies have demonstrated that organic extracts of urban air PM can induce oncogenic transformation of cultured animal cells. Moreover, some studies have demonstrated that the resultant transformed cells can form malignant tumours in vivo.

4.4 Susceptible populations

4.4.1 Polymorphisms in carcinogen-metabolizing genes

Studies on the influence of genotype and lung cancer risk, and of genotype and biomarkers, are summarized in Supplemental Tables S26 and S27 (available online), respectively.

Data on the roles of genotypes/phenotypes related to outdoor air exposure are sparse. Hosgood et al. (2007) carried out a meta-analysis of six studies evaluating the associations of *GSTM1* null, *GSTT1* null, and *GSTP1* (105 Val polymorphism) genotypes and their association with risk of lung cancer in regions where exposure to indoor burning of coal, wood, and biomass fuels is common. The risk of lung cancer was elevated for carriers of the *GSTM1* null and *GSTT1* null genotypes, with the strongest association observed for *GSTM1* null carriers in four Asian regions where coal smoke is used for heating and cooking. However, individual data on exposure to air pollution were not available.

4.4.2 Age and sex

The potentially different effects of exposure early in life and greater life expectancy from childhood could influence the risk of cancer associated with air pollution. Indeed, children have different susceptibilities owing to their dynamic growth and developmental processes as well as physiological, metabolic, and behavioural differences. From conception through adolescence, rapid growth and developmental processes occur that can be disrupted by exposures to environmental chemicals. These include anatomical, physiological, metabolic, functional, toxicokinetic, and toxicodynamic processes (IPCS, 2008). Exposure pathways and exposure patterns may also be different in different stages of childhood. Children have a higher inhalation rate and a higher ratio of body surface area to body weight, which may lead to increased exposures. Children's metabolic pathways may differ from those of adults. Children have more years of future life and thus more time to develop chronic diseases that take decades to appear and that may be triggered by early environmental exposures (IPCS, 2008). However, aside from the studies of childhood cancer reviewed in Section 2.5, the Working Group found no specific studies of susceptibility in relation to age at exposure.

Men and women differ in body size, conductive airway size, and ventilatory parameters; therefore, sex differences in deposition might be expected. Since women are generally smaller than men, the increased minute ventilation of women compared with their normal ventilation could affect deposition patterns. This may help explain why sex-related effects on deposition have been observed in some studies.

Kim & Hu (1998) assessed the regional deposition patterns of particles of 1 μm, 3 μm, and 5 μm mass median aerodynamic diameter in healthy adult men and women using controlled breathing. The total fractional deposition in the lungs was similar in men and women for the 1 μm particles but was greater in women for the 3 μm and 5 μm particles. Deposition also appeared to be more localized in the lungs of women compared with those of men. Jaques & Kim (2000) measured deposition in healthy adults using sizes in the ultrafine mode (0.04–0.1 μm). Total fractional lung deposition was greater in women than in men for 0.04 μm and 0.06 μm particles. The region of peak fractional deposition was shifted closer to the mouth, and the peak height was slightly greater for women than for men for all exposure conditions. These

differences were generally attributed to the smaller size of the upper airways, particularly of the laryngeal structure, in women. In another study (Bennett et al., 1996), the total respiratory tract deposition of 2 µm particles was examined in adult men and women aged 18–80 years who breathed with a normal resting pattern. There was a tendency for greater deposition fractions in women than in men. However, since men had greater minute ventilation, the deposition rate (i.e. deposition per unit time) was greater in men than in women. More recently, Bennett & Zeman (2004) found no difference in the deposition of 2 µm particles in boys versus girls aged 6–13 years (n = 36) (EPA, 2009).

4.4.3 Socioeconomic status

Social inequalities in risk factors may account for more than half of the inequalities in outcomes of major noncommunicable diseases, including lung cancer (Di Cesare et al., 2013). Social inequalities in exposure to air pollution have been documented and are discussed in Section 1.4.3. Research into whether the effects of air pollution also differ by socioeconomic status has been focused largely on general mortality and cardiorespiratory diseases (O'Neill et al. 2003). However, several studies of cancer and air pollution have considered socioeconomic status as a potential confounder or effect modifier.

A cohort study in Rome, Italy, reported increased relative risks for the association of NO_2 and $PM_{2.5}$ and lung cancer with lower levels of education and an area-based socioeconomic index, but the trends were not statistically significant (Cesaroni et al., 2013). Several other studies of air pollution and lung cancer in Europe and North America adjusted for individual or aggregate indicators of socioeconomic status in addition to other covariates, but did not report results according to the levels of socioeconomic indicators (e.g. Jerrett et al., 2013; Carey et al. 2013; Raaschou-Nielsen et al. 2013b; Krewski et al., 2009; Beelen et al., 2008a). In general, the study results were not notably changed by adjustment for socioeconomic indicators.

4.4.4 Increased risk in diseased populations

Several studies have shown that non-malignant respiratory disease, particularly COPD, increases the risk of developing lung cancer (e.g. Skillrud et al., 1986; Turner et al., 2007). Abnormal regulation of the immune system and chronic inflammation appear to be key events in this process. In addition, the possibility of a genetic basis for lung cancer susceptibility in the context of COPD is becoming clear (Rooney & Sethi, 2011). Although there is evidence that comorbid conditions increase the risk of acute mortality associated with exposure to air pollution, the role of comorbidity has not been investigated in studies of air pollution and cancer.

4.5 Mechanistic considerations

Outdoor air pollution consists of a mixture of complex components, including diesel and gasoline engine exhausts, biomass combustion emissions, geological dust, and industrial emissions. The complexity of this "mixture of mixtures" contributes to the complexity of the mechanisms underlying the genetic and related effects reported in humans and experimental systems. The compiled evidence supports the operation of multiple mechanisms that involve DNA damage (e.g. formation of bulky adducts, strand breaks, oxidatively damaged DNA, and induction or alteration of DNA repair pathways), cytogenetic damage (e.g. chromosome breaks and aneuploidy), somatic- and germ-cell mutation, oncogenic cell transformation, epigenetic changes, changes in gene expression, and induction of oxidative stress and inflammation.

Table 4.19 Summary of genetic effects reported in molecular epidemiology studies of outdoor air pollution

Exposure scenario	End-point reported			
	DNA adducts[a]	Cytogenetic damage[b]	DNA strand breaks[c]	Changes in gene expression[d]
Traffic police	+	+	+	NA
Taxi drivers	+	+	+	NA
Bus drivers	+	+	NA	NA
Mail carriers	+	+	NA	NA
Urban residents	+	+	+	+
Urban children	+	+	+	+

[a] Refer to Section 4.2.3.
[b] Refer to Section 4.2.2.
[c] Refer to Section 4.2.3.
[d] Refer to Section 4.2.4.
+, positive; NA, not available.

4.5.1 Outdoor air pollution

Molecular epidemiology studies have been conducted in occupationally exposed populations, the general population in different geographical areas, and susceptible populations of children. Occupational (e.g. traffic police, bus drivers, and mail carriers) or environmental (e.g. urban residents exposed to traffic and residents exposed to combustion sources) exposure studies have confirmed significantly increased levels of biomarkers of exposure of mutagenicity (e.g. urinary 1-OHP and 8-oxodG, urinary mutagenic activity, and protein adducts), biologically active internal exposures (e.g. DNA adducts and strand breaks in target tissues), cytogenetic damage (e.g. MN and CAs), mutations (in human newborns), and changes in gene expression. Table 4.19 provides an overview of the mechanistically important/relevant genetic effects in humans exposed to elevated levels of outdoor air pollution.

Increased susceptibility in humans to the effects of outdoor air pollution has been associated with genetic polymorphisms, for example *GSTM1* null, alone or in combination with selected *CYP1A1* genotypes (Hosgood et al. 2007).

A relatively small number of studies have shown that animals exposed to outdoor air pollution in situ have elevated levels of DNA damage, cytogenetic damage, and heritable mutations. Pedigree analyses of herring gulls collected from urban/industrial areas, as well as pedigree and male germ-cell analyses of both inbred and outbred mice housed in an area with elevated levels of outdoor air pollution (Yauk, 2004; Yauk et al., 2008), showed increased levels of germ-cell mutations associated with the polluted sites, and elimination of the effect via PM removal. The hypervariable repeat sequence loci examined are not associated with any known phenotype. Nevertheless, significant increases in paternal germline and germ-cell mutation rates in organisms exposed to outdoor air pollution confirm the ability of genetic damage induced by outdoor air pollution to be transmitted between generations. The phenotypic consequences of this transmission remain unknown. The most recent investigation of heritable germ-cell mutations in mice exposed to outdoor air pollution detected DNA damage in lung tissue, but not in gonadal tissue, suggesting a mechanism independent of bulky adduct formation. In situ exposures of cattle and mice to polluted outdoor air showed significant increases in cytogenetic damage in haematopoietic tissues. Numerous studies have also documented mutations and cytogenetic damage in plants exposed to elevated levels of outdoor air pollution.

In addition, there is relatively good evidence for the induction of genetic and related effects after controlled experimental exposures to components of outdoor air pollution (e.g. VOCs,

SVOCs, PM, and PM extracts). Some of these components have been previously evaluated for their carcinogenic risk to humans (IARC, 2010c, 2012c, 2013).

4.5.2 Gases

The gaseous portion of outdoor air pollution contains well-known pulmonary irritants such as ozone and NO_x. Although no studies have investigated genetic and related effects induced by exposures to the gaseous portion of outdoor air pollution specifically, reviews of the scientific literature indicate that these agents can induce genetic effects in vivo (Victorin, 1994, 1996). Recent literature suggests that abnormal immune system regulation and chronic inflammation are key mechanistic features of obstructive pulmonary disorders that enhance lung cancer risk (Turner et al., 2007).

4.5.3 Volatile organic compounds

The VOC portion of outdoor air pollution can contain a wide range of substances, and the occurrence of these substances in outdoor air is reviewed in Section 1 of this *Monograph*. The types and concentrations of these substances (e.g. benzene, formaldehyde, 1,3-butadiene, and styrene) vary with respect to the type of sample, the atmospheric conditions, the physical–chemical properties of the compound, and the proximity to known sources. The carcinogenicity of these substances and the mechanisms underlying their carcinogenic activity are addressed in detail in *IARC Monograph* Volume 100F (IARC, 2012b).

4.5.4 Semivolatile organic compounds

The SVOC fraction of outdoor air pollution also contains a wide range of substances, and the occurrence of these substances is also reviewed in Section 1 of this *Monograph*. The types and concentrations of these substances in outdoor air samples, which can include PAHs and nitroarenes that are known mutagenic carcinogens (IARC, 2010c, 2013), vary with respect to the sample collection method, the physical–chemical properties of the compounds, the PM concentration and composition, the atmospheric conditions, and the proximity to known sources. Some components of outdoor polluted air, such as PAHs and nitro-PAHs, whether in the vapour phase or adsorbed to suspended PM, can be metabolically converted to reactive species that will bind covalently to DNA in human tissues and experimental systems. These substances have been previously reviewed by IARC (see Table 1.2 in Section 1) and several have been classified as Group 1, 2A, or 2B agents. The carcinogenicity of PAHs and selected nitro-PAHs and the mechanisms underlying PAH- and nitro-PAH-induced carcinogenesis are extensively reviewed in *IARC Monographs* Volumes 92 and 105 (IARC, 2010c, 2013). Volume 105 also provides an evaluation of diesel and gasoline engine emissions, important components of outdoor air pollution (IARC, 2013).

A small number of studies provide evidence that the SVOC portion of filtered outdoor air contains substances that induce mutations in bacteria and plants, DNA damage in bacteria, and mitotic recombination in *Drosophila*. Although the identity of the implicated substances and their mechanisms of action remain unclear, some studies have documented the presence of PAHs and nitro-PAHs that are known or suspected mutagenic carcinogens (Pyysalo et al., 1987; Sera et al., 1994; Gupta et al., 1996; Du Four et al., 2004; Škarek et al., 2007).

4.5.5 Airborne particulate matter

The adsorption of substances with a range of physical–chemical properties will influence the biological properties of PM in polluted outdoor air. Studies on model particles such as carbon black and titanium dioxide have shown inverse

correlations between particle size and inflammogenicity (Duffin et al., 2007; Stoeger et al., 2006). Metals ions are involved in generating oxidative processes associated with particles deposited in the respiratory tract, and thus are a source of oxidative stress and inflammation (Tao et al., 2003; Li et al., 2008). Absorption onto carbonaceous particles of high-molecular-weight organic compounds, such as PAHs and nitro-PAHs, provides a mechanism whereby these semivolatile or non-volatile compounds can be delivered into the airways, where they can be absorbed and metabolically converted into reactive intermediates. For genotoxic organic compounds adsorbed to PM to manifest their genotoxic activity, they must be bioavailable. It is clear that organic solvents can effectively remove organic compounds from PM in outdoor air, and based on results obtained in vitro with bacteria and human cells, there is some evidence that biological fluids can effectively remove genotoxic compounds adsorbed to airborne PM. Ohsawa et al. (1983) and Takeda et al. (1983) showed that bovine serum extracts of airborne PM can induce mutations in *Salmonella*, but the potency of the extracts is low relative to organic solvent extracts. Yuan & Xun (1994) and Yuan et al. (1994) showed that PM extracts prepared using simulated lung fluid can cause DNA damage in cultured human amnion cells. Lei et al. (1993) showed that simulated lung fluid PM extracts can induce chromosomal damage in mouse haematopoietic cells.

Controlled short-term human exposures to concentrated airborne particles have been shown to be associated with pulmonary inflammation (Ghio et al. 2000; Samet et al., 2009). Nevertheless, the studies did not investigate the degree of sustained inflammation observed in rodents, most notably rats, at high lung PM burdens.

The information available indicates that inhalation exposure to PM promotes a pro-oxidant and pro-inflammatory milieu. Concomitant ROS production, together with exposure to mutagenic carcinogens such as PAHs, can be expected to give rise to a multitude of DNA lesions. If left unrepaired, these lesions can be expected to contribute to mutations and chromosomal damage that initiate and promote carcinogenesis.

Experimental exposures of rodents to organic PM extracts induced chromosomal damage. However, only a few studies used a route of exposure (inhalation) that is relevant to elevated human lung cancer risk attributable to PM exposures (Izzotti et al., 1996; Zhao et al., 2001). Intratracheal exposures of rats to PM suspensions induced DNA damage measured as strand breaks and oxidatively damaged DNA (Meng & Zhang, 2007; Lin et al., 2009; Danielsen et al., 2010; Zhang et al., 2011).

Exposure of *Drosophila* to PM extracts (larval exposure via feed) induced elevations in both somatic and germ mutation frequency. In addition, in vitro exposures to PM suspensions induced chromosomal damage in human lymphocytes and mutations in rat primary hepatocytes and *Salmonella* (Wei & Meng, 2006a, c; Alfaro-Moreno et al., 1997; Du Four et al., 2004). Collectively, these studies indicate that pulmonary exposure to PM or PM extracts causes genetic damage.

The bulk of published studies that assessed the induction of genetic and related effects in experimental systems were performed with organic solvent extracts of PM collected from polluted locations. Chemical analysis of extracts of PM collected from a wide range of locations clearly indicates that the matrix contains numerous PAHs and nitro-PAHs that are classified by IARC as known or probable human carcinogens (Yang et al., 1994; Durant et al., 1998; Pedersen et al., 1999, 2004, 2005; Brits et al., 2004; Calderón-Segura et al., 2004). Not surprisingly, organic solvent extracts of outdoor air can induce CAs, MN, SCEs, DNA strand breaks, unscheduled DNA synthesis, bulky adducts, and oxidative DNA lesions in cultured human cells. In addition, organic solvent extracts of PM can induce

Table 4.20 Summary by end-point of genetic and related effects induced in humans and experimental systems by exposure to outdoor air pollution or samples derived from outdoor air pollution

End-point	Humans	Experimental animals	Mammalian cells	Plants	Bacteria	Acellular
Mutations	(+)[a]	+	+	+	+	–[b]
Cytogenetic damage (CAs, MN, SCEs)	+	+	+	+	NA	NA
Stable DNA adducts	+	+	+	NE	NE	+
DNA strand breaks	+	+/–[c]	+	NE	NE	+
Oxidatively damaged DNA	+	+/–[d]	+	NE	NE	+
Oxidative stress and inflammation	+	+	+	NE	NE	+
Cell transformation	NA	NA	+	NA	NA	NA
Epigenetic changes	+	+	NE	NE	NE	NA

[a] Limited information available.
[b] Not applicable.
[c] Few studies, conflicting results. See Table 4.10, Section 4.2.3c.
[d] Few studies, conflicting results. See Supplemental Table S14 (available online), Section 4.2.3e.
+, positive; –, negative; CAs, chromosomal aberration; MN, micronuclei; NA, not available; NE, not evaluated; SCEs, sister chromatid exchanges.

mutations, CAs, aneuploidy, MN, SCEs, DNA strand breaks, bulky adducts, and oxidative DNA lesions in cultured mammalian cells, as well as nuclear and mitochondrial DNA mutations and gene conversion in yeast, and mutations and DNA damage in bacteria. Finally, organic PM extracts can induce bulky adducts, DNA strand breaks, and oxidative lesions in naked DNA in solution.

In addition, effects induced by water and/or acid extracts of PM are also well documented. For example, acid extracts of PM induce MN and DNA strand breaks in cultured human lymphocytes (Yuan et al., 1999a, b; Jayasekher, 2009). Acid extracts contained transition metals, including nickel and chromium, which are known to participate in Fenton reactions that generate reactive peroxide and hydroxyl radicals, which contribute to the formation of ROS. Indeed, aqueous extracts of airborne PM have been shown to induce DNA strand breaks in rat lung, oxidative lesions on naked DNA in solution, the formation of ROS in vitro, and the formation of ROS in cultured mammalian cells.

Genetic and related effects of outdoor air pollution and other mechanistic events

Tables 4.20 and 4.21 summarize the genetic and related effects in humans and experimental systems induced by exposures to outdoor air pollution and samples derived from outdoor air pollution. A large body of evidence clearly indicates that humans exposed to elevated levels of outdoor air pollution have increased levels of chromosomal damage, including chromosome breaks and aneuploidy. Similar effects in experimental systems, both in vivo and in vitro, are also well documented. In addition, a variety of other genotoxic effects in humans and experimental animals exposed to elevated levels of outdoor air pollution or samples derived from outdoor air pollution (e.g. PM and PM extracts) are also well documented (e.g. mutations, DNA strand breaks, stable DNA adducts, and oxidized nucleobases). Sustained inflammation is induced in humans and experimental animals exposed to elevated levels of outdoor air pollution or samples derived from outdoor air pollution.

Table 4.21 Summary by exposure of genetic and related effects in humans and experimental systems induced by exposure to outdoor air pollution or samples derived from outdoor air pollution

Agent	End-point induced	
	Human	Experimental systems[a]
Outdoor air pollution	Mutations[b], CAs, MN, SCEs, DNA strand breaks, oxidative DNA lesions, bulky DNA adducts	Somatic[c] and gametic[d] mutations, CAs in mice, MN in plants, bulky DNA adducts, DNA strand breaks, oxidized nucleobases
VOCs[e]	NE	
SVOCs[f]	NA	Somatic[b] and gametic[g] mutations, SCEs in murine cells, mutations in bacteria, DNA damage in bacteria
Airborne suspension of concentrated PM (CAPs)	DNA strand breaks, oxidative DNA lesions, oxidative stress, inflammation	NA
PM suspensions	Inflammation	Mutations in bacteria and mammalian cells, CAs in human cells, strand breaks in mammalian cells and naked DNA, ROS formation
Aqueous or acidic PM extracts	NA	DNA strand breaks in rat lung, MN and DNA strand breaks in human cells, oxidative DNA lesions, ROS formation in mammalian cells and in vitro
Organic solvent PM extracts	NA	CAs, MN, SCEs, DNA strand breaks, unscheduled DNA synthesis, bulky adducts, and oxidative DNA lesions in human cells. Mutations, CAs, aneuploidy, MN, SCEs, bulky adducts, oxidative DNA lesions, and ROS formation in mammalian cells. Nuclear and mitochondrial DNA mutations and mitotic recombination in yeast. Mutations and DNA damage in bacteria

[a] Includes experimental and in situ plant, animal, and insect exposures, exposures of human cells in vitro, exposures of mammalian cells in vitro, exposures of yeast and bacteria, and exposures of naked DNA.
[b] Single study of newborns in the Cracow region of Poland (see Section 4.2.1a).
[c] *Drosophila* and *Tradescantia* only.
[d] Mice and herring gulls exposed in situ.
[e] Operationally defined as substances that exist in vapour phase at ambient temperatures and pressures.
[f] Operationally defined as substances that exist partially in vapour phase at ambient temperatures and pressures. Balance adsorbed to PM.
[g] *Arabidopsis* only (seed exposure).
CAs, chromosomal aberrations; CAPs, concentrated ambient particles; MN, micronuclei; NA, not applicable; NE, not evaluated; PM, particulate matter; ROS, reactive oxygen species; SCEs, sister chromatid exchanges; SVOCs, semivolatile organic compounds; VOCs, volatile organic compounds.

Cross-sectional studies of humans lend support to the contention that alterations in the pattern of DNA methylation in circulating lymphocytes can be induced by exposure to high levels of outdoor air pollution (Hou et al., 2011).

Polluted outdoor air can contain a wide range of agents, and the PM fraction is known to contain several substances that can initiate tumour formation via genetic damage and mutation (e.g. PAHs and transition metals), as well as less-harmful constituents that induce responses that contribute to tumour promotion (e.g. inflammation). In sum, there is compelling evidence across species and experimental systems that exposure to air pollution PM is associated with increased levels of DNA damage, mutations, and chromosomal damage. Other mechanistic events include sustained proliferative signalling, evasion of growth suppression, resistance to cell death, stimulation of angiogenesis, replicative immortality, activation of invasion, and metastasis (see Supplemental Figure S4).

References

Abou Chakra OR, Joyeux M, Nerrière E, Strub MP, Zmirou-Navier D (2007). Genotoxicity of organic extracts of urban airborne particulate matter: an assessment within a personal exposure study. *Chemosphere*, 66(7):1375–81. doi:10.1016/j.chemosphere.2006.06.066 PMID:16901531

Adamkiewicz G, Ebelt S, Syring M, Slater J, Speizer FE, Schwartz J et al. (2004). Association between air pollution exposure and exhaled nitric oxide in an elderly population. *Thorax*, 59(3):204–9. doi:10.1136/thorax.2003.006445 PMID:14985553

Adamson IY, Prieditis H, Hedgecock C, Vincent R (2000). Zinc is the toxic factor in the lung response to an atmospheric particulate sample. *Toxicol Appl Pharmacol*, 166(2):111–9. doi:10.1006/taap.2000.8955 PMID:10896852

Adamson IY, Prieditis H, Vincent R (1999a). Pulmonary toxicity of an atmospheric particulate sample is due to the soluble fraction. *Toxicol Appl Pharmacol*, 157(1):43–50. doi:10.1006/taap.1999.8658 PMID:10329506

Adamson IY, Vincent R, Bjarnason SG (1999b). Cell injury and interstitial inflammation in rat lung after inhalation of ozone and urban particulates. *Am J Respir Cell Mol Biol*, 20(5):1067–72. doi:10.1165/ajrcmb.20.5.3468 PMID:10226078

Adonis M, Gil L (1993). Mutagenicity of organic extracts from Santiago (Chile) airborne particulate matter. *Mutat Res*, 292(1):51–61. doi:10.1016/0165-1161(93)90007-M PMID:7688097

Adonis M, Gil L (2000). Polycyclic aromatic hydrocarbon levels and mutagenicity of inhalable particulate matter in Santiago, Chile. *Inhal Toxicol*, 12(12):1173–83. doi:10.1080/08958370050198520 PMID:11114787

Agurell E, Stensman C (1992). *Salmonella* mutagenicity of three complex mixtures assayed with the microsuspension technique. A WHO/IPCS/CSCM study. *Mutat Res*, 276(1–2):87–91. doi:10.1016/0165-1110(92)90057-G PMID:1370111

al-Khodairy F, Al-Dakan A, Akel M, Hannan MA (1998). A comparative analysis of mutagenic activities of air samples collected from Riyadh before, during and after the Gulf War. *Int J Environ Health Res*, 8(1):15–22. doi:10.1080/09603129873615

Alfaro Moreno E, Flores Rojas G, Hartasanchez Frenk F, Orozco de la Huerta A, Quintana Belmares R, Osornio Vargas AR (1997). In vitro induction of abnormal anaphases by contaminating atmospheric dust from the City of Mexicali, Baja California, Mexico. *Arch Med Res*, 28(4):549–53. PMID:9428582

Alfaro-Moreno E, Martínez L, García-Cuellar C, Bonner JC, Murray JC, Rosas I et al. (2002). Biologic effects induced in vitro by PM_{10} from three different zones of Mexico City. *Environ Health Perspect*, 110(7):715–20. doi:10.1289/ehp.02110715 PMID:12117649

Alfheim I, Hongslo J, Moller M, Ramdahl T, Sortland B, Wikström L et al. (1984). Air pollution from aluminum smelting plants. II. The contribution from an aluminum smelting plant using the Söderberg process to polycyclic aromatic hydrocarbons and mutagens in ambient air. *Toxicol Environ Chem*, 8(2–3):195–212. doi:10.1080/02772248409357052

Alfheim I, Löfroth G, Møller M (1983). Bioassay of extracts of ambient particulate matter. *Environ Health Perspect*, 47:227–38. doi:10.1289/ehp.8347227 PMID:6186477

Alink GM, Smit HA, van Houdt JJ, Kolkman JR, Boleij JS (1983). Mutagenic activity of airborne particulates at non-industrial locations. *Mutat Res*, 116(1):21–34. doi:10.1016/0165-1218(83)90003-4 PMID:6828047

Allen RW, Carlsten C, Karlen B, Leckie S, van Eeden S, Vedal S et al. (2011). An air filter intervention study of endothelial function among healthy adults in a woodsmoke-impacted community. *Am J Respir Crit Care Med*, 183(9):1222–30. doi:10.1164/rccm.201010-1572OC PMID:21257787

American Thoracic Society (2000). American Thoracic Society. What constitutes an adverse health effect of air pollution? Official statement of the American Thoracic Society. *Am J Respir Crit Care Med*, 161(2 Pt 1):665–73. doi:10.1164/ajrccm.161.2.ats4-00 PMID:10673213

Anbazhagan M, Arumugam P, Ramesh A, Santhiya ST (2010). Genetic risk assessment in traffic policemen of Chennai City by sister chromatid exchange analysis. *Int J Hum Genet*, 10(4):251–5.

André E, Stoeger T, Takenaka S, Bahnweg M, Ritter B, Karg E et al. (2006). Inhalation of ultrafine carbon particles triggers biphasic pro-inflammatory response in the mouse lung. *Eur Respir J*, 28(2):275–85. doi:10.1183/09031936.06.00071205 PMID:16641123

Andrysík Z, Vondráček J, Marvanová S, Ciganek M, Neča J, Pěnčíková K et al. (2011). Activation of the aryl hydrocarbon receptor is the major toxic mode of action of an organic extract of a reference urban dust particulate matter mixture: the role of polycyclic aromatic hydrocarbons. *Mutat Res*, 714(1–2):53–62. doi:10.1016/j.mrfmmm.2011.06.011 PMID:21762708

Anwar WA, Kamal AAM (1988). Cytogenetic effects in a group of traffic policemen in Cairo. *Mutat Res*, 208(3–4):225–31. doi:10.1016/0165-7992(88)90065-6 PMID:2456461

Arayasiri M, Mahidol C, Navasumrit P, Autrup H, Ruchirawat M (2010). Biomonitoring of benzene and 1,3-butadiene exposure and early biological effects in traffic policemen. *Sci Total Environ*, 408(20):4855–62. doi:10.1016/j.scitotenv.2010.06.033 PMID:20627202

Ares J, Eckl PM, Raffelsberger I (2000). Genotoxicity at low level dose of inspirable urban ambient air particulate in a semiarid regime. *Environ Monit Assess*, 63(3):388–408. doi:10.1023/A:1006202307463

Arey J, Harger WP, Helmig D, Atkinson R (1992). Bioassay-directed fractionation of mutagenic PAH atmospheric photooxidation products and ambient particulate extracts. *Mutat Res*, 281(1):67–76. doi:10.1016/0165-7992(92)90038-J PMID:1371594

Arey J, Zielinska B, Harger WP, Atkinson R, Winer AM (1988). The contribution of nitrofluoranthenes and nitropyrenes to the mutagenic activity of ambient particulate organic matter collected in southern California. *Mutat Res*, 207(2):45–51. doi:10.1016/0165-7992(88)90040-1 PMID:3340093

Athanasiou K, Arzimanoglou I, Piccoli C, Yamasaki H, Arzimanoglou II (1987). Mutagenicity, sister chromatid exchange inducibility and in vitro cell transforming ability of particulates from Athens air. *Cell Biol Toxicol*, 3(3):251–61. doi:10.1007/BF00117863 PMID:3333731

Athanasiou K, Viras LG, Siskos PA (1986). Mutagenicity and polycyclic aromatic hydrocarbons analysis of ambient airborne particles collected in Athens, Greece. *Sci Total Environ*, 52(3):201–9. doi:10.1016/0048-9697(86)90120-8 PMID:3526551

Auger F, Gendron MC, Chamot C, Marano F, Dazy AC (2006). Responses of well-differentiated nasal epithelial cells exposed to particles: role of the epithelium in airway inflammation. *Toxicol Appl Pharmacol*, 215(3):285–94. doi:10.1016/j.taap.2006.03.002 PMID:16647095

Autrup H, Vestergaard AB (1996). Transplacental transfer of environmental genotoxins – polycyclic aromatic hydrocarbon-albumin in nonsmoking women. *Environ Health Perspect*, 104:Suppl 3: 625–7. PMID:8781394

Avogbe PH, Ayi-Fanou L, Autrup H, Loft S, Fayomi B, Sanni A et al. (2005). Ultrafine particulate matter and high-level benzene urban air pollution in relation to oxidative DNA damage. *Carcinogenesis*, 26(3):613–20. doi:10.1093/carcin/bgh353 PMID:15591089

Ayi-Fanou L, Avogbe PH, Fayomi B, Keith G, Hountondji C, Creppy EE et al. (2011). DNA-adducts in subjects exposed to urban air pollution by benzene and polycyclic aromatic hydrocarbons (PAHs) in Cotonou, Benin. *Environ Toxicol*, 26(1):93–102. doi:10.1002/tox.20533 PMID:20014405

Ayi Fanou L, Mobio TA, Creppy EE, Fayomi B, Fustoni S, Møller P et al. (2006). Survey of air pollution in Cotonou, Benin – air monitoring and biomarkers. *Sci Total Environ*, 358(1–3):85–96. doi:10.1016/j.scitotenv.2005.03.025 PMID:15916795

Baccarelli A, Wright RO, Bollati V, Tarantini L, Litonjua AA, Suh HH et al. (2009). Rapid DNA methylation changes after exposure to traffic particles. *Am J Respir Crit Care Med*, 179(7):572–8. doi:10.1164/rccm.200807-1097OC PMID:19136372

Bagate K, Meiring JJ, Gerlofs-Nijland ME, Vincent R, Cassee FR, Borm PJ (2004). Vascular effects of ambient particulate matter instillation in spontaneous hypertensive rats. *Toxicol Appl Pharmacol*, 197(1):29–39. doi:10.1016/j.taap.2004.02.005 PMID:15126072

Bagley ST, Stoltz SL, Becker DM, Keen RE (1992). Characterization of organic extracts from standard reference materials 1649, 'urban dust/organics,' and 1650, 'diesel particulate matter', using a microsuspension assay. A WHO/IPCS/CSCM study. *Mutat Res*, 276(1–2):81–6. doi:10.1016/0165-1110(92)90056-F PMID:1370110

Bagryantseva Y, Novotna B, Rossner P Jr, Chvatalova I, Milcova A, Svecova V et al. (2010). Oxidative damage to biological macromolecules in Prague bus drivers and garagemen: impact of air pollution and genetic polymorphisms. *Toxicol Lett*, 199(1):60–8. doi:10.1016/j.toxlet.2010.08.007 PMID:20723587

Bai JY, Zhang JD, Wang HB, Yang WM (1999). The genetic toxicity of the organic fractions of air particulates. *Chin J Publ Health*, 18:89–91.

Bai YP, Li J, Fan XY, Yao SQ, Jiang SF, Jin YL (2005). Effects of traffic air pollution on the rate of micronucleus and sister chromatid exchange of traffic police in a city. *Carcinog Teratog Mutagen*, 17(4):250–3. Available from: http://www.egh.net.cn/EN/Y2005/V17/I4/250.

Bailey MR, Fry FA, James AC (1985). Long-term retention of particles in the human respiratory tract. *J Aerosol Sci*, 16(4):295–305. doi:10.1016/0021-8502(85)90037-0

Ball JC, Straccia AM, Young WC, Aust AE (2000). The formation of reactive oxygen species catalyzed by neutral, aqueous extracts of NIST ambient particulate matter and diesel engine particles. *J Air Waste Manag Assoc*, 50(11):1897–903. doi:10.1080/10473289.2000.10464231 PMID:11111334

Barraza-Villarreal A, Sunyer J, Hernandez-Cadena L, Escamilla-Nuñez MC, Sienra-Monge JJ, Ramírez-Aguilar M et al. (2008). Air pollution, airway inflammation, and lung function in a cohort study of Mexico City schoolchildren. *Environ Health Perspect*, 116(6):832–8. doi:10.1289/ehp.10926 PMID:18560490

Baulig A, Sourdeval M, Meyer M, Marano F, Baeza-Squiban A (2003). Biological effects of atmospheric particles on human bronchial epithelial cells. Comparison with diesel exhaust particles. *Toxicol In Vitro*, 17(5–6):567–73. doi:10.1016/S0887-2333(03)00115-2 PMID:14599446

Becher R, Bucht A, Øvrevik J, Hongslo JK, Dahlman HJ, Samuelsen JT et al. (2007). Involvement of NADPH oxidase and iNOS in rodent pulmonary cytokine responses to urban air and mineral particles. *Inhal Toxicol*, 19(8):645–55. doi:10.1080/08958370701353528 PMID:17510837

Becker S, Dailey LA, Soukup JM, Grambow SC, Devlin RB, Huang YC (2005). Seasonal variations in air pollution particle-induced inflammatory mediator release and oxidative stress. *Environ Health Perspect*, 113(8):1032–8. doi:10.1289/ehp.7996 PMID:16079075

Becker S, Soukup JM, Gilmour MI, Devlin RB (1996). Stimulation of human and rat alveolar macrophages by urban air particulates: effects on oxidant radical generation and cytokine production. *Toxicol Appl*

Pharmacol, 141(2):637–48. doi:10.1006/taap.1996.0330 PMID:8975789

Beelen R, Hoek G, van den Brandt PA, Goldbohm RA, Fischer P, Schouten LJ et al. (2008a). Long-term exposure to traffic-related air pollution and lung cancer risk. *Epidemiology*, 19(5):702–10. doi:10.1097/EDE.0b013e318181b3ca PMID:18633326

Bennett WD, Zeman KL (2004). Effect of body size on breathing pattern and fine-particle deposition in children *J Appl Physiol (1985)*, 97(3):821–6.

Bennett WD, Zeman KL, Kim C (1996). Variability of fine particle deposition in healthy adults: effect of age and gender. *Am J Respir Crit Care Med*, 153(5):1641–7. doi:10.1164/ajrccm.153.5.8630615 PMID:8630615

Beskid O, Binkova B, Dusek Z, Rössner P, Solansky I, Kalina I et al. (2007). Chromosomal aberrations by fluorescence in situ hybridization (FISH) – biomarker of exposure to carcinogenic PAHs. *Mutat Res*, 620(1–2):62–70. doi:10.1016/j.mrfmmm.2007.02.023 PMID:17412370

Bind M-A, Baccarelli A, Zanobetti A, Tarantini L, Suh H, Vokonas P et al. (2012). Air pollution and markers of coagulation, inflammation, and endothelial function: associations and epigene-environment interactions in an elderly cohort. *Epidemiology*, 23(2):332–40. doi:10.1097/EDE.0b013e31824523f0 PMID:22237295

Binková B, Cerná M, Pastorková A, Jelínek R, Benes I, Novák J et al. (2003). Biological activities of organic compounds adsorbed onto ambient air particles: comparison between the cities of Teplice and Prague during the summer and winter seasons 2000–2001. *Mutat Res*, 525(1–2):43–59. doi:10.1016/S0027-5107(02)00312-3 PMID:12650904

Binková B, Chvatalova I, Lnenickova Z, Milcova A, Tulupova E, Farmer PB et al. (2007). PAH-DNA adducts in environmentally exposed population in relation to metabolic and DNA repair gene polymorphisms. *Mutat Res*, 620(1–2):49–61. doi:10.1016/j.mrfmmm.2007.02.022 PMID:17412371

Binková B, Lewtas J, Mísková I, Rössner P, Cerná M, Mrácková G et al. (1996). Biomarker studies in northern Bohemia. *Environ Health Perspect*, 104:Suppl 3: 591–7. doi:10.1289/ehp.96104s3591 PMID:8781388

Bolognesi C, Gallerani E, Bonatti S, De Ferrari M, Fontana V, Valerio F et al. (1997b). Sister chromatid exchange induction in peripheral blood lymphocytes of traffic police workers. *Mutat Res*, 394(1–3):37–44. doi:10.1016/S1383-5718(97)00121-6 PMID:9434841

Bolognesi C, Merlo F, Rabboni R, Valerio F, Abbondandolo A (1997a). Cytogenetic biomonitoring in traffic police workers: micronucleus test in peripheral blood lymphocytes. *Environ Mol Mutagen*, 30(4):396–402. doi:10.1002/(SICI)1098-2280(1997)30:4<396::AID-EM4>3.0.CO;2-H PMID:9435880

Bonetta S, Gianotti V, Bonetta S, Gosetti F, Oddone M, Gennaro MC et al. (2009). DNA damage in A549 cells exposed to different extracts of $PM_{2.5}$ from industrial, urban and highway sites. *Chemosphere*, 77(7):1030–4. doi:10.1016/j.chemosphere.2009.07.076 PMID:19729187

Bos I, De Boever P, Emmerechts J, Buekers J, Vanoirbeek J, Meeusen R et al. (2012). Changed gene expression in brains of mice exposed to traffic in a highway tunnel. *Inhal Toxicol*, 24(10):676–86. doi:10.3109/08958378.2012.714004 PMID:22906174

Brain JD (1970). Free cells in the lungs. Some aspects of their role, quantitation, and regulation. *Arch Intern Med*, 126(3):477–87. doi:10.1001/archinte.1970.00310090107013 PMID:4915938

Bräuner EV, Forchhammer L, Møller P, Barregard L, Gunnarsen L, Afshari A et al. (2008a). Indoor particles affect vascular function in the aged: an air filtration-based intervention study. *Am J Respir Crit Care Med*, 177(4):419–25. doi:10.1164/rccm.200704-632OC PMID:17932377

Bräuner EV, Forchhammer L, Møller P, Simonsen J, Glasius M, Wåhlin P et al. (2007). Exposure to ultrafine particles from ambient air and oxidative stress-induced DNA damage. *Environ Health Perspect*, 115(8):1177–82. doi:10.1289/ehp.9984 PMID:17687444

Bräuner EV, Møller P, Barregard L, Dragsted LO, Glasius M, Wåhlin P et al. (2008b). Exposure to ambient concentrations of particulate air pollution does not influence vascular function or inflammatory pathways in young healthy individuals. *Part Fibre Toxicol*, 5(1):13 doi:10.1186/1743-8977-5-13 PMID:18837984

Bridgewater LC, Manning FC, Patierno SR (1994). Base-specific arrest of *in vitro* DNA replication by carcinogenic chromium: relationship to DNA interstrand crosslinking. *Carcinogenesis*, 15(11):2421–7. doi:10.1093/carcin/15.11.2421 PMID:7955085

Brits E, Schoeters G, Verschaeve L (2004). Genotoxicity of PM_{10} and extracted organics collected in an industrial, urban and rural area in Flanders, Belgium. *Environ Res*, 96(2):109–18. doi:10.1016/j.envres.2004.03.006 PMID:15325871

Bronzetti G, Cini M, Paoli M, Ciacchini G, Giaconi V, Morichetti E (1997). Mutagenicity and chemical analysis of airborne particulate matter collected in Pisa. *J Environ Pathol Toxicol Oncol*, 16(2–3):147–56. PMID:9275995

Brook RD (2008). Cardiovascular effects of air pollution. *Clin Sci (Lond)*, 115(6):175–87. doi:10.1042/CS20070444 PMID:18691154

Brook RD, Brook JR, Urch B, Vincent R, Rajagopalan S, Silverman F (2002). Inhalation of fine particulate air pollution and ozone causes acute arterial vasoconstriction in healthy adults. *Circulation*, 105(13):1534–6. doi:10.1161/01.CIR.0000013838.94747.64 PMID:11927516

Brook RD, Jerrett M, Brook JR, Bard RL, Finkelstein MM (2008). The relationship between diabetes mellitus and traffic-related air pollution. *J Occup Environ*

Med, 50(1):32–8. doi:10.1097/JOM.0b013e31815dba70 PMID:18188079

Brook RD, Rajagopalan S, Pope CA 3rd, Brook JR, Bhatnagar A, Diez-Roux AV et al.; American Heart Association Council on Epidemiology and Prevention, Council on the Kidney in Cardiovascular Disease, and Council on Nutrition, Physical Activity and Metabolism (2010). Particulate matter air pollution and cardiovascular disease: an update to the scientific statement from the American Heart Association. *Circulation*, 121(21):2331–78. doi:10.1161/CIR.0b013e3181dbece1 PMID:20458016

Brown DM, Donaldson K, Stone V (2004). Effects of PM_{10} in human peripheral blood monocytes and J774 macrophages. *Respir Res*, 5(1):29 doi:10.1186/1465-9921-5-29 PMID:15613243

Brown DM, Hutchison L, Donaldson K, Stone V (2007). The effects of PM_{10} particles and oxidative stress on macrophages and lung epithelial cells: modulating effects of calcium-signaling antagonists. *Am J Physiol Lung Cell Mol Physiol*, 292(6):L1444–51. doi:10.1152/ajplung.00162.2006 PMID:17369293

Brown LE, Trought KR, Bailey CI, Clemons JH (2005). 2,3,7,8-TCDD equivalence and mutagenic activity associated with PM_{10} from three urban locations in New Zealand. *Sci Total Environ*, 349(1–3):161–74. doi:10.1016/j.scitotenv.2005.01.008 PMID:16198678

Burgaz S, Demircigil GC, Karahalil B, Karakaya AE (2002). Chromosomal damage in peripheral blood lymphocytes of traffic policemen and taxi drivers exposed to urban air pollution. *Chemosphere*, 47(1):57–64. doi:10.1016/S0045-6535(01)00185-0 PMID:11996136

Buschini A, Cassoni F, Anceschi E, Pasini L, Poli P, Rossi C (2001). Urban airborne particulate: genotoxicity evaluation of different size fractions by mutagenesis tests on microorganisms and comet assay. *Chemosphere*, 44(8):1723–36. doi:10.1016/S0045-6535(00)00550-6 PMID:11534904

Buthbumrung N, Mahidol C, Navasumrit P, Promvijit J, Hunsonti P, Autrup H et al. (2008). Oxidative DNA damage and influence of genetic polymorphisms among urban and rural schoolchildren exposed to benzene. *Chem Biol Interact*, 172(3):185–94. doi:10.1016/j.cbi.2008.01.005 PMID:18282563

Byun H-M, Panni T, Motta V, Hou L, Nordio F, Apostoli P et al. (2013). Effects of airborne pollutants on mitochondrial DNA methylation. *Part Fibre Toxicol*, 10(1):18 doi:10.1186/1743-8977-10-18 PMID:23656717

Calderón-Garcidueñas L, Osnaya N, Rodríguez-Alcaraz A, Villarreal-Calderón A (1997). DNA damage in nasal respiratory epithelium from children exposed to urban pollution. *Environ Mol Mutagen*, 30(1):11–20. doi:10.1002/(SICI)1098-2280(1997)30:1<11::AID-EM3>3.0.CO;2-F PMID:9258325

Calderón-Garcidueñas L, Osnaya-Brizuela N, Ramirez-Martinez L, Villarreal-Calderon A (1996). DNA strand breaks in human nasal respiratory epithelium are induced upon exposure to urban pollution. *Environ Health Perspect*, 104(2):160–8. PMID:8820583

Calderón-Garcidueñas L, Villarreal-Calderon R, Valencia-Salazar G, Henríquez-Roldán C, Gutiérrez-Castrellón P, Torres-Jardón R et al. (2008). Systemic inflammation, endothelial dysfunction, and activation in clinically healthy children exposed to air pollutants. *Inhal Toxicol*, 20(5):499–506. doi:10.1080/08958370701864797 PMID:18368620

Calderón-Garcidueñas L, Wen-Wang L, Zhang YJ, Rodriguez-Alcaraz A, Osnaya N, Villarreal-Calderón A et al. (1999). 8-hydroxy-2′-deoxyguanosine, a major mutagenic oxidative DNA lesion, and DNA strand breaks in nasal respiratory epithelium of children exposed to urban pollution. *Environ Health Perspect*, 107(6):469–74. PMID:10339447

Calderón-Segura ME, Gómez-Arroyo S, Villalobos-Pietrini R, Butterworth FM, Amador-Muñoz O (2004). The effects of seasonal weather on the genotoxicity, cytokinetic properties, cytotoxicity and organochemical content of extracts of airborne particulates in Mexico City. *Mutat Res*, 558(1–2):7–17. doi:10.1016/j.mrgentox.2003.10.018 PMID:15036114

Carere A, Andreoli C, Galati R, Leopardi P, Marcon F, Rosati MV et al. (2002). Biomonitoring of exposure to urban air pollutants: analysis of sister chromatid exchanges and DNA lesions in peripheral lymphocytes of traffic policemen. *Mutat Res*, 518(2):215–24. doi:10.1016/S1383-5718(02)00108-0 PMID:12113772

Carey IM, Atkinson RW, Kent AJ, van Staa T, Cook DG, Anderson HR (2013). Mortality associations with long-term exposure to outdoor air pollution in a national English cohort. *Am J Respir Crit Care Med*, 187(11):1226–33. doi:10.1164/rccm.201210-1758OC PMID:23590261

Carreras HA, Calderón-Segura ME, Gómez-Arroyo S, Murillo-Tovar MA, Amador-Muñoz O (2013). Composition and mutagenicity of PAHs associated with urban airborne particles in Córdoba, Argentina. *Environ Pollut*, 178:403–10. doi:10.1016/j.envpol.2013.03.016 PMID:23624338

Casellas M, Fernandez P, Bayona JM, Solanas AM (1995). Bioassay-directed chemical analysis of genotoxic components in urban airborne particulate matter from Barcelona (Spain). *Chemosphere*, 30(4):725–40. doi:10.1016/0045-6535(94)00438-Z PMID:7889349

Cassee FR, Boere AJ, Fokkens PH, Leseman DL, Sioutas C, Kooter IM et al. (2005). Inhalation of concentrated particulate matter produces pulmonary inflammation and systemic biological effects in compromised rats. *J Toxicol Environ Health A*, 68(10):773–96. doi:10.1080/15287390590930171 PMID:16020176

Cavallo D, Ursini CL, Carelli G, Iavicoli I, Ciervo A, Perniconi B et al. (2006). Occupational exposure in airport personnel: characterization and evaluation

of genotoxic and oxidative effects. *Toxicology*, 223(1–2):26–35. doi:10.1016/j.tox.2006.03.003 PMID:16621217

Cavanagh JA, Trought K, Brown L, Duggan S (2009). Exploratory investigation of the chemical characteristics and relative toxicity of ambient air particulates from two New Zealand cities. *Sci Total Environ*, 407(18):5007–18. doi:10.1016/j.scitotenv.2009.05.020 PMID:19570565

Cebulska-Wasilewska A, Wiecheć A, Panek A, Binková B, Srám RJ, Farmer PB (2005). Influence of environmental exposure to PAHs on the susceptibility of lymphocytes to DNA-damage induction and on their repair capacity. *Mutat Res*, 588(2):73–81. doi:10.1016/j.mrgentox.2005.08.013 PMID:16311068

Černá M, Pastorková A, Vrbíková V, Smíd J, Rössner P (1999). Mutagenicity monitoring of airborne particulate matter (PM_{10}) in the Czech Republic. *Mutat Res*, 444(2):373–86. doi:10.1016/S1383-5718(99)00107-2 PMID:10521677

Cesaroni G, Badaloni C, Gariazzo C, Stafoggia M, Sozzi R, Davoli M et al. (2013). Long-term exposure to urban air pollution and mortality in a cohort of more than a million adults in Rome. *Environ Health Perspect*, 121(3):324–31. doi:10.1289/ehp.1205862 PMID:23308401

Chandrasekaran R, Samy PL, Murthy PB (1996). Increased sister chromatid exchange (SCE) frequencies in lymphocytes from traffic policemen exposed to automobile exhaust pollution. *Hum Exp Toxicol*, 15(4):301–4. doi:10.1177/096032719601500405 PMID:8845219

Chen CH, Lu YM, Zhang KJ (1999). Analysis of chromosome aberration in peripheral blood lymphocytes from traffic policemen. *J Hyg Res*, 6:324–5.

Chen ST, Lin CC, Liu YS, Lin C, Hung PT, Jao CW et al. (2013). Airborne particulate collected from central Taiwan induces DNA strand breaks, poly(ADP-ribose) polymerase-1 activation, and estrogen-disrupting activity in human breast carcinoma cell lines. *J Environ Sci Health A Tox Hazard Subst Environ Eng*, 48(2):173–81. doi:10.1080/10934529.2012.717809 PMID:23043339

Ciganek M, Neca J, Adamec V, Janosek J, Machala M (2004). A combined chemical and bioassay analysis of traffic-emitted polycyclic aromatic hydrocarbons. *Sci Total Environ*, 334–335:141–8. doi:10.1016/j.scitotenv.2004.04.034 PMID:15504500

Clarke RW, Catalano P, Coull B Koutrakis P, Krishna Murthy GG, Rice T et al. (2000a). Age-related responses in rats to concentrated urban air particles (CAPs). *Inhal Toxicol*, 12:73–84. doi:10.1080/089583700196400

Clarke RW, Catalano PJ, Koutrakis P, Murthy GG, Sioutas C, Paulauskis J et al. (1999). Urban air particulate inhalation alters pulmonary function and induces pulmonary inflammation in a rodent model of chronic bronchitis. *Inhal Toxicol*, 11(8):637–56. doi:10.1080/089583799196781 PMID:10477440

Clarke RW, Coull B, Reinisch U, Catalano P, Killingsworth CR, Koutrakis P et al. (2000b). Inhaled concentrated ambient particles are associated with hematologic and bronchoalveolar lavage changes in canines. *Environ Health Perspect*, 108(12):1179–87. doi:10.1289/ehp.001081179 PMID:11133399

Claxton LD, Warren S, Zweidinger R, Creason J (2001). A comparative assessment of Boise, Idaho, ambient air fine particle samples using the plate and microsuspension *Salmonella* mutagenicity assays. *Sci Total Environ*, 275(1–3):95–108. doi:10.1016/S0048-9697(00)00857-3 PMID:11482407

Claxton LD, Woodall GM Jr (2007). A review of the mutagenicity and rodent carcinogenicity of ambient air. *Mutat Res*, 636(1–3):36–94. doi:10.1016/j.mrrev.2007.01.001 PMID:17451995

Commoner B, Madyastha R, Bronsdon A, Vithayathil AJ (1978). Environmental mutagens in urban air particulates. *J Toxicol Environ Health*, 4(1):59–77. doi:10.1080/15287397809529645 PMID:633412

Coronas MV, Pereira TS, Rocha JA, Lemos AT, Fachel JM, Salvadori DM et al. (2009). Genetic biomonitoring of an urban population exposed to mutagenic airborne pollutants. *Environ Int*, 35(7):1023–9. doi:10.1016/j.envint.2009.05.001 PMID:19500845

Costa DL, Lehmann JR, Winsett D, Richards J, Ledbetter AD, Dreher KL (2006). Comparative pulmonary toxicological assessment of oil combustion particles following inhalation or instillation exposure. *Toxicol Sci*, 91(1):237–46. doi:10.1093/toxsci/kfj123 PMID:16449252

Courtois YA, Min S, Lachenal C, Jacquot-Deschamps JM, Callais F, Festy B (1988). Genotoxicity of organic extracts from atmospheric particles. *Ann N Y Acad Sci*, 534:724–40. doi:10.1111/j.1749-6632.1988.tb30162.x PMID:3389685

Crebelli R (1989). Monitoring of urban air pollution by mutagenicity assays. *Ann Ist Super Sanita*, 25(4):591–4. PMID:2631625

Crebelli R, Fuselli S, Baldassarri LT, Ziemacki G, Carere A, Benigni R (1995). Genotoxicity of urban air particulate matter: correlations between mutagenicity data, airborne micropollutants, and meteorological parameters. *Int J Environ Health Res*, 5(1):19–34. doi:10.1080/09603129509356830

Crebelli R, Fuselli S, Meneguz A, Aquilina G, Conti L, Leopardi P et al. (1988). In vitro and in vivo mutagenicity studies with airborne particulate extracts. *Mutat Res*, 204(4):565–75. doi:10.1016/0165-1218(88)90059-6 PMID:3280990

Cui YQ, Ji XY, Chen CM et al. (1991). Mutagenic effect of air pollution on human chorionic villi: an analysis of 2698 cases. *Carcinog Teratog Mutagen*, 13:6–10.

Daisey JM, Kneip TJ, Hawryluk I, Mukai F (1980). Seasonal variations in the bacterial mutagenicity of airborne particulate organic matter in New York City. *Environ*

Sci Technol, 14(12):1487–90. doi:10.1021/es60172a001 PMID:22279993

Danielsen PH, Loft S, Jacobsen NR, Jensen KA, Autrup H, Ravanat JL et al. (2010). Oxidative stress, inflammation, and DNA damage in rats after intratracheal instillation or oral exposure to ambient air and wood smoke particulate matter. *Toxicol Sci*, 118(2):574–85. doi:10.1093/toxsci/kfq290 PMID:20864625

Danielsen PH, Loft S, Kocbach A, Schwarze PE, Møller P (2009). Oxidative damage to DNA and repair induced by Norwegian wood smoke particles in human A549 and THP-1 cell lines. *Mutat Res*, 674(1–2):116–22. doi:10.1016/j.mrgentox.2008.10.014 PMID:19041418

Danielsen PH, Loft S, Møller P (2008). DNA damage and cytotoxicity in type II lung epithelial (A549) cell cultures after exposure to diesel exhaust and urban street particles. *Part Fibre Toxicol*, 5(1):6 doi:10.1186/1743-8977-5-6 PMID:18397523

Danielsen PH, Møller P, Jensen KA, Sharma AK, Wallin H, Bossi R et al. (2011). Oxidative stress, DNA damage, and inflammation induced by ambient air and wood smoke particulate matter in human A549 and THP-1 cell lines. *Chem Res Toxicol*, 24(2):168–84. doi:10.1021/tx100407m PMID:21235221

de Andrade SJ, Varella SD, Pereira GT, Zocolo GJ, de Marchi MR, Varanda EA (2011). Mutagenic activity of airborne particulate matter (PM_{10}) in a sugarcane farming area (Araraquara city, southeast Brazil). *Environ Res*, 111(4):545–50. doi:10.1016/j.envres.2011.03.004 PMID:21481367

De Coster S, Koppen G, Bracke M, Schroijen C, Den Hond E, Nelen V et al. (2008). Pollutant effects on genotoxic parameters and tumor-associated protein levels in adults: a cross sectional study. *Environ Health*, 7(1):26 doi:10.1186/1476-069X-7-26 PMID:18522717

De Flora S, Bagnasco M, Izzotti A, D'Agostini F, Pala M, Valerio F (1989). Mutagenicity of polycyclic aromatic hydrocarbon fractions extracted from urban air particulates. *Mutat Res*, 224(2):305–18. doi:10.1016/0165-1218(89)90169-9 PMID:2677712

de Raat WK (1983). Genotoxicity of aerosol extracts. Some methodological aspects and the contribution of urban and industrial locations. *Mutat Res*, 116(1):47–63. doi:10.1016/0165-1218(83)90005-8 PMID:6298617

de Raat WK, de Meijere FA (1988). The mutagenicity of ambient air particles from local traffic and distant sources during episodes with moderate photochemical air pollution. *Sci Total Environ*, 73(3):159–79. doi:10.1016/0048-9697(88)90426-3 PMID:2463671

de Raat WK, de Meijere FA, Kooijman SALM (1985). Mutagenicity of ambient aerosol collected in an urban and industrial area of The Netherlands. *Sci Total Environ*, 44(1):17–33. doi:10.1016/0048-9697(85)90048-8 PMID:3895434

De Vizcaya-Ruiz A, Gutierrez-Castillo ME, Uribe-Ramirez M, Cebrián ME, Mugica-Alvarez V, Sepúlveda J et al. (2006). Characterization and in vitro biological effects of concentrated particulate matter from Mexico City. *Atmos Environ*, 40:Suppl 2: 583–92. doi:10.1016/j.atmosenv.2005.12.073

Delfino RJ, Sioutas C, Malik S (2005). Potential role of ultrafine particles in associations between airborne particle mass and cardiovascular health. *Environ Health Perspect*, 113(8):934–46. doi:10.1289/ehp.7938 PMID:16079061

Delfino RJ, Staimer N, Tjoa T, Arhami M, Polidori A, Gillen DL et al. (2010a). Associations of primary and secondary organic aerosols with airway and systemic inflammation in an elderly panel cohort. *Epidemiology*, 21(6):892–902. doi:10.1097/EDE.0b013e3181f20e6c PMID:20811287

Delfino RJ, Staimer N, Tjoa T, Arhami M, Polidori A, Gillen DL et al. (2010b). Association of biomarkers of systemic inflammation with organic components and source tracers in quasi-ultrafine particles. *Environ Health Perspect*, 118(6):756–62. doi:10.1289/ehp.0901407 PMID:20123637

Delfino RJ, Staimer N, Tjoa T, Gillen DL, Polidori A, Arhami M et al. (2009). Air pollution exposures and circulating biomarkers of effect in a susceptible population: clues to potential causal component mixtures and mechanisms. *Environ Health Perspect*, 117(8):1232–8. doi:10.1289/ehp.0800194 PMID:19672402

Delfino RJ, Staimer N, Tjoa T, Polidori A, Arhami M, Gillen DL et al. (2008). Circulating biomarkers of inflammation, antioxidant activity, and platelet activation are associated with primary combustion aerosols in subjects with coronary artery disease. *Environ Health Perspect*, 116(7):898–906. doi:10.1289/ehp.11189 PMID:18629312

Dellinger B, Pryor WA, Cueto R, Squadrito GL, Hegde V, Deutsch WA (2001). Role of free radicals in the toxicity of airborne fine particulate matter. *Chem Res Toxicol*, 14(10):1371–7. doi:10.1021/tx010050x PMID:11599928

Demetriou CA, Raaschou-Nielsen O, Loft S, Møller P, Vermeulen R, Palli D et al. (2012). Biomarkers of ambient air pollution and lung cancer: a systematic review. *Occup Environ Med*, 69(9):619–27. doi:10.1136/oemed-2011-100566 PMID:22773658

Di Cesare M, Khang YH, Asaria P, Blakely T, Cowan MJ, Farzadfar F et al.; Lancet NCD Action Group (2013). Inequalities in non-communicable diseases and effective responses. *Lancet*, 381(9866):585–97. doi:10.1016/S0140-6736(12)61851-0 PMID:23410608

Di Pietro A, Visalli G, Munaò F, Baluce B, La Maestra S, Primerano P et al. (2009). Oxidative damage in human epithelial alveolar cells exposed in vitro to oil fly ash transition metals. *Int J Hyg Environ Health*, 212(2):196–208. doi:10.1016/j.ijheh.2008.05.005 PMID:18667355

Dick CA, Singh P, Daniels M, Evansky P, Becker S, Gilmour MI (2003). Murine pulmonary inflammatory responses following instillation of size-fractionated

ambient particulate matter. *J Toxicol Environ Health A*, 66(23):2193–207. doi:10.1080/716100636 PMID:14669776

Ding GW, Wang WJ, Wang XY (1999). Research on genotoxicity of TSP in various functional regions of Lanzhou city. *J Lanzhou Med Coll*, 25:17–20.

Don Porto Carero A, Hoet PHM, Verschaeve L, Schoeters G, Nemery B (2001). Genotoxic effects of carbon black particles, diesel exhaust particles, and urban air particulates and their extracts on a human alveolar epithelial cell line (A549) and a human monocytic cell line (THP-1). *Environ Mol Mutagen*, 37(2):155–63. doi:10.1002/em.1023 PMID:11246222

Donaldson K, Brown DM, Mitchell C, Dineva M, Beswick PH, Gilmour P et al. (1997). Free radical activity of PM_{10}: iron-mediated generation of hydroxyl radicals. *Environ Health Perspect*, 105:Suppl 5: 1285–9. PMID:9400739

Du Four VA, Van Larebeke N, Janssen CR (2004). Genotoxic and mutagenic activity of environmental air samples in Flanders, Belgium. *Mutat Res*, 558(1–2):155–67. doi:10.1016/j.mrgentox.2003.12.002 PMID:15036129

Duffin R, Tran L, Brown D, Stone V, Donaldson K (2007). Proinflammogenic effects of low-toxicity and metal nanoparticles in vivo and in vitro: highlighting the role of particle surface area and surface reactivity. *Inhal Toxicol*, 19(10):849–56. doi:10.1080/08958370701479323 PMID:17687716

Durant JL, Lafleur AL, Plummer EF, Taghizadeh K, Busby WF, Thilly WG (1998). Human lymphoblast mutagens in urban airborne particles. *Environ Sci Technol*, 32(13):1894–906. doi:10.1021/es9706965

Dwivedi S, Saquib Q, Al-Khedhairy AA, Ali AY, Musarrat J (2012). Characterization of coal fly ash nanoparticles and induced oxidative DNA damage in human peripheral blood mononuclear cells. *Sci Total Environ*, 437:331–8. doi:10.1016/j.scitotenv.2012.08.004 PMID:22960109

Dybdahl M, Risom L, Bornholdt J, Autrup H, Loft S, Wallin H (2004). Inflammatory and genotoxic effects of diesel particles in vitro and in vivo. *Mutat Res*, 562(1–2):119–31. doi:10.1016/j.mrgentox.2004.05.010 PMID:15279835

Eichhorn GL, Shin YA (1968). Interaction of metal ions with polynucleotides and related compounds. XII. The relative effect of various metal ions on DNA helicity. *J Am Chem Soc*, 90(26):7323–8. doi:10.1021/ja01028a024 PMID:5725551

Elassouli SM, Alqahtani MH, Milaat W (2007). Genotoxicity of air borne particulates assessed by comet and the *Salmonella* mutagenicity test in Jeddah, Saudi Arabia. *Int J Environ Res Public Health*, 4(3):216–33. doi:10.3390/ijerph2007030004 PMID:17911660

EPA (2006). Report on air quality in nonattainment areas for 2003–2005 covering ozone, particulate matter, carbon monoxide, sulfur dioxide, nitrogen dioxide, and lead. Research Triangle Park (NC): US Environmental Protection Agency.

EPA (2009). Integrated science assessment for particulate matter (Final Report). EPA/600/R-08/139F. Washington (DC): US Environmental Protection Agency. Available from: http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=216546.

Epton MJ, Dawson RD, Brooks WM, Kingham S, Aberkane T, Cavanagh JA et al. (2008). The effect of ambient air pollution on respiratory health of school children: a panel study. *Environ Health*, 7(1):16 doi:10.1186/1476-069X-7-16 PMID:18479529

Erdinger L, Dürr M, Höpker KA (2005). Correlations between mutagenic activity of organic extracts of airborne particulate matter, NO_x and sulphur dioxide in southern Germany: results of a two-year study. *Environ Sci Pollut Res Int*, 12(1):10–20. doi:10.1065/espr2004.04.196 PMID:15768736

Espinosa-Aguirre JJ, Reyes RE, Rubio J, Ostrosky-Wegman P, Martinez G (1993). Mutagenic activity of urban air samples and its modulation by chili extracts. *Mutat Res*, 303(2):55–61. doi:10.1016/0165-7992(93)90095-D PMID:7692278

Evenson DP, Jost LK, Marshall D, Zinaman MJ, Clegg E, Purvis K et al. (1999). Utility of the sperm chromatin structure assay as a diagnostic and prognostic tool in the human fertility clinic. *Hum Reprod*, 14(4):1039–49. doi:10.1093/humrep/14.4.1039 PMID:10221239

Fabiani R, De Bartolomeo A, Rosignoli P, Morozzi G, Cecinato A, Balducci C (2008). Chemical and toxicological characterization of airborne total suspended particulate (TSP) and PM_{10} organic extracts. *Polycycl Aromat Compd*, 28(4–5):486–99. doi:10.1080/10406630802377948

Farina F, Sancini G, Mantecca P, Gallinotti D, Camatini M, Palestini P (2011). The acute toxic effects of particulate matter in mouse lung are related to size and season of collection. *Toxicol Lett*, 202(3):209–17. doi:10.1016/j.toxlet.2011.01.031 PMID:21371539

Ferreira MI, Domingos M, Gomes HA, Saldiva PH, de Assunção JV (2007). Evaluation of mutagenic potential of contaminated atmosphere at Ibirapuera Park, São Paulo - SP, Brazil, using the *Tradescantia* stamen-hair assay. *Environ Pollut*, 145(1):219–24. doi:10.1016/j.envpol.2006.03.013 PMID:16777295

Ferreira MI, Petrenko H, Lobo DJ, Rodrigues GS, Moreira A, Saldiva PH (2000). In situ monitoring of the mutagenic effects of the gaseous emissions of a solid waste incinerator in metropolitan São Paulo, Brazil, using the *Tradescantia* stamen-hair assay. *J Air Waste Manag Assoc*, 50(10):1852–6. doi:10.1080/10473289.2000.10464217 PMID:11288313

Ferreira MI, Rodrigues GS, Domingos M, Saldiva PHDN (2003). In situ monitoring of mutagenicity of air pollutants in São Paulo city using *Tradescantia*-SHM bioassay.

Braz Arch Biol Technol, 46(2):253–8. doi:10.1590/S1516-89132003000200017

Festy B, Coviaux F, Le Moullec Y, Juguet B, Person A, Courtois Y (1984). Potential toxicity of urban air contamination by inert particles [in French]. *Ann Pharm Fr*, 42(6):519–28. PMID:6535427

Festy LP (1980). Biological activity of organic extracts from atmospheric particles in urban air. *Pollut Atmos*, 22:50–7.

Flessel CP, Guirguis GN, Cheng JC, Chang K-I, Hahn ES, Twiss SA et al. (1985). Sources of mutagens in Contra Costa County community aerosols during pollution episodes: diurnal variations and relations to source emissions tracers. *Environ Int*, 11(2–4):293–301. doi:10.1016/0160-4120(85)90021-2

Fortoul TI, Rojas-Lemus M, Avila-Casado MC, Rodriguez-Lara V, Montaño LF, Muñoz-Comonfort A et al. (2010). Endogenous antioxidants and nasal human epithelium response to air pollutants: genotoxic and inmmuno-cytochemical evaluation. *J Appl Toxicol*, 30(7):661–5. doi:10.1002/jat.1538 PMID:20981858

Frampton MW, Ghio AJ, Samet JM, Carson JL, Carter JD, Devlin RB (1999). Effects of aqueous extracts of PM_{10} filters from the Utah Valley on human airway epithelial cells. *Am J Physiol*, 277(5 Pt 1):L960–7. PMID:10564181

Freedman AP, Robinson SE (1988). Noninvasive magnetopneumographic studies of lung dust retention and clearance in coal miners. In: Frantz RL, Ramani RV, editors. Respirable dust in the mineral industries: health effects, characterization and control. University Park (PA): Penn State University Press; pp. 181–6.

Fujii T, Hayashi S, Hogg JC, Mukae H, Suwa T, Goto Y et al. (2002). Interaction of alveolar macrophages and airway epithelial cells following exposure to particulate matter produces mediators that stimulate the bone marrow. *Am J Respir Cell Mol Biol*, 27(1):34–41. doi:10.1165/ajrcmb.27.1.4787 PMID:12091243

Fujii T, Hayashi S, Hogg JC, Vincent R, Van Eeden SF (2001). Particulate matter induces cytokine expression in human bronchial epithelial cells. *Am J Respir Cell Mol Biol*, 25(3):265–71. doi:10.1165/ajrcmb.25.3.4445 PMID:11588002

Gábelová A, Valovicová Z, Horváthová E, Slamenová D, Binková B, Srám RJ et al. (2004). Genotoxicity of environmental air pollution in three European cities: Prague, Kosice and Sofia. *Mutat Res*, 563(1):49–59. doi:10.1016/j.mrgentox.2004.05.014 PMID:15324748

Gábelová A, Valovicová Z, Lábaj J, Bacová G, Binková B, Farmer PB (2007). Assessment of oxidative DNA damage formation by organic complex mixtures from airborne particles PM_{10}. *Mutat Res*, 620(1–2):135–44. doi:10.1016/j.mrfmmm.2007.03.003 PMID:17403525

Galati R, Zijno A, Crebelli R, Falasca G, Tomei F, Iecher F et al. (2001). Detection of antibodies to the benzo(a) pyrene diol epoxide-DNA adducts in sera from individuals exposed to low doses of polycyclic aromatic hydrocarbons. *J Exp Clin Cancer Res*, 20(3):359–64. PMID:11718215

Gallagher JE, Jackson MA, George MH, Lewtas J (1990). Dose-related differences in DNA adduct levels in rodent tissues following skin application of complex mixtures from air pollution sources. *Carcinogenesis*, 11(1):63–8. doi:10.1093/carcin/11.1.63 PMID:2403860

García-Suástegui WA, Huerta-Chagoya A, Carrasco-Colín KL, Pratt MM, John K, Petrosyan P et al. (2011). Seasonal variations in the levels of PAH-DNA adducts in young adults living in Mexico City. *Mutagenesis*, 26(3):385–91. doi:10.1093/mutage/geq104 PMID:21193517

Garçon G, Dagher Z, Zerimech F, Ledoux F, Courcot D, Aboukais A et al. (2006). Dunkerque City air pollution particulate matter-induced cytotoxicity, oxidative stress and inflammation in human epithelial lung cells (L132) in culture. *Toxicol In Vitro*, 20(4):519–28. doi:10.1016/j.tiv.2005.09.012 PMID:16298102

Gavett SH, Haykal-Coates N, Highfill JW, Ledbetter AD, Chen LC, Cohen MD et al. (2003). World Trade Center fine particulate matter causes respiratory tract hyperresponsiveness in mice. *Environ Health Perspect*, 111(7):981–91. doi:10.1289/ehp.5931 PMID:12782502

Gerlofs-Nijland ME, Boere AJ, Leseman DL, Dormans JA, Sandström T, Salonen RO et al. (2005). Effects of particulate matter on the pulmonary and vascular system: time course in spontaneously hypertensive rats. *Part Fibre Toxicol*, 2(1):2 doi:10.1186/1743-8977-2-2 PMID:15813961

Gerlofs-Nijland ME, Dormans JA, Bloemen HJ, Leseman DL, John A, Boere F et al. (2007). Toxicity of coarse and fine particulate matter from sites with contrasting traffic profiles. *Inhal Toxicol*, 19(13):1055–69. doi:10.1080/08958370701626261 PMID:17957546

Ghio AJ, Devlin RB (2001). Inflammatory lung injury after bronchial instillation of air pollution particles. *Am J Respir Crit Care Med*, 164(4):704–8. doi:10.1164/ajrccm.164.4.2011089 PMID:11520740

Ghio AJ, Kim C, Devlin RB (2000). Concentrated ambient air particles induce mild pulmonary inflammation in healthy human volunteers. *Am J Respir Crit Care Med*, 162(3 Pt 1):981–8. doi:10.1164/ajrccm.162.3.9911115 PMID:10988117

Ghio AJ, Stonehuerner J, Dailey LA, Carter JD (1999). Metals associated with both the water-soluble and insoluble fractions of an ambient air pollution particle catalyze an oxidative stress. *Inhal Toxicol*, 11(1):37–49. doi:10.1080/089583799197258 PMID:10380158

Ghio AJ, Suliman HB, Carter JD, Abushamaa AM, Folz RJ (2002). Overexpression of extracellular superoxide dismutase decreases lung injury after exposure to oil fly ash. *Am J Physiol Lung Cell Mol Physiol*, 283(1):L211–8. doi:10.1152/ajplung.00409.2001 PMID:12060579

Gil L, Cáceres D, Adonis M (1997). Influence of atmospheric air pollution on indoor air quality: comparison

of chemical pollutants and mutagenicity levels in Santiago (Chile). *Indoor Built Environ*, 6(6):320–30. doi:10.1177/1420326X9700600602

Gilmour PS, Brown DM, Lindsay TG, Beswick PH, MacNee W, Donaldson K (1996). Adverse health effects of PM_{10} particles: involvement of iron in generation of hydroxyl radical. *Occup Environ Med*, 53(12):817–22. doi:10.1136/oem.53.12.817 PMID:8994401

Giovannelli L, Pitozzi V, Moretti S, Boddi V, Dolara P (2006). Seasonal variations of DNA damage in human lymphocytes: correlation with different environmental variables. *Mutat Res*, 593(1–2):143–52. doi:10.1016/j.mrfmmm.2005.07.002 PMID:16095632

Goldsmith CA, Frevert C, Imrich A, Sioutas C, Kobzik L (1997). Alveolar macrophage interaction with air pollution particulates. *Environ Health Perspect*, 105:Suppl 5: 1191–5. doi:10.1289/ehp.97105s51191 PMID:9400722

Gong J, Zhu T, Kipen H, Wang G, Hu M, Ohman-Strickland P et al. (2013). Malondialdehyde in exhaled breath condensate and urine as a biomarker of air pollution induced oxidative stress. *J Expo Sci Environ Epidemiol*, 23(3):322–7. doi:10.1038/jes.2012.127 PMID:23321859

Gordon T, Nadziejko C, Schlesinger R, Chen LC (1998). Pulmonary and cardiovascular effects of acute exposure to concentrated ambient particulate matter in rats. *Toxicol Lett*, 96–97(1–2):285–8. doi:10.1016/S0378-4274(98)00084-8 PMID:9820679

Goto S, Kato Y, Orii A (1982). Daily variation of mutagenicities of airborne particulates. *J Jpn Soc Air Pollut*, 17:295–303.

Greenberg A, Lwo J, Antherholt TB, Rosen R, Hartman T, Butler J et al. (1993). Bioassay-directed fractionation of organic compounds associated with airborne particulate matter: an interseasonal study. *Atmos Environ Part A Gen Top*, 27(10):1609–26. doi:10.1016/0960-1686(93)90160-Z

Greenwell LL, Moreno T, Jones TP, Richards RJ (2002). Particle-induced oxidative damage is ameliorated by pulmonary antioxidants. *Free Radic Biol Med*, 32(9):898–905. doi:10.1016/S0891-5849(02)00782-7 PMID:11978491

Gualtieri M, Ovrevik J, Mollerup S, Asare N, Longhin E, Dahlman HJ et al. (2011). Airborne urban particles (Milan winter-$PM_{2.5}$) cause mitotic arrest and cell death: effects on DNA, mitochondria, AhR binding and spindle organization. *Mutat Res*, 713(1–2):18–31. doi:10.1016/j.mrfmmm.2011.05.011 PMID:21645525

Gundel LA, Dalsey JM, De Carvalho LRF, Kado NY, Schuetzle D (1993). Polar organic matter in airborne particles: chemical characterization and mutagenic activity. *Environ Sci Technol*, 27(10):2112–9. doi:10.1021/es00047a017

Gupta P, Harger WP, Arey J (1996). The contribution of nitro- and methylnitronaphthalenes to the vapor-phase mutagenicity of ambient air samples. *Atmos Environ*, 30(18):3157–66. doi:10.1016/1352-2310(96)00024-6

Gurgueira SA, Lawrence J, Coull B, Murthy GG, González-Flecha B (2002). Rapid increases in the steady-state concentration of reactive oxygen species in the lungs and heart after particulate air pollution inhalation. *Environ Health Perspect*, 110(8):749–55. doi:10.1289/ehp.02110749 PMID:12153754

Gutiérrez-Castillo ME, Roubicek DA, Cebrián-García ME, De Vizcaya-Ruíz A, Sordo-Cedeño M, Ostrosky-Wegman P (2006). Effect of chemical composition on the induction of DNA damage by urban airborne particulate matter. *Environ Mol Mutagen*, 47(3):199–211. doi:10.1002/em.20186 PMID:16355389

Hadnagy W, Seemayer NH (1987). Comparative investigation on the genotoxicity of city smog and automobile exhaust particulates. *J Aerosol Sci*, 18(6):697–9. doi:10.1016/0021-8502(87)90100-5

Hadnagy W, Seemayer NH (1991). In vitro cytogenetic assays for the detection of mitotic aneuploidy by particulate pollutants. *Toxicol In Vitro*, 5(5–6):507–10. doi:10.1016/0887-2333(91)90082-O PMID:20732066

Hadnagy W, Seemayer NH, Tomingas R (1986). Cytogenetic effects of airborne particulate matter in human lymphocytes in vitro. *Mutat Res*, 175(2):97–101. doi:10.1016/0165-7992(86)90131-4 PMID:3762578

Hadnagy W, Seemayer NH, Tomingas R, Ivanfy K (1989). Comparative study of sister-chromatid exchanges and chromosomal aberrations induced by airborne particulates from an urban and a highly industrialized location in human lymphocyte cultures. *Mutat Res*, 225(1–2):27–32. doi:10.1016/0165-7992(89)90028-6 PMID:2913489

Halatek T, Stepnik M, Stetkiewicz J, Krajnow A, Kur B, Szymczak W et al. (2011). The inflammatory response in lungs of rats exposed on the airborne particles collected during different seasons in four European cities. *J Environ Sci Health A Tox Hazard Subst Environ Eng*, 46(13):1469–81. doi:10.1080/10978526.2011.609064 PMID:21961642

Hallare AV, Gervasio MKR, Gervasio PLG, Acacio-Claro PJB (2009). Monitoring genotoxicity among gasoline station attendants and traffic enforcers in the City of Manila using the micronucleus assay with exfoliated epithelial cells. *Environ Monit Assess*, 156(1–4):331–41. doi:10.1007/s10661-008-0488-y PMID:18712612

Hannigan MP, Busby WF Jr, Cass GR (2005). Source contributions to the mutagenicity of urban particulate air pollution. *J Air Waste Manag Assoc*, 55(4):399–410. doi:10.1080/10473289.2005.10464633 PMID:15887882

Hannigan MP, Cass GR, Penman BW, Crespi CL, Lafleur AL, Busby WF et al. (1997). Human cell mutagens in Los Angeles air. *Environ Sci Technol*, 31(2):438–47. doi:10.1021/es960266z

Hannigan MP, Cass GR, Penman BW, Crespi CL, Lafleur AL, Busby WF et al. (1998). Bioassay-directed chemical

analysis of Los Angeles airborne particulate matter using a human cell mutagenicity assay. *Environ Sci Technol*, 32(22):3502–14. doi:10.1021/es9706561

Hansen AM, Wallin H, Binderup ML, Dybdahl M, Autrup H, Loft S et al. (2004). Urinary 1-hydroxypyrene and mutagenicity in bus drivers and mail carriers exposed to urban air pollution in Denmark. *Mutat Res*, 557(1):7–17. doi:10.1016/j.mrgentox.2003.09.007 PMID:14706514

Happo MS, Salonen RO, Hälinen AI, Jalava PI, Pennanen AS, Kosma VM et al. (2007). Dose and time dependency of inflammatory responses in the mouse lung to urban air coarse, fine, and ultrafine particles from six European cities. *Inhal Toxicol*, 19(3):227–46. doi:10.1080/08958370601067897 PMID:17365027

Healey K, Lingard JJ, Tomlin AS, Hughes A, White KL, Wild CP et al. (2005). Genotoxicity of size-fractionated samples of urban particulate matter. *Environ Mol Mutagen*, 45(4):380–7. doi:10.1002/em.20105 PMID:15662658

Hebels DG, Jennen DG, van Herwijnen MH, Moonen EJ, Pedersen M, Knudsen LE et al. (2011). Whole-genome gene expression modifications associated with nitrosamine exposure and micronucleus frequency in human blood cells. *Mutagenesis*, 26(6):753–61. doi:10.1093/mutage/ger043 PMID:21724973

Heidenfelder BL, Reif DM, Harkema JR, Cohen Hubal EA, Hudgens EE, Bramble LA et al. (2009). Comparative microarray analysis and pulmonary changes in Brown Norway rats exposed to ovalbumin and concentrated air particulates. *Toxicol Sci*, 108(1):207–21. doi:10.1093/toxsci/kfp005 PMID:19176365

Hemminki K, Zhang LF, Krüger J, Autrup H, Törnqvist M, Norbeck HE (1994). Exposure of bus and taxi drivers to urban air pollutants as measured by DNA and protein adducts. *Toxicol Lett*, 72(1–3):171–4. doi:10.1016/0378-4274(94)90025-6 PMID:7515516

Herbstman JB, Tang D, Zhu D, Qu L, Sjödin A, Li Z et al. (2012). Prenatal exposure to polycyclic aromatic hydrocarbons, benzo[*a*]pyrene-DNA adducts, and genomic DNA methylation in cord blood. *Environ Health Perspect*, 120(5):733–8. doi:10.1289/ehp.1104056 PMID:22256332

Heuser VD, da Silva J, Moriske HJ, Dias JF, Yoneama ML, de Freitas TR (2002). Genotoxicity biomonitoring in regions exposed to vehicle emissions using the comet assay and the micronucleus test in native rodent *Ctenomys minutus*. *Environ Mol Mutagen*, 40(4):227–35. doi:10.1002/em.10115 PMID:12489112

Hoek G, Krishnan RM, Beelen R, Peters A, Ostro B, Brunekreef B et al. (2013). Long-term air pollution exposure and cardio-respiratory mortality: a review. *Environ Health*, 12(1):43 doi:10.1186/1476-069X-12-43 PMID:23714370

Holloway JW, Savarimuthu Francis S, Fong KM, Yang IA (2012). Genomics and the respiratory effects of air pollution exposure. *Respirology*, 17(4):590–600. doi:10.1111/j.1440-1843.2012.02164.x PMID:22404320

Hornberg C, Maciuleviciute L, Seemayer NH (1996). Sister chromatid exchanges in rodent tracheal epithelium exposed in vitro to environmental pollutants. *Toxicol Lett*, 88(1–3):45–53. doi:10.1016/0378-4274(96)03717-4 PMID:8920716

Hornberg C, Maciuleviciute L, Seemayer NH (1997). Comparative analysis of cyto- and genotoxic effects of airborne particulates on human and rodent respiratory cells in vitro. *Toxicol In Vitro*, 11(5):711–5. doi:10.1016/S0887-2333(97)00065-9 PMID:20654375

Hornberg C, Maciuleviciute L, Seemayer NH, Kainka E (1998). Induction of sister chromatid exchanges (SCE) in human tracheal epithelial cells by the fractions PM_{10} and $PM_{2.5}$ of airborne particulates. *Toxicol Lett*, 96–97(1–2):215–20. doi:10.1016/S0378-4274(98)00075-7 PMID:9820670

Hornberg C, Seemayer NH (1995). Induction of sister chromatid exchanges in rodent tracheal epithelial cells as a sensitive bioassay for detection of genotoxic activity of airborne particulates. *Exp Toxicol Pathol*, 47(4):241–3. doi:10.1016/S0940-2993(11)80257-5 PMID:8855119

Hosgood HD 3rd, Berndt SI, Lan Q (2007). GST genotypes and lung cancer susceptibility in Asian populations with indoor air pollution exposures: a meta-analysis. *Mutat Res*, 636(1–3):134–43. doi:10.1016/j.mrrev.2007.02.002 PMID:17428724

Hou L, Wang S, Dou C, Zhang X, Yu Y, Zheng Y et al. (2012). Air pollution exposure and telomere length in highly exposed subjects in Beijing, China: a repeated-measure study. *Environ Int*, 48:71–7. doi:10.1016/j.envint.2012.06.020 PMID:22871507

Hou L, Zhang X, Tarantini L, Nordio F, Bonzini M, Angelici L et al. (2011). Ambient PM exposure and DNA methylation in tumor suppressor genes: a cross-sectional study. *Part Fibre Toxicol*, 8(1):25 doi:10.1186/1743-8977-8-25 PMID:21878113

Hoxha M, Dioni L, Bonzini M, Pesatori AC, Fustinoni S, Cavallo D et al. (2009). Association between leukocyte telomere shortening and exposure to traffic pollution: a cross-sectional study on traffic officers and indoor office workers. *Environ Health*, 8(1):41 doi:10.1186/1476-069X-8-41 PMID:19772576

Hsiao WL, Mo ZY, Fang M, Shi XM, Wang F (2000). Cytotoxicity of $PM_{2.5}$ and $PM_{2.5-10}$ ambient air pollutants assessed by the MTT and the Comet assays. *Mutat Res*, 471(1–2):45–55. doi:10.1016/S1383-5718(00)00116-9 PMID:11080660

Huang YC, Schmitt M, Yang Z, Que LG, Stewart JC, Frampton MW et al. (2010). Gene expression profile in circulating mononuclear cells after exposure to ultrafine carbon particles. *Inhal Toxicol*, 22(10):835–46. doi:10.3109/08958378.2010.486419 PMID:20507211

Huang W, Wang G, Lu SE, Kipen H, Wang Y, Hu M et al. (2012). Inflammatory and oxidative stress responses of

healthy young adults to changes in air quality during the Beijing Olympics. *Am J Respir Crit Care Med*, 186(11):1150–9. doi:10.1164/rccm.201205-0850OC PMID:22936356

Huang YC, Karoly ED, Dailey LA, Schmitt MT, Silbajoris R, Graff DW et al. (2011). Comparison of gene expression profiles induced by coarse, fine, and ultrafine particulate matter. *J Toxicol Environ Health A*, 74(5):296–312. doi:10.1080/15287394.2010.516238 PMID:21240730

Huttunen K, Siponen T, Salonen I, Yli-Tuomi T, Aurela M, Dufva H et al. (2012). Low-level exposure to ambient particulate matter is associated with systemic inflammation in ischemic heart disease patients. *Environ Res*, 116:44–51. doi:10.1016/j.envres.2012.04.004 PMID:22541720

IARC (1982). Some industrial chemicals and dyestuffs. *IARC Monogr Eval Carcinog Risk Chem Hum*. 29:1–398. Available from: http://monographs.iarc.fr/ENG/Monographs/vol1-42/mono29.pdf. PMID:6957379

IARC (1984). Polynuclear aromatic hydrocarbons, Part 2, carbon blacks, mineral oils (lubricant base oils and derived products) and some nitroarenes. *IARC Monogr Eval Carcinog Risk Chem Hum*. 33:1–222. Available from: http://monographs.iarc.fr/ENG/Monographs/vol1-42/mono33.pdf. PMID:6590450

IARC (1989). Diesel and gasoline engine exhausts and some nitroarenes. *IARC Monogr Eval Carcinog Risks Hum*, 46:1–458. Available from: http://monographs.iarc.fr/ENG/Monographs/vol46/index.php. PMID:2483415

IARC (1992). Occupational exposures to mists and vapours from strong inorganic acids and other industrial chemicals. *IARC Monogr Eval Carcinog Risks Hum*, 54:1–310. Available from: http://monographs.iarc.fr/ENG/Monographs/vol54/index.php. PMID:1345371

IARC (2006). Formaldehyde, 2-butoxyethanol and 1-tert-butoxypropan-2-ol. *IARC Monogr Eval Carcinog Risks Hum*, 88:1–478. Available from: http://monographs.iarc.fr/ENG/Monographs/vol88/index.php. PMID:17366697

IARC (2010a). Carbon black, titanium dioxide, and talc. *IARC Monogr Eval Carcinog Risks Hum*, 93:1–413. Available from: http://monographs.iarc.fr/ENG/Monographs/vol93/index.php. PMID:21449489

IARC (2010b). Household use of solid fuels and high-temperature frying. *IARC Monogr Eval Carcinog Risks Hum*, 95:1–430. Available from: http://monographs.iarc.fr/ENG/Monographs/vol95/index.php. PMID:20701241

IARC (2010c). Some non-heterocyclic polycyclic aromatic hydrocarbons and some related exposures. *IARC Monogr Eval Carcinog Risks Hum*, 92:1–853. Available from: http://monographs.iarc.fr/ENG/Monographs/vol92/index.php. PMID:21141735

IARC (2012a). Arsenic, metals, fibres, and dusts. *IARC Monogr Eval Carcinog Risks Hum*, 100C:1–499. Available from: http://monographs.iarc.fr/ENG/Monographs/vol100C/index.php. PMID:23189751

IARC (2012b). Chemical agents and related occupations. *IARC Monogr Eval Carcinog Risks Hum*, 100F:1–599. Available from: http://monographs.iarc.fr/ENG/Monographs/vol100F/index.php. PMID:23189753

IARC (2012c). Personal habits and indoor combustions. *IARC Monogr Eval Carcinog Risks Hum*, 100E:1–575. Available from: http://monographs.iarc.fr/ENG/Monographs/vol100E/index.php. PMID:23193840

IARC (2013). Diesel and gasoline engine exhausts and some nitroarenes. *IARC Monogr Eval Carcinog Risks Hum*, 105:1–704. Available from: http://monographs.iarc.fr/ENG/Monographs/vol105/index.php.

Ichinose T, Yoshida S, Sadakane K, Takano H, Yanagisawa R, Inoue K et al. (2008). Effects of Asian sand dust, Arizona sand dust, amorphous silica and aluminum oxide on allergic inflammation in the murine lung. *Inhal Toxicol*, 20(7):685–94. doi:10.1080/08958370801935133 PMID:18464056

ICRP (1994). Human respiratory tract model for radiological protection. A report of a task group of the International Commission on Radiological Protection (ICRP Publication 66). *Ann ICRP*, 24(1–3).

Ieradi LA, Cristaldi M, Mascanzoni D, Cardarelli E, Grossi R, Campanella L (1996). Genetic damage in urban mice exposed to traffic pollution. *Environ Pollut*, 92(3):323–8. doi:10.1016/0269-7491(95)00109-3 PMID:15091385

Imlay JA, Chin SM, Linn S (1988). Toxic DNA damage by hydrogen peroxide through the Fenton reaction in vivo and in vitro. *Science*, 240(4852):640–2. doi:10.1126/science.2834821 PMID:2834821

Imrich A, Ning Y, Lawrence J, Coull B, Gitin E, Knutson M et al. (2007). Alveolar macrophage cytokine response to air pollution particles: oxidant mechanisms. *Toxicol Appl Pharmacol*, 218(3):256–64. doi:10.1016/j.taap.2006.11.033 PMID:17222881

IPCS (2008). Principles for evaluating health risks in children associated with exposure to chemicals. *Environ Health Criteria*, No. 237. Geneva, Switzerland: International Programme on Chemical Safety. Available from: http://www.inchem.org/documents/ehc/ehc/ehc237.pdf.

Ishikawa H, Tian Y, Piao F, Duan Z, Zhang Y, Ma M et al. (2006). Genotoxic damage in female residents exposed to environmental air pollution in Shenyang city, China. *Cancer Lett*, 240(1):29–35. doi:10.1016/j.canlet.2005.08.023 PMID:16246488

Israël GW, Busing JB (1983). Investigations on the mutagenic action of urban particulates. *J Aerosol Sci*, 14(3):192–4. doi:10.1016/0021-8502(83)90025-3

Izzotti A, Camoirano A, D'Agostini F, Sciacca S, De Naro Papa F, Cesarone CF et al. (1996). Biomarker alterations produced in rat lung by intratracheal instillations of air particulate extracts and chemoprevention

with oral *N*-acetylcysteine. *Cancer Res*, 56(7):1533–8. PMID:8603398

Jadczyk P, Kucharczyk J (2005). Mutagenicity of pollutants and their fractions adsorbed on airborne particulate in the centre of Wrocław (Poland). *Environ Prot Eng*, 31(2):77–92.

Jalava P, Salonen RO, Hälinen AI, Sillanpää M, Sandell E, Hirvonen MR (2005). Effects of sample preparation on chemistry, cytotoxicity, and inflammatory responses induced by air particulate matter. *Inhal Toxicol*, 17(2):107–17. doi:10.1080/08958370590899550 PMID:15764488

Jansen KL, Larson TV, Koenig JQ, Mar TF, Fields C, Stewart J et al. (2005). Associations between health effects and particulate matter and black carbon in subjects with respiratory disease. *Environ Health Perspect*, 113(12):1741–6. doi:10.1289/ehp.8153 PMID:16330357

Janssen BG, Godderis L, Pieters N, Poels K, Kiciński M, Cuypers A et al. (2013). Placental DNA hypomethylation in association with particulate air pollution in early life. *Part Fibre Toxicol*, 10(1):22 doi:10.1186/1743-8977-10-22 PMID:23742113

Jaques PA, Kim CS (2000). Measurement of total lung deposition of inhaled ultrafine particles in healthy men and women. *Inhal Toxicol*, 12(8):715–31. doi:10.1080/08958370050085156 PMID:10880153

Jayasekher T (2009). Aerosols near by a coal fired thermal power plant: chemical composition and toxic evaluation. *Chemosphere*, 75(11):1525–30. doi:10.1016/j.chemosphere.2009.02.001 PMID:19264341

Jerrett M, Burnett RT, Beckerman BS, Turner MC, Krewski D, Thurston G et al. (2013). Spatial analysis of air pollution and mortality in California. *Am J Respir Crit Care Med*, 188(5):593–9. doi:10.1164/rccm.201303-0609OC PMID:23805824

Jiang LP, Dai H, Sun Q, Geng CY, Yang Y, Wu T et al. (2011). Ambient particulate matter on DNA damage in HepG2 cells. *Toxicol Ind Health*, 27(1):87–95. doi:10.1177/0748233710387001 PMID:20947658

Kadiiska MB, Ghio AJ, Mason RP (2004). ESR investigation of the oxidative damage in lungs caused by asbestos and air pollution particles. *Spectrochim Acta A Mol Biomol Spectrosc*, 60(6):1371–7. doi:10.1016/j.saa.2003.10.035 PMID:15134737

Kado NY, Guirguis GN, Flessel CP, Chan RC, Chang KI, Wesolowski JJ (1986). Mutagenicity of fine (less than 2.5 microns) airborne particles: diurnal variation in community air determined by a *Salmonella* micro preincubation (microsuspension) procedure. *Environ Mutagen*, 8(1):53–66. doi:10.1002/em.2860080106 PMID:3510862

Kado NY, Langley D, Eisenstadt E (1983). A simple modification of the *Salmonella* liquid-incubation assay. Increased sensitivity for detecting mutagens in human urine. *Mutat Res*, 121(1):25–32. doi:10.1016/0165-7992(83)90082-9 PMID:6306458

Kamdar O, Le W, Zhang J, Ghio AJ, Rosen GD, Upadhyay D (2008). Air pollution induces enhanced mitochondrial oxidative stress in cystic fibrosis airway epithelium. *FEBS Lett*, 582(25–26):3601–6. doi:10.1016/j.febslet.2008.09.030 PMID:18817777

Kameda T, Inazu K, Bandow H Sanukida S, Maeda Y (2004). Diurnal change of direct-acting mutagenicity of soluble organic fraction of airborne particles collected at Southern Osaka: correlation between the mutagenicity, particles-associated nitroarenes, and gaseous emission. *Atmos Environ*, 38(13):1903–12. doi:10.1016/j.atmosenv.2004.01.007

Karahalil B, Karakaya AE, Burgaz S (1999). The micronucleus assay in exfoliated buccal cells: application to occupational exposure to polycyclic aromatic hydrocarbons. *Mutat Res*, 442(1):29–35. doi:10.1016/S1383-5718(99)00055-8 PMID:10366770

Karlsson HL, Holgersson A, Möller L (2008). Mechanisms related to the genotoxicity of particles in the subway and from other sources. *Chem Res Toxicol*, 21(3):726–31. doi:10.1021/tx7003568 PMID:18260651

Karlsson HL, Ljungman AG, Lindbom J, Möller L (2006). Comparison of genotoxic and inflammatory effects of particles generated by wood combustion, a road simulator and collected from street and subway. *Toxicol Lett*, 165(3):203–11. doi:10.1016/j.toxlet.2006.04.003 PMID:16716543

Karlsson HL, Nilsson L, Möller L (2005). Subway particles are more genotoxic than street particles and induce oxidative stress in cultured human lung cells. *Chem Res Toxicol*, 18(1):19–23. doi:10.1021/tx049723c PMID:15651844

Karlsson HL, Nygren J, Möller L (2004). Genotoxicity of airborne particulate matter: the role of cell-particle interaction and of substances with adduct-forming and oxidizing capacity. *Mutat Res*, 565(1):1–10. doi:10.1016/j.mrgentox.2004.07.015 PMID:15576234

Kawanaka Y, Matsumoto E, Sakamoto K, Wang N, Yun S-J (2004). Size distributions of mutagenic compounds and mutagenicity in atmospheric particulate matter collected with a low-pressure cascade impactor. *Atmos Environ*, 38(14):2125–32. doi:10.1016/j.atmosenv.2004.01.021

Kawanishi S, Inoue S, Yamamoto K (1989). Site-specific DNA damage induced by nickel(II) ion in the presence of hydrogen peroxide. *Carcinogenesis*, 10(12):2231–5. doi:10.1093/carcin/10.12.2231 PMID:2686851

Kelvin EA, Edwards S, Jedrychowski W, Schleicher RL, Camann D, Tang D et al. (2009). Modulation of the effect of prenatal PAH exposure on PAH-DNA adducts in cord blood by plasma antioxidants. *Cancer Epidemiol Biomarkers Prev*, 18(8):2262–8. doi:10.1158/1055-9965.EPI-09-0316 PMID:19661084

Ketelslegers HB, Gottschalk RW, Koppen G, Schoeters G, Baeyens WF, van Larebeke NA et al. (2008). Multiplex genotyping as a biomarker for susceptibility

to carcinogenic exposure in the FLEHS biomonitoring study. *Cancer Epidemiol Biomarkers Prev*, 17(8):1902–12. doi:10.1158/1055-9965.EPI-08-0045 PMID:18708379

Kim CS, Hu SC (1998). Regional deposition of inhaled particles in human lungs: comparison between men and women. *J Appl Physiol (1985)*, 84(6):1834–44. PMID:9609774

Kimura KC, Fukumasu H, Chaible LM, Lima CE, Horst MA, Matsuzaki P et al. (2010). Evaluation of DNA damage by the alkaline comet assay of the olfactory and respiratory epithelia of dogs from the city of São Paulo, Brazil. *Exp Toxicol Pathol*, 62(3):209–19. doi:10.1016/j.etp.2009.03.008 PMID:19447591

Knaapen AM, Shi T, Borm PJA, Schins RPF (2002). Soluble metals as well as the insoluble particle fraction are involved in cellular DNA damage induced by particulate matter. *Mol Cell Biochem*, 234(–):235(1–2):317–26. doi:10.1023/A:1015970023889 PMID:12162450

Knudsen LE, Norppa H, Gamborg MO, Nielsen PS, Okkels H, Soll-Johanning H et al. (1999). Chromosomal aberrations in humans induced by urban air pollution: influence of DNA repair and polymorphisms of glutathione *S*-transferase M1 and *N*-acetyltransferase 2. *Cancer Epidemiol Biomarkers Prev*, 8(4 Pt 1):303–10. PMID:10207633

Kodavanti UP, Mebane R, Ledbetter A, Krantz T, McGee J, Jackson MC et al. (2000). Variable pulmonary responses from exposure to concentrated ambient air particles in a rat model of bronchitis. *Toxicol Sci*, 54(2):441–51. doi:10.1093/toxsci/54.2.441 PMID:10774827

Koenig JQ, Jansen K, Mar TF, Lumley T, Kaufman J, Trenga CA et al. (2003). Measurement of offline exhaled nitric oxide in a study of community exposure to air pollution. *Environ Health Perspect*, 111(13):1625–9. doi:10.1289/ehp.6160 PMID:14527842

Koenig JQ, Mar TF, Allen RW, Jansen K, Lumley T, Sullivan JH et al. (2005). Pulmonary effects of indoor- and outdoor-generated particles in children with asthma. *Environ Health Perspect*, 113(4):499–503. doi:10.1289/ehp.7511 PMID:15811822

Kong XH, Yi XJ, Wang H (1994). **A** study on mutagenicity of NO_2-PAHs in airborne particulates in Shenyang. *J Health Toxicol*, 8:160–3.

Kooter I, Pennings J, Opperhuizen A, Cassee F (2005). Gene expression pattern in spontaneously hypertensive rats exposed to urban particulate matter (EHC-93). *Inhal Toxicol*, 17(1):53–65. doi:10.1080/08958370590885717 PMID:15764483

Kooter IM, Boere AJ, Fokkens PH, Leseman DL, Dormans JA, Cassee FR (2006). Response of spontaneously hypertensive rats to inhalation of fine and ultrafine particles from traffic: experimental controlled study. *Part Fibre Toxicol*, 3(1):7 doi:10.1186/1743-8977-3-7 PMID:16700918

Koppen G, Verheyen G, Maes A, Van Gorp U, Schoeters G, Hond ED et al. (2007). A battery of DNA effect biomarkers to evaluate environmental exposure of Flemish adolescents. *J Appl Toxicol*, 27(3):238–46. doi:10.1002/jat.1174 PMID:17226746

Koshy L, Jones T, BéruBé K (2009). Characterization and bioreactivity of respirable airborne particles from a municipal landfill. *Biomarkers*, 14:Suppl 1: 49–53. doi:10.1080/13547500902965351 PMID:19604059

Krewski D, Jerrett M, Burnett RT, Ma R, Hughes E, Shi Y et al. (2009). Extended follow-up and spatial analysis of the American Cancer Society study linking particulate air pollution and mortality. *Res Rep Health Eff Inst*, 140(140):5–114, discussion 115–36. PMID:19627030

Krishna G, Nath J, Ong T (1984). Correlative genotoxicity studies of airborne particles in *Salmonella typhimurium* and cultured human lymphocytes. *Environ Mutagen*, 6(4):585–92. doi:10.1002/em.2860060411 PMID:6381042

Krishna G, Nath J, Soler L, Ong T (1986). Comparative in vivo and in vitro genotoxicity studies of airborne particle extract in mice. *Mutat Res*, 171(2–3):157–63. doi:10.1016/0165-1218(86)90049-2 PMID:3528836

Kuo CY, Cheng YW, Chen CY, Lee H (1998). Correlation between the amounts of polycyclic aromatic hydrocarbons and mutagenicity of airborne particulate samples from Taichung City, Taiwan. *Environ Res*, 78(1):43–9. doi:10.1006/enrs.1998.3838 PMID:9630444

Kure EH, Andreassen A, Ovrebø S, Grzybowska E, Fiala Z, Strózyk M et al. (1997). Benzo(a)pyrene-albumin adducts in humans exposed to polycyclic aromatic hydrocarbons in an industrial area of Poland. *Occup Environ Med*, 54(9):662–6. doi:10.1136/oem.54.9.662 PMID:9423579

Lai HK, Tsang H, Wong CM (2013). Meta-analysis of adverse health effects due to air pollution in Chinese populations. *BMC Public Health*, 13(1):360 doi:10.1186/1471-2458-13-360 PMID:23594435

Larsson BM, Grunewald J, Sköld CM, Lundin A, Sandström T, Eklund A et al. (2010). Limited airway effects in mild asthmatics after exposure to air pollution in a road tunnel. *Respir Med*, 104(12):1912–8. doi:10.1016/j.rmed.2010.06.014 PMID:20621461

Lazarová M, Slamenová D (2004). Genotoxic effects of a complex mixture adsorbed onto ambient air particles on human cells in vitro; the effects of vitamins E and C. *Mutat Res*, 557(2):167–75. doi:10.1016/j.mrgentox.2003.10.011 PMID:14729371

Lee H, Su SY, Liu KS, Chou MC (1994). Correlation between meteorological conditions and mutagenicity of airborne particulate samples in a tropical monsoon climate area from Kaohsiung City, Taiwan. *Environ Mol Mutagen*, 23(3):200–7. doi:10.1002/em.2850230309 PMID:8162895

Lei XF, Wang L, Xun Z (1993). Study of the genotoxicity and concentration of five metals in airborne particles. *Chin J Environ Sci*, 14:30–3.

Lemos AT, Coronas MV, Rocha JAV, Vargas VMF (2012). Mutagenicity of particulate matter fractions in areas under the impact of urban and industrial activities. *Chemosphere*, 89(9):1126–34. doi:10.1016/j.chemosphere.2012.05.100 PMID:22795069

Li N, Xia T, Nel AE (2008). The role of oxidative stress in ambient particulate matter-induced lung diseases and its implications in the toxicity of engineered nanoparticles. *Free Radic Biol Med*, 44(9):1689–99. doi:10.1016/j.freeradbiomed.2008.01.028 PMID:18313407

Li PK, Gao ZY, Jiang RF, Gai BB, Qin YQ, Song WM (2010). DNA damage in population exposed to fine particulate [in Chinese]. *J Environ Occup Med*, 27:254–6. Available from: http://www.cnki.net/kcms/detail/detail.aspx?filename=LDYX201005004&dbcode=CJFQ&dbname=CJFD2010.

Li Y, Rittenhouse-Olson K, Scheider WL, Mu L (2012). Effect of particulate matter air pollution on C-reactive protein: a review of epidemiologic studies. *Rev Environ Health*, 27(2–3):133–49. doi:10.1515/reveh-2012-0012 PMID:23023922

Li Z, Hyseni X, Carter JD, Soukup JM, Dailey LA, Huang YC (2006). Pollutant particles enhanced H_2O_2 production from NAD(P)H oxidase and mitochondria in human pulmonary artery endothelial cells. *Am J Physiol Cell Physiol*, 291(2):C357–65. doi:10.1152/ajpcell.00365.2005 PMID:16571865

Lim SS, Vos T, Flaxman AD, Danaei G, Shibuya K, Adair-Rohani H et al. (2012). A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990–2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet*, 380(9859):2224–60. doi:10.1016/S0140-6736(12)61766-8 PMID:23245609

Lin ZQ, Xi ZG, Yang DF, Chao FH, Zhang HS, Zhang W et al. (2009). Oxidative damage to lung tissue and peripheral blood in endotracheal $PM_{2.5}$-treated rats. *Biomed Environ Sci*, 22(3):223–8. doi:10.1016/S0895-3988(09)60049-0 PMID:19725465

Lindbom J, Gustafsson M, Blomqvist G, Dahl A, Gudmundsson A, Swietlicki E et al. (2007). Wear particles generated from studded tires and pavement induces inflammatory reactions in mouse macrophage cells. *Chem Res Toxicol*, 20(6):937–46. doi:10.1021/tx700018z PMID:17516662

Lingard JJN, Tomlin AS, Clarke AG, Healey K, Hay AWM, Wild CP et al. (2005). A study of trace metal concentration of urban airborne particulate matter and its role in free radical activity as measured by plasmid strand break assay. *Atmos Environ*, 39(13):2377–84. doi:10.1016/j.atmosenv.2004.05.063

Liu L, Poon R, Chen L, Frescura AM, Montuschi P, Ciabattoni G et al. (2009a). Acute effects of air pollution on pulmonary function, airway inflammation, and oxidative stress in asthmatic children. *Environ Health Perspect*, 117(4):668–74. doi:10.1289/ehp.11813 PMID:19440509

Liu L, Ruddy T, Dalipaj M, Poon R, Szyszkowicz M, You H et al. (2009b). Effects of indoor, outdoor, and personal exposure to particulate air pollution on cardiovascular physiology and systemic mediators in seniors. *J Occup Environ Med*, 51(9):1088–98. doi:10.1097/JOM.0b013e3181b35144 PMID:19701101

Liu L, Ruddy TD, Dalipaj M, Szyszkowicz M, You H, Poon R et al. (2007). Influence of personal exposure to particulate air pollution on cardiovascular physiology and biomarkers of inflammation and oxidative stress in subjects with diabetes. *J Occup Environ Med*, 49(3):258–65. doi:10.1097/JOM.0b013e31803220ef PMID:17351511

Ljubimova JY, Kleinman MT, Karabalin NM, Inoue S, Konda B, Gangalum P et al. (2013). Gene expression changes in rat brain after short and long exposures to particulate matter in Los Angeles basin air: comparison with human brain tumors. *Exp Toxicol Pathol*, 65(7–8):1063–71. doi:10.1016/j.etp.2013.04.002 PMID:23688656

Löfroth G, Lazaridis G, Agurell E (1985). The use of the *Salmonella*/microsome mutagenicity test for the characterization of organic extracts from ambient air particulate matter. *Environ Int*, 11(2–4):161–7. doi:10.1016/0160-4120(85)90009-1

Longhin E, Pezzolato E, Mantecca P, Holme JA, Franzetti A, Camatini M et al. (2013). Season linked responses to fine and quasi-ultrafine Milan PM in cultured cells. *Toxicol In Vitro*, 27(2):551–9. doi:10.1016/j.tiv.2012.10.018 PMID:23159502

Ma TH, Cabrera GL, Chen R, Gill BS, Sandhu SS, Vandenberg AL et al. (1994). *Tradescantia* micronucleus bioassay. *Mutat Res*, 310(2):221–30. doi:10.1016/0027-5107(94)90115-5 PMID:7523893

Madrigano J, Baccarelli A, Mittleman MA, Sparrow D, Spiro A 3rd, Vokonas PS et al. (2012). Air pollution and DNA methylation: interaction by psychological factors in the VA Normative Aging Study. *Am J Epidemiol*, 176(3):224–32. doi:10.1093/aje/kwr523 PMID:22798479

Madrigano J, Baccarelli A, Mittleman MA, Wright RO, Sparrow D, Vokonas PS et al. (2011). Prolonged exposure to particulate pollution, genes associated with glutathione pathways, and DNA methylation in a cohort of older men. *Environ Health Perspect*, 119(7):977–82. doi:10.1289/ehp.1002773 PMID:21385671

Manney S, Meddings CM, Harrison RM, Mansur AH, Karakatsani A, Analitis A et al. (2012). Association between exhaled breath condensate nitrate + nitrite levels with ambient coarse particle exposure in subjects with airways disease. *Occup Environ Med*, 69(9):663–9. doi:10.1136/oemed-2011-100255 PMID:22767867

Mao DJ, Shi YM, Zhou YF,, Ding DH, Ma YS, Peng LH et al. (1993). The study of sperm abnormality in mice exposed to organic extracts of total suspended particulates in Shanghai. *Carcinog Teratog Mutagen*, 5:9

Mar TF, Jansen K, Shepherd K, Lumley T, Larson TV, Koenig JQ (2005). Exhaled nitric oxide in children with asthma and short-term $PM_{2.5}$ exposure in Seattle. *Environ Health Perspect*, 113(12):1791–4. doi:10.1289/ehp.7883 PMID:16330366

Martin S, Dawidowski L, Mandalunis P, Cereceda-Balic F, Tasat DR (2007). Characterization and biological effect of Buenos Aires urban air particles on mice lungs. *Environ Res*, 105(3):340–9. doi:10.1016/j.envres.2007.04.009 PMID:17628521

Martin S, Fernandez-Alanis E, Delfosse V, Evelson P, Yakisich JS, Saldiva PH et al. (2010). Low doses of urban air particles from Buenos Aires promote oxidative stress and apoptosis in mice lungs. *Inhal Toxicol*, 22(13):1064–71. doi:10.3109/08958378.2010.523030 PMID:21047167

Massolo L, Müller A, Tueros M, Rehwagen M, Franck U, Ronco A et al. (2002). Assessment of mutagenicity and toxicity of different-size fractions of air particulates from La Plata, Argentina, and Leipzig, Germany. *Environ Toxicol*, 17(3):219–31. doi:10.1002/tox.10054 PMID:12112630

Matsumoto Y, Sakai S, Kato T, Nakajima T, Satoh H (1998). Long-term trends of particulate mutagenic activity in atmosphere of Sapporo. 1. Determination of mutagenic activity by the conventional tester strains TA98 and TA100 during an 18-year period (1974–1992). *Environ Sci Technol*, 32(18):2665–71. doi:10.1021/es9801036

Maynard AM, Kuempel ED (2005). Airborne nanostructured particles and occupational health. *J Nanopart Res*, 7(6):587–614. doi:10.1007/s11051-005-6770-9

McCracken J, Baccarelli A, Hoxha M, Dioni L, Melly S, Coull B et al. (2010). Annual ambient black carbon associated with shorter telomeres in elderly men: Veterans Affairs Normative Aging Study. *Environ Health Perspect*, 118(11):1564–70. doi:10.1289/ehp.0901831 PMID:21465749

Melville GN (1970). Changes in specific airway conductance in healthy volunteers following nasal and oral inhalation of SO_2. *West Indian Med J*, 19(4):231–5. PMID:5504408

Mendez R, Zheng Z, Fan Z, Rajagopalan S, Sun Q, Zhang K (2013). Exposure to fine airborne particulate matter induces macrophage infiltration, unfolded protein response, and lipid deposition in white adipose tissue. *Am J Transl Res*, 5(2):224–34. PMID:23573366

Meng Z, Zhang Q (2007). Damage effects of dust storm $PM_{2.5}$ on DNA in alveolar macrophages and lung cells of rats. *Food Chem Toxicol*, 45(8):1368–74. doi:10.1016/j.fct.2007.01.014 PMID:17336437

Meng ZQ, Zhang QX (2005). Damage of atmospheric fine particles on DNA in alveolar macrophages of rats *Chin Environ Sci*, 25(1):15–7.

Meng ZQ, Zhang QX (2006b). Damaging effects of dust storm fine particles instillation on DNA in lung cells of rats [in Chinese]. *Chin J Publ Health*, 22:1458–9. Available from: http://www.cnki.net/kcms/detail/detail.aspx?filename=ZGGW200612027&dbcode=CJFQ&dbname=cjfd2006.

Meng ZQ, Zhang QX, Geng H (2006a). DNA damage effects of fine particles from sandstorm in rat alveolar macrophages [in Chinese]. *J Environ Occup Med*, 23:185–8. Available from: http://www.cnki.net/.

Mercer RR, Crapo JD (1989). Anatomical modeling of microdosimetry of inhaled particles and gases in the lung. In: Crapo JD, Smolko ED, Miller FJ, Graham JA, Hayes AW, editors. Extrapolation of dosimetry relationships for inhaled particles and gases. San Diego (CA): Academic Press; pp. 69–78.

Merlo F, Bolognesi C, Peluso M, Valerio F, Abbondandolo A, Puntoni R (1997). Airborne levels of polycyclic aromatic hydrocarbons: ^{32}P-postlabeling DNA adducts and micronuclei in white blood cells from traffic police workers and urban residents. *J Environ Pathol Toxicol Oncol*, 16(2–3):157–62. PMID:9275996

Miller FJ (1995). Uptake and fate of ozone in the respiratory tract. *Toxicol Lett*, 82–83:277–85. doi:10.1016/0378-4274(95)03562-1 PMID:8597066

Miller FJ, Overton JH Jr, Jaskot RH, Menzel DB (1985). A model of the regional uptake of gaseous pollutants in the lung. I. The sensitivity of the uptake of ozone in the human lung to lower respiratory tract secretions and exercise. *Toxicol Appl Pharmacol*, 79(1):11–27. doi:10.1016/0041-008X(85)90364-3 PMID:3840292

Mills NL, Robinson SD, Fokkens PH, Leseman DL, Miller MR, Anderson D et al. (2008). Exposure to concentrated ambient particles does not affect vascular function in patients with coronary heart disease. *Environ Health Perspect*, 116(6):709–15. doi:10.1289/ehp.11016 PMID:18560524

Møller M, Alfheim I (1980). Mutagenicity and PAH-analysis of airborne particulate matter. *Atmos Environ*, 14(1):83–8. doi:10.1016/0004-6981(80)90111-0 PMID:6986880

Møller M, Alfheim I, Larssen S, Mikalsen A (1982). Mutagenicity of airborne particles in relation to traffic and air pollution parameters. *Environ Sci Technol*, 16(4):221–5. doi:10.1021/es00098a010

Møller P, Loft S (2010). Oxidative damage to DNA and lipids as biomarkers of exposure to air pollution. *Environ Health Perspect*, 118(8):1126–36. doi:10.1289/ehp.0901725 PMID:20423813

Monarca S, Crebelli R, Feretti D, Zanardini A, Fuselli S, Filini L et al. (1997). Mutagens and carcinogens in size-classified air particulates of a northern Italian

town. *Sci Total Environ*, 205(2–3):137–44. doi:10.1016/S0048-9697(97)00194-0 PMID:9372626

Moreno T, Merolla L, Gibbons W, Greenwell L, Jones T, Richards R (2004). Variations in the source, metal content and bioreactivity of technogenic aerosols: a case study from Port Talbot, Wales, UK. *Sci Total Environ*, 333(1–3):59–73. doi:10.1016/j.scitotenv.2004.04.019 PMID:15364519

Moriske HJ, Block I, Schleibinger H, Rüden H (1985). Polar neutral organic compounds in urban aerosols. 1. Chemical characterization and mutagenic effect in relation to various sources [in German]. *Zentralbl Bakteriol Mikrobiol Hyg B*, 181(3–5):240–71. PMID:4096145

Morozzi G, Mastrandrea V, Trotta F, Tonti A, Scardazza F, Cenci E (1992). Chemical characterization and biological properties of airborne particulate matter. *Aerobiologia*, 8(3):451–7. doi:10.1007/BF02272915

Morris WA, Versteeg JK, Bryant DW, Legzdins AE, McCarry BE, Marvin CH (1995). Preliminary comparisons between mutagenicity and magnetic susceptibility of respirable airborne particulate. *Atmos Environ*, 29(23):3441–50. doi:10.1016/1352-2310(95)00203-B

Motta S, Federico C, Saccone S, Librando V, Mosesso P (2004). Cytogenetic evaluation of extractable agents from airborne particulate matter generated in the city of Catania (Italy). *Mutat Res*, 561(1–2):45–52. doi:10.1016/j.mrgentox.2004.03.008 PMID:15238229

Motykiewicz G, Hadnagy W, Seemayer NH, Szeliga J, Tkocz A, Chorazy M (1991). Influence of airborne suspended matter on mitotic cell division. *Mutat Res*, 260(2):195–202. doi:10.1016/0165-1218(91)90008-A PMID:2046700

Motykiewicz G, Michalska J, Szeliga J, Cimander B (1988). Mutagenic and clastogenic activity of direct-acting components from air pollutants of the Silesian industrial region. *Mutat Res*, 204(2):289–96. doi:10.1016/0165-1218(88)90102-4 PMID:3278218

Motykiewicz G, Michalska J, Szeliga J, Konopacka M, Tkocz A, Hadnagy W et al. (1990). Genotoxicity of airborne suspended matter determined by in vitro and in vivo short-term assays. In: Seemayer NH, Hadnagy W, editors. Environmental hygiene II. Berlin: Springer-Verlag; pp. 17–21.

Motykiewicz G, Perera FP, Santella RM, Hemminki K, Seemayer NH, Chorazy M (1996). Assessment of cancer hazard from environmental pollution in Silesia. *Toxicol Lett*, 88(1–3):169–73. doi:10.1016/0378-4274(96)03734-4 PMID:8920733

Müller A, Alzuet P, Herbarth O, Ronco A (2001). Assessment of toxicity and mutagenicity in air particulate matter from an urban industrial area in the coast of the Rio de la Plata. *Environ Toxicol*, 16(2):151–7. doi:10.1002/tox.1019 PMID:11339715

Mustafic H, Jabre P, Caussin C, Murad MH, Escolano S, Tafflet M et al. (2012). Main air pollutants and myocardial infarction: a systematic review and meta-analysis. *JAMA*, 307(7):713–21. doi:10.1001/jama.2012.126 PMID:22337682

NCRP (1997). Deposition, retention, and dosimetry of inhaled radioactive substances (Report No. 125). Bethesda (MD): National Council on Radiation Protection and Measurements.

Nielsen PS, de Pater N, Okkels H, Autrup H (1996a). Environmental air pollution and DNA adducts in Copenhagen bus drivers – effect of *GSTM1* and *NAT2* genotypes on adduct levels. *Carcinogenesis*, 17(5):1021–7. doi:10.1093/carcin/17.5.1021 PMID:8640907

Nielsen PS, Okkels H, Sigsgaard T, Kyrtopoulos S, Autrup H (1996b). Exposure to urban and rural air pollution: DNA and protein adducts and effect of glutathione-*S*-transferase genotype on adduct levels. *Int Arch Occup Environ Health*, 68(3):170–6. doi:10.1007/BF00381627 PMID:8919845

Nodelman V, Ultman JS (1999). Longitudinal distribution of chlorine absorption in human airways: a comparison to ozone absorption. *J Appl Physiol (1985)*, 87(6):2073–80. PMID:10601152

Norppa H, Bonassi S, Hansteen IL, Hagmar L, Strömberg U, Rössner P et al. (2006). Chromosomal aberrations and SCEs as biomarkers of cancer risk. *Mutat Res*, 600(1–2):37–45. doi:10.1016/j.mrfmmm.2006.05.030 PMID:16814813

Novotna B, Topinka J, Solansky I, Chvatalova I, Lnenickova Z, Sram RJ (2007). Impact of air pollution and genotype variability on DNA damage in Prague policemen. *Toxicol Lett*, 172(1–2):37–47. doi:10.1016/j.toxlet.2007.05.013 PMID:17590289

O'Neill MS, Jerrett M, Kawachi I, Levy JI, Cohen AJ, Gouveia N et al.; Workshop on Air Pollution and Socioeconomic Conditions (2003). Health, wealth, and air pollution: advancing theory and methods. *Environ Health Perspect*, 111(16):1861–70. doi:10.1289/ehp.6334 PMID:14644658

Oberdörster G (1988). Lung clearance of inhaled insoluble and soluble particles. *J Aerosol Med*, 1(4):289–330. doi:10.1089/jam.1988.1.289

Obolenskaya MY, Teplyuk NM, Divi RL, Poirier MC, Filimonova NB, Zadrozna M et al. (2010). Human placental glutathione *S*-transferase activity and polycyclic aromatic hydrocarbon DNA adducts as biomarkers for environmental oxidative stress in placentas from pregnant women living in radioactivity- and chemically-polluted regions. *Toxicol Lett*, 196(2):80–6. doi:10.1016/j.toxlet.2010.03.1115 PMID:20380873

Oh SM, Kim HR, Park YJ, Lee SY, Chung KH (2011). Organic extracts of urban air pollution particulate matter ($PM_{2.5}$)-induced genotoxicity and oxidative stress in human lung bronchial epithelial cells (BEAS-2B cells). *Mutat Res*, 723(2):142–51. doi:10.1016/j.mrgentox.2011.04.003 PMID:21524716

Ohsawa M, Ochi T, Hayashi H (1983). Mutagenicity in *Salmonella typhimurium* mutants of serum extracts

from airborne particulates. *Mutat Res*, 116(2):83–90. doi:10.1016/0165-1218(83)90099-X PMID:6338361

Ohtani Y, Shimada Y, Ujiiye A (1985). Comparison between mutagenic activities of airborne particulates in Maebashi and in Minato-ku Tokyo. *J Jpn Soc Air Pollut*, 20:463–9.

Ohyama M, Otake T, Adachi S, Kobayashi T, Morinaga K (2007). A comparison of the production of reactive oxygen species by suspended particulate matter and diesel exhaust particles with macrophages. *Inhal Toxicol*, 19:Suppl 1: 157–60. doi:10.1080/08958370701496103 PMID:17886063

Pacini S, Giovannelli L, Gulisano M, Peruzzi B, Polli G, Boddi V et al. (2003). Association between atmospheric ozone levels and damage to human nasal mucosa in Florence, Italy. *Environ Mol Mutagen*, 42(3):127–35. doi:10.1002/em.10188 PMID:14556220

Pagano P, De Zaiacomo T, Scarcella E, Bruni S, Calamosca M (1996). Mutagenic activity of total and particle-sized fractions of urban particulate matter. *Environ Sci Technol*, 30(12):3512–6. doi:10.1021/es960182q

Palli D, Russo A, Masala G, Saieva C, Guarrera S, Carturan S et al. (2001). DNA adduct levels and DNA repair polymorphisms in traffic-exposed workers and a general population sample. *Int J Cancer*, 94(1):121–7. doi:10.1002/ijc.1433 PMID:11668486

Palli D, Saieva C, Grechi D, Masala G, Zanna I, Barbaro A et al. (2004). DNA bulky adducts in a Mediterranean population correlate with environmental ozone concentration, an indicator of photochemical smog. *Int J Cancer*, 109(1):17–23. doi:10.1002/ijc.11687 PMID:14735463

Palli D, Saieva C, Munnia A, Peluso M, Grechi D, Zanna I et al. (2008). DNA adducts and PM_{10} exposure in traffic-exposed workers and urban residents from the EPIC-Florence City study. *Sci Total Environ*, 403(1–3):105–12. doi:10.1016/j.scitotenv.2008.05.041 PMID:18603281

Palli D, Sera F, Giovannelli L, Masala G, Grechi D, Bendinelli B et al. (2009). Environmental ozone exposure and oxidative DNA damage in adult residents of Florence, Italy. *Environ Pollut*, 157(5):1521–5. doi:10.1016/j.envpol.2008.09.011 PMID:18954923

Pastorelli R, Restano J, Guanci M, Maramonte M, Magagnotti C, Allevi R et al. (1996). Hemoglobin adducts of benzo[*a*]pyrene diolepoxide in newspaper vendors: association with traffic exhaust. *Carcinogenesis*, 17(11):2389–94. doi:10.1093/carcin/17.11.2389 PMID:8968053

Pedersen DU, Durant JL, Penman BW, Crespi CL, Hemond HF, Lafleur AL et al. (1999). Seasonal and spatial variations in human cell mutagenicity of respirable airborne particles in the northeastern United States. *Environ Sci Technol*, 33(24):4407–15. doi:10.1021/es9905997

Pedersen DU, Durant JL, Penman BW, Crespi CL, Hemond HF, Lafleur AL et al. (2004). Human-cell mutagens in respirable airborne particles in the northeastern United States. 1. Mutagenicity of fractionated samples. *Environ Sci Technol*, 38(3):682–9. doi:10.1021/es0347282 PMID:14968851

Pedersen DU, Durant JL, Taghizadeh K, Hemond HF, Lafleur AL, Cass GR (2005). Human cell mutagens in respirable airborne particles from the northeastern United States. 2. Quantification of mutagens and other organic compounds. *Environ Sci Technol*, 39(24):9547–60. doi:10.1021/es050886c PMID:16475335

Pedersen M, Vinzents P, Petersen JH, Kleinjans JC, Plas G, Kirsch-Volders M et al. (2006). Cytogenetic effects in children and mothers exposed to air pollution assessed by the frequency of micronuclei and fluorescence in situ hybridization (FISH): a family pilot study in the Czech Republic. *Mutat Res*, 608(2):112–20. doi:10.1016/j.mrgentox.2006.02.013 PMID:16829164

Pedersen M, Wichmann J, Autrup H, Dang DA, Decordier I, Hvidberg M et al. (2009). Increased micronuclei and bulky DNA adducts in cord blood after maternal exposures to traffic-related air pollution. *Environ Res*, 109(8):1012–20. doi:10.1016/j.envres.2009.08.011 PMID:19783246

Peluso M, Bollati V, Munnia A, Srivatanakul P, Jedpiyawongse A, Sangrajrang S et al. (2012). DNA methylation differences in exposed workers and nearby residents of the Ma Ta Phut industrial estate, Rayong, Thailand. *Int J Epidemiol*, 41(6):1753–60, discussion 1761–3. doi:10.1093/ije/dys129 PMID:23064502

Peluso M, Merlo F, Munnia A, Valerio F, Perrotta A, Puntoni R et al. (1998). ^{32}P-postlabeling detection of aromatic adducts in the white blood cell DNA of nonsmoking police officers. *Cancer Epidemiol Biomarkers Prev*, 7(1):3–11. PMID:9456236

Peluso M, Srivatanakul P, Munnia A, Jedpiyawongse A, Ceppi M, Sangrajrang S et al. (2010). Malondialdehyde-deoxyguanosine adducts among workers of a Thai industrial estate and nearby residents. *Environ Health Perspect*, 118(1):55–9. PMID:20056580

Peluso M, Srivatanakul P, Munnia A, Jedpiyawongse A, Meunier A, Sangrajrang S et al. (2008). DNA adduct formation among workers in a Thai industrial estate and nearby residents. *Sci Total Environ*, 389(2–3):283–8. doi:10.1016/j.scitotenv.2007.09.012 PMID:17935758

Peng BC, Ye SH (1995). Investigation on the cytogenetic effects in tunnel bus drivers and conductors. *Chinese J Indust Med*, 6:334–6.

Pereira CE, Heck TG, Saldiva PH, Rhoden CR (2007). Ambient particulate air pollution from vehicles promotes lipid peroxidation and inflammatory responses in rat lung. *Braz J Med Biol Res*, 40(10):1353–9. doi:10.1590/S0100-879X2006005000164 PMID:17713644

Pereira TS, Beltrami LS, Rocha JAV, Broto FP, Comellas LR, Salvadori DM et al. (2013). Toxicogenetic monitoring in urban cities exposed to different airborne

contaminants. *Ecotoxicol Environ Saf*, 90:174–82. doi:10.1016/j.ecoenv.2012.12.029 PMID:23395453

Pereira TS, Gotor GN, Beltrami LS, Nolla CG, Rocha JA, Broto FP et al. (2010). *Salmonella* mutagenicity assessment of airborne particulate matter collected from urban areas of Rio Grande do Sul State, Brazil, differing in anthropogenic influences and polycyclic aromatic hydrocarbon levels. *Mutat Res*, 702(1):78–85. doi:10.1016/j.mrgentox.2010.07.003 PMID:20643224

Perera F, Hemminki K, Jedrychowski W, Whyatt R, Campbell U, Hsu Y et al. (2002). In utero DNA damage from environmental pollution is associated with somatic gene mutation in newborns. *Cancer Epidemiol Biomarkers Prev*, 11(10 Pt 1):1134–7. PMID:12376523

Perera F, Li TY, Zhou ZJ, Yuan T, Chen YH, Qu L et al. (2008). Benefits of reducing prenatal exposure to coal-burning pollutants to children's neurodevelopment in China. *Environ Health Perspect*, 116(10):1396–400. doi:10.1289/ehp.11480 PMID:18941584

Perera FP, Hemminki K, Gryzbowska E, Motykiewicz G, Michalska J, Santella RM et al. (1992). Molecular and genetic damage in humans from environmental pollution in Poland. *Nature*, 360(6401):256–8. doi:10.1038/360256a0 PMID:1436106

Perera FP, Jedrychowski W, Rauh V, Whyatt RM (1999). Molecular epidemiologic research on the effects of environmental pollutants on the fetus. *Environ Health Perspect*, 107:Suppl 3: 451–60. doi:10.1289/ehp.99107s3451 PMID:10346993

Perrone MG, Gualtieri M, Consonni V, Ferrero L, Sangiorgi G, Longhin E et al. (2013). Particle size, chemical composition, seasons of the year and urban, rural or remote site origins as determinants of biological effects of particulate matter on pulmonary cells. *Environ Pollut*, 176:215–27. doi:10.1016/j.envpol.2013.01.012 PMID:23434772

Petruzzelli S, Celi A, Pulerà N, Baliva F, Viegi G, Carrozzi L et al. (1998). Serum antibodies to benzo(a)pyrene diol epoxide-DNA adducts in the general population: effects of air pollution, tobacco smoking, and family history of lung diseases. *Cancer Res*, 58(18):4122–6. PMID:9751623

Piekarska K, Zaciera M, Czarny A, Zaczyńska E (2009). Mutagenic and cytotoxic properties of extracts of suspended particulate matter collected in Wrocław city area. *Environ Prot Eng*, 35:37–48.

Piekarska K, Zaciera M, Czarny A, Zaczyńska E (2011). Application of short-term tests in assessment of atmospheric air pollution. *Environ Prot Eng*, 37:85–98.

Pieters N, Plusquin M, Cox B, Kicinski M, Vangronsveld J, Nawrot TS (2012). An epidemiological appraisal of the association between heart rate variability and particulate air pollution: a meta-analysis. *Heart*, 98(15):1127–35. doi:10.1136/heartjnl-2011-301505 PMID:22628541

Pinkerton KE, Menache M, Plopper CG (1995). Changes in the tracheobronchial epithelium, pulmonary acinus, and lung antioxidant enzyme activity. In: Consequences of prolonged inhalation of ozone on F344 rats: collaborative studies. Research Report No. 65, Parts VIII and IX. Cambridge (MA): Health Effects Institute; pp. 41–98.

Pinkerton KE, Mercer RR, Plopper CG, Crapo JD (1992). Distribution of injury and microdosimetry of ozone in the ventilatory unit of the rat. *J Appl Physiol (1985)*, 73(3):817–24. PMID:1400043

Piperakis SM, Petrakou E, Tsilimigaki S (2000). Effects of air pollution and smoking on DNA damage of human lymphocytes. *Environ Mol Mutagen*, 36(3):243–9. doi:10.1002/1098-2280(2000)36:3<243::AID-EM8>3.0.CO;2-9 PMID:11044906

Pitts JN Jr, Sweetman JA Jr, Harger W, Fitz DR, Paur HR, Winer AM (1985). Diurnal mutagenicity of airborne particulate organic matter adjacent to a heavily traveled West Los Angeles freeway. *J Air Pollut Control Assoc*, 35(6):638–43. doi:10.1080/00022470.1985.10465939 PMID:2410469

Poli P, Buschini A, Restivo FM, Ficarelli A, Cassoni F, Ferrero I et al. (1999). Comet assay application in environmental monitoring: DNA damage in human leukocytes and plant cells in comparison with bacterial and yeast tests. *Mutagenesis*, 14(6):547–56. doi:10.1093/mutage/14.6.547 PMID:10567029

Poma A, Arrizza L, Picozzi P, Spanò L (2002). Monitoring urban air particulate matter (fractions $PM_{2.5}$ and PM_{10}) genotoxicity by plant systems and human cells in vitro: a comparative analysis. *Teratog Carcinog Mutagen*, 22(4):271–84. doi:10.1002/tcm.10020 PMID:12111711

Poma A, Limongi T, Pisani C, Granato V, Picozzi P (2006). Genotoxicity induced by fine urban air particulate matter in the macrophages cell line RAW 264.7. *Toxicol In Vitro*, 20(6):1023–9. doi:10.1016/j.tiv.2006.01.014 PMID:16504459

Postlethwait EM, Bidani A (1990). Reactive uptake governs the pulmonary air space removal of inhaled nitrogen dioxide. *J Appl Physiol (1985)*, 68(2):594–603. PMID:2318771

Postlethwait EM, Langford SD, Bidani A (1991). Transfer of NO_2 through pulmonary epithelial lining fluid. *Toxicol Appl Pharmacol*, 109(3):464–71. doi:10.1016/0041-008X(91)90009-4 PMID:1853345

Postlethwait EM, Langford SD, Jacobson LM, Bidani A (1995). NO_2 reactive absorption substrates in rat pulmonary surface lining fluids. *Free Radic Biol Med*, 19(5):553–63. doi:10.1016/0891-5849(95)00058-6 PMID:8529914

Pratt MM, King LC, Adams LD, John K, Sirajuddin P, Olivero OA et al. (2011). Assessment of multiple types of DNA damage in human placentas from smoking and nonsmoking women in the Czech Republic. *Environ Mol Mutagen*, 52(1):58–68. doi:10.1002/em.20581 PMID:20839217

Preidecker BLB (1980). I. Bacterial mutagenicity of particulates from Houston air. *Environ Mutagen*, 2(1):75–83. doi:10.1002/em.2860020111 PMID:7035159

Pyysalo H, Tuominen J, Wickstrom K, Skyttä E, Tikkanen L, Salomaa S et al. (1987). Polycyclic organic material (POM) in urban air. Fractionation, chemical analysis and genotoxicity of particulate and vapour phases in an industrial town in Finland. *Atmos Environ*, 21(5):1167–80. doi:10.1016/0004-6981(87)90244-7

Qian LM, Zhang BQ (1997). Breakpoint regression in research of airborne particulate genotoxicity. *J Biomath*, 12:93–6.

Qian LM, Zhang BQ, Wang WX (1997). Correlation between genotoxicity of airborne particulates and meteorological condition in Guangzhou city. *Chin J Publ Health*, 16:160–2.

Qin HY, Peng XW, Meng ZJ,, Huang JL, Li Q, Yang XB et al. (2012). $PM_{2.5}$ induced DNA damage in human bronchial epithelial cells [in Chinese]. *J Environ Health*, 29:391–3. Available from: http://www.cnki.net/.

Raaschou-Nielsen O, Andersen ZJ, Beelen R, Samoli E, Stafoggia M, Weinmayr G et al. (2013b). Air pollution and lung cancer incidence in 17 European cohorts: prospective analyses from the European Study of Cohorts for Air Pollution Effects (ESCAPE). *Lancet Oncol*, 14(9):813–22. doi:10.1016/S1470-2045(13)70279-1 PMID:23849838

Raaschou-Nielsen O, Sørensen M, Ketzel M, Hertel O, Loft S, Tjønneland A et al. (2013a). Long-term exposure to traffic-related air pollution and diabetes-associated mortality: a cohort study. *Diabetologia*, 56(1):36–46. PMID:22918192

Rahman MH, Arslan MI, Chen Y, Ali S, Parvin T, Wang LW et al. (2003). Polycyclic aromatic hydrocarbon-DNA adducts among rickshaw drivers in Dhaka City, Bangladesh. *Int Arch Occup Environ Health*, 76(7):533–8. doi:10.1007/s00420-003-0431-z PMID:12827370

Reche C, Moreno T, Amato F, Viana M, van Drooge BL, Chuang HC et al. (2012). A multidisciplinary approach to characterise exposure risk and toxicological effects of PM_{10} and $PM_{2.5}$ samples in urban environments. *Ecotoxicol Environ Saf*, 78:327–35. doi:10.1016/j.ecoenv.2011.11.043 PMID:22177483

Reliene R, Hlavacova A, Mahadevan B, Baird WM, Schiestl RH (2005). Diesel exhaust particles cause increased levels of DNA deletions after transplacental exposure in mice. *Mutat Res*, 570(2):245–52. doi:10.1016/j.mrfmmm.2004.11.010 PMID:15708583

Rhoden CR, Lawrence J, Godleski JJ, González-Flecha B (2004). *N*-acetylcysteine prevents lung inflammation after short-term inhalation exposure to concentrated ambient particles. *Toxicol Sci*, 79(2):296–303. doi:10.1093/toxsci/kfh122 PMID:15056806

Richter E, Rösler S, Scherer G, Gostomzyk JG, Grübl A, Krämer U et al. (2001). Haemoglobin adducts from aromatic amines in children in relation to area of residence and exposure to environmental tobacco smoke. *Int Arch Occup Environ Health*, 74(6):421–8. doi:10.1007/s004200100243 PMID:11563605

Riva DR, Magalhães CB, Lopes AA, Lanças T, Mauad T, Malm O et al. (2011). Low dose of fine particulate matter ($PM_{2.5}$) can induce acute oxidative stress, inflammation and pulmonary impairment in healthy mice. *Inhal Toxicol*, 23(5):257–67. doi:10.3109/08958378.2011.566290 PMID:21506876

Roberts ES, Richards JH, Jaskot R, Dreher KL (2003). Oxidative stress mediates air pollution particle-induced acute lung injury and molecular pathology. *Inhal Toxicol*, 15(13):1327–46. doi:10.1080/08958370390241795 PMID:14569496

Rojas E, Valverde M, Lopez MC, Naufal I, Sanchez I, Bizarro P et al. (2000). Evaluation of DNA damage in exfoliated tear duct epithelial cells from individuals exposed to air pollution assessed by single cell gel electrophoresis assay. *Mutat Res*, 468(1):11–7. doi:10.1016/S1383-5718(00)00035-8 PMID:10863153

Romieu I, Barraza-Villarreal A, Escamilla-Nuñez C, Almstrand AC, Diaz-Sanchez D, Sly PD et al. (2008). Exhaled breath malondialdehyde as a marker of effect of exposure to air pollution in children with asthma. *J Allergy Clin Immunol*, 121(4):903–9.e6. doi:10.1016/j.jaci.2007.12.004 PMID:18234317

Rooney C, Sethi T (2011). The epithelial cell and lung cancer: the link between chronic obstructive pulmonary disease and lung cancer. *Respiration*, 81(2):89–104. doi:10.1159/000323946 PMID:21311210

Rossi C, Poli P, Buschini A, Cassoni F, Cattani S, DeMunari E (1995). Comparative investigations among meteorological conditions, air chemical-physical pollutants and airborne particulate mutagenicity: a long-term study (1990–1994) from a northern Italian town. *Chemosphere*, 30(10):1829–45. doi:10.1016/0045-6535(95)00065-G PMID:7780721

Rossner P Jr, Rossnerova A, Spatova M, Beskid O, Uhlirova K, Libalova H et al. (2013b). Analysis of biomarkers in a Czech population exposed to heavy air pollution. Part II: chromosomal aberrations and oxidative stress. *Mutagenesis*, 28(1):97–106. doi:10.1093/mutage/ges058 PMID:23053823

Rossner P Jr, Rossnerova A, Sram RJ (2011c). Oxidative stress and chromosomal aberrations in an environmentally exposed population. *Mutat Res*, 707(1–2):34–41. doi:10.1016/j.mrfmmm.2010.12.005 PMID:21167186

Rossner P Jr, Svecova V, Milcova A, Lnenickova Z, Solansky I, Santella RM et al. (2007). Oxidative and nitrosative stress markers in bus drivers. *Mutat Res*, 617(1–2):23–32. doi:10.1016/j.mrfmmm.2006.11.033 PMID:17328930

Rossner P Jr, Svecova V, Schmuczerova J, Milcova A, Tabashidze N, Topinka J et al. (2013a). Analysis of biomarkers in a Czech population exposed to

heavy air pollution. Part I: bulky DNA adducts. *Mutagenesis*, 28(1):89–95. doi:10.1093/mutage/ges057 PMID:23047913

Rossner P Jr, Tabashidze N, Dostal M, Novakova Z, Chvatalova I, Spatova M et al. (2011a). Genetic, biochemical, and environmental factors associated with pregnancy outcomes in newborns from the Czech Republic. *Environ Health Perspect*, 119(2):265–71. doi:10.1289/ehp.1002470 PMID:20923744

Rossner P Jr, Uhlirova K, Beskid O, Rossnerova A, Svecova V, Sram RJ (2011b). Expression of *XRCC5* in peripheral blood lymphocytes is upregulated in subjects from a heavily polluted region in the Czech Republic. *Mutat Res*, 713(1–2):76–82. doi:10.1016/j.mrfmmm.2011.06.001 PMID:21684294

Rossnerova A, Spatova M, Rossner P, Solansky I, Sram RJ (2009). The impact of air pollution on the levels of micronuclei measured by automated image analysis. *Mutat Res*, 669(1–2):42–7. doi:10.1016/j.mrfmmm.2009.04.008 PMID:19409399

Rossnerova A, Tulupova E, Tabashidze N, Schmuczerova J, Dostal M, Rossner P Jr et al. (2013). Factors affecting the 27K DNA methylation pattern in asthmatic and healthy children from locations with various environments. *Mutat Res*, 741–742:18–26. doi:10.1016/j.mrfmmm.2013.02.003 PMID:23458556

Roubicek DA, Gutiérrez-Castillo ME, Sordo M, Cebrián-García ME, Ostrosky-Wegman P (2007). Micronuclei induced by airborne particulate matter from Mexico City. *Mutat Res*, 631(1):9–15. doi:10.1016/j.mrgentox.2007.04.001 PMID:17500027

Rowan-Carroll A, Halappanavar S, Williams A, Somers CM, Yauk CL (2013). Mice exposed in situ to urban air pollution exhibit pulmonary alterations in gene expression in the lipid droplet synthesis pathways. *Environ Mol Mutagen*, 54(4):240–9. doi:10.1002/em.21768 PMID:23536514

Rubes J, Lower X, Cassel M, Moore D, Perreault SD, Slott VL et al. (1996). Aneuploidy detection in sperm nuclei using fluorescence in situ hybridization. *Arch Zootec.*, 45(170–171):323–7.

Rubeš J, Pokorná Z, Borkovec L, Urbanová J, Strnadová V (1997). Dairy cattle as a bioindicator of exposure to genotoxic substances in a heavily polluted area in Northern Bohemia. *Mutat Res*, 391(1–2):57–70. doi:10.1016/S0165-1218(97)00032-3 PMID:9219549

Rubes J, Rybar R, Prinosilova P, Veznik Z, Chvatalova I, Solansky I et al. (2010). Genetic polymorphisms influence the susceptibility of men to sperm DNA damage associated with exposure to air pollution. *Mutat Res*, 683(1–2):9–15. doi:10.1016/j.mrfmmm.2009.09.010 PMID:19800896

Rubes J, Selevan SG, Evenson DP, Zudova D, Vozdova M, Zudova Z et al. (2005). Episodic air pollution is associated with increased DNA fragmentation in human sperm without other changes in semen quality. *Hum Reprod*, 20(10):2776–83. doi:10.1093/humrep/dei122 PMID:15980006

Rubes J, Selevan SG, Sram RJ, Evenson DP, Perreault SD (2007). *GSTM1* genotype influences the susceptibility of men to sperm DNA damage associated with exposure to air pollution. *Mutat Res*, 625(1–2):20–8. doi:10.1016/j.mrfmmm.2007.05.012 PMID:17714740

Ruchirawat M, Mahidol C, Tangjarukij C, Pui-ock S, Jensen O, Kampeerawipakorn O et al. (2002). Exposure to genotoxins present in ambient air in Bangkok, Thailand – particle associated polycyclic aromatic hydrocarbons and biomarkers. *Sci Total Environ*, 287(1–2):121–32. doi:10.1016/S0048-9697(01)01008-7 PMID:11883753

Ruchirawat M, Navasumrit P, Settachan D, Autrup H (2006). Environmental impacts on children's health in Southeast Asia: genotoxic compounds in urban air. *Ann N Y Acad Sci*, 1076(1):678–90. doi:10.1196/annals.1371.037 PMID:17119245

Ruchirawat M, Settachan D, Navasumrit P, Tuntawiroon J, Autrup H (2007). Assessment of potential cancer risk in children exposed to urban air pollution in Bangkok, Thailand. *Toxicol Lett*, 168(3):200–9. doi:10.1016/j.toxlet.2006.09.013 PMID:17157453

Rückerl R, Greven S, Ljungman P, Aalto P, Antoniades C, Bellander T et al.; AIRGENE Study Group (2007). Air pollution and inflammation (interleukin-6, C-reactive protein, fibrinogen) in myocardial infarction survivors. *Environ Health Perspect*, 115(7):1072–80. doi:10.1289/ehp.10021 PMID:17637925

Rückerl R, Ibald-Mulli A, Koenig W, Schneider A, Woelke G, Cyrys J et al. (2006). Air pollution and markers of inflammation and coagulation in patients with coronary heart disease. *Am J Respir Crit Care Med*, 173(4):432–41. doi:10.1164/rccm.200507-1123OC PMID:16293802

Rundell KW, Slee JB, Caviston R, Hollenbach AM (2008). Decreased lung function after inhalation of ultrafine and fine particulate matter during exercise is related to decreased total nitrate in exhaled breath condensate. *Inhal Toxicol*, 20(1):1–9. doi:10.1080/08958370701758593 PMID:18236215

Sakitani T, Hayashi K (1986). Mutagenic activities of air pollutants. Part 1. Diurnal and seasonal variations in the concentrations of six polycyclic aromatic hydrocarbons in the urban air and the relationship between their concentrations and mutagenic activities. *Tokyo Jikeikai Med J*, 101:247–57.

Sakitani T, Suzuki Y (1986). Mutagenic activities of air pollutants. Part 2. Mutagenic activities of air pollutants observed by micronucleus test. *Tokyo Jikeikai Med J*, 101:259–66.

Salam MT, Byun H-M, Lurmann F, Breton CV, Wang X, Eckel SP et al. (2012). Genetic and epigenetic variations in inducible nitric oxide synthase promoter, particulate pollution, and exhaled nitric oxide levels in children. *J*

Allergy Clin Immunol, 129(1):232–9.e1–7. doi:10.1016/j.jaci.2011.09.037 PMID:22055874

Saldiva PH, Clarke RW, Coull BA, Stearns RC, Lawrence J, Murthy GG et al. (2002). Lung inflammation induced by concentrated ambient air particles is related to particle composition. *Am J Respir Crit Care Med*, 165(12):1610–7. doi:10.1164/rccm.2106102 PMID:12070061

Samet J, Krewski D (2007). Health effects associated with exposure to ambient air pollution. *J Toxicol Environ Health A*, 70(3–4):227–42. doi:10.1080/15287390600884644 PMID:17365585

Samet JM, DeMarini DM, Malling HV (2004). Biomedicine. Do airborne particles induce heritable mutations? *Science*, 304(5673):971–2. doi:10.1126/science.1097441 PMID:15143266

Samet JM, Rappold A, Graff D, Cascio WE, Berntsen JH, Huang YC et al. (2009). Concentrated ambient ultrafine particle exposure induces cardiac changes in young healthy volunteers. *Am J Respir Crit Care Med*, 179(11):1034–42. doi:10.1164/rccm.200807-1043OC PMID:19234105

Sato MIZ, Valent GU, Coimbrão CA, Coelho MC, Sanchez Sanchez P, Alonso CD et al. (1995). Mutagenicity of airborne particulate organic material from urban and industrial areas of São Paulo, Brazil. *Mutat Res*, 335(3):317–30. doi:10.1016/0165-1161(95)00035-6 PMID:8524347

Schaumann F, Borm PJ, Herbrich A, Knoch J, Pitz M, Schins RP et al. (2004). Metal-rich ambient particles (particulate matter$_{2.5}$) cause airway inflammation in healthy subjects. *Am J Respir Crit Care Med*, 170(8):898–903. doi:10.1164/rccm.200403-423OC PMID:15229099

Scherer G, Frank S, Riedel K, Meger-Kossien I, Renner T (2000). Biomonitoring of exposure to polycyclic aromatic hydrocarbons of nonoccupationally exposed persons. *Cancer Epidemiol Biomarkers Prev*, 9(4):373–80. PMID:10794481

Schilderman PA, Hoogewerff JA, van Schooten FJ, Maas LM, Moonen EJ, van Os BJ et al. (1997). Possible relevance of pigeons as an indicator species for monitoring air pollution. *Environ Health Perspect*, 105(3):322–30. doi:10.1289/ehp.97105322 PMID:9171994

Schins RP, Lightbody JH, Borm PJ, Shi T, Donaldson K, Stone V (2004). Inflammatory effects of coarse and fine particulate matter in relation to chemical and biological constituents. *Toxicol Appl Pharmacol*, 195(1):1–11. doi:10.1016/j.taap.2003.10.002 PMID:14962500

Schneider JC, Card GL, Pfau JC, Holian A (2005). Air pollution particulate SRM 1648 causes oxidative stress in RAW 264.7 macrophages leading to production of prostaglandin E2, a potential Th2 mediator. *Inhal Toxicol*, 17(14):871–7. doi:10.1080/08958370500244498 PMID:16282164

Schwartz J (2004). Air pollution and children's health. *Pediatrics*, 113(4):Suppl): 1037–43. PMID:15060197

Seemayer NH, Hadnagy W, Behrendt H, Tomingas R (1989). Inhalation hazards from airborne particulates evaluated by in vitro cyto- and genotoxicity testing: a long-term study over a period of 14 years (1975–1988) from a highly industrialized area. *Exp Pathol*, 37(1–4):228–30. doi:10.1016/S0232-1513(89)80054-4 PMID:2637158

Seemayer NH, Hadnagy W, Tomingas R (1987a). Mutagenic and carcinogenic effects of airborne particulate matter from polluted areas on human and rodent tissue cultures. *Experientia Suppl*, 51:231–4. doi:10.1007/978-3-0348-7491-5_39 PMID:2958327

Seemayer NH, Hadnagy W, Tomingas R (1990a). Enhancement of cell transformation and induction of sister chromatid exchanges as test systems for detection of seasonal and local differences in genotoxicity of airborne particulates. In: Seemayer NH, Hadnagy W, editors. Environmental hygiene II. Berlin, Germany: Springer-Verlag; pp. 50–3. Available from: doi:10.1007/978-3-642-46712-7_12.

Seemayer NH, Hadnagy W, Tomingas R (1990b). Evaluation of health risks by airborne particulates from in vitro cyto- and genotoxicity testing on human and rodent tissue culture cells: a longitudinal study from 1975 until now. *J Aerosol Sci*, 21:S501–4. doi:10.1016/0021-8502(90)90290-E

Seemayer NH, Hadnagy W, Tomingas R, Manojlovic N (1987b). Cell- and genotoxic activities of airborne particulates from a highly industrialized region: a survey over a period of 11 years (1975–1986). *J Aerosp Sci*, 18(6):721–4. doi:10.1016/0021-8502(87)90106-6

Seemayer NH, Hornberg C, Hadnagy W (1994). Comparative genotoxicity testing of airborne particulates using rodent tracheal epithelial cells and human lymphocytes in vitro. *Toxicol Lett*, 72(1–3):95–103. doi:10.1016/0378-4274(94)90015-9 PMID:8202962

Seemayer NH, Manojlovic N, Konig H, Tomingas R (1988). Comparative investigation of carcinogenic and mutagenic activity of airborne particulate matter from polluted areas using human and rodent tissue culture cells. *Ann Occup Hyg*, 32:247–56. doi:10.1093/annhyg/32.inhaled_particles_VI.247

Seemayer NH, Manojlovic N, Schurer CC, Tomingas R (1984). Cell cultures as a tool for detection of cytotoxic, mutagenic and carcinogenic activity of airborne particulate matter. *J Aerosol Sci*, 15(3):426–30. doi:10.1016/0021-8502(84)90135-6

Selevan SG, Borkovec L, Slott VL, Zudová Z, Rubes J, Evenson DP et al. (2000). Semen quality and reproductive health of young Czech men exposed to seasonal air pollution. *Environ Health Perspect*, 108(9):887–94. doi:10.1289/ehp.00108887 PMID:11017895

Sellappa S, Sadhanandhan B, Francis A, Vasudevan SG (2010). Evaluation of genotoxicity in petrol station workers in South India using micronucleus assay. *Ind*

Health, 48(6):852–6. doi:10.2486/indhealth.MS1055 PMID:20616461

Senlin L, Zhenkun Y, Xiaohui C, Minghong W, Guoying S, Jiamo F et al. (2008). The relationship between physicochemical characterization and the potential toxicity of fine particulates ($PM_{2.5}$) in Shanghai atmosphere. *Atmos Environ*, 42(31):7205–14. doi:10.1016/j.atmosenv.2008.07.030

Sera N, Fukuhara K, Miyata N, Tokiwa H (1994). Detection of nitro-azabenzo[*a*]pyrene derivatives in the semivolatile phase originating from airborne particulate matter, diesel and gasoline vehicles. *Mutagenesis*, 9(1):47–52. doi:10.1093/mutage/9.1.47 PMID:8208130

Sevastyanova O, Binkova B, Topinka J, Sram RJ, Kalina I, Popov T et al. (2007). In vitro genotoxicity of PAH mixtures and organic extract from urban air particles: part II: human cell lines. *Mutat Res*, 620(1–2):123–34. doi:10.1016/j.mrfmmm.2007.03.002 PMID:17420030

Shah PS, Balkhair T; Knowledge Synthesis Group on Determinants of Preterm/LBW Births (2011). Air pollution and birth outcomes: a systematic review. *Environ Int*, 37(2):498–516. doi:10.1016/j.envint.2010.10.009 PMID:21112090

Shang Y, Fan L, Feng J, Lv S, Wu M, Li B et al. (2013). Genotoxic and inflammatory effects of organic extracts from traffic-related particulate matter in human lung epithelial A549 cells: the role of quinones. *Toxicol In Vitro*, 27(2):922–31. doi:10.1016/j.tiv.2013.01.008 PMID:23333790

Shao L, Shi Z, Jones TP, Li J, Whittaker AG, Bérubé KA (2006). Bioreactivity of particulate matter in Beijing air: results from plasmid DNA assay. *Sci Total Environ*, 367(1):261–72. doi:10.1016/j.scitotenv.2005.10.009 PMID:16313948

Sharma AK, Jensen KA, Rank J, White PA, Lundstedt S, Gagne R et al. (2007). Genotoxicity, inflammation and physico-chemical properties of fine particle samples from an incineration energy plant and urban air. *Mutat Res*, 633(2):95–111. doi:10.1016/j.mrgentox.2007.05.013 PMID:17683978

Shen H, Anastasio C (2011). Formation of hydroxyl radical from San Joaquin Valley particles extracted in a cell-free surrogate lung fluid. *Atmos Chem Phys*, 11(18):9671–82. doi:10.5194/acp-11-9671-2011 PMID:22121357

Shen H, Anastasio C (2012). A comparison of hydroxyl radical and hydrogen peroxide generation in ambient particle extracts and laboratory metal solutions. *Atmos Environ (1994)*, 46:665–8. doi:10.1016/j.atmosenv.2011.10.006 PMID:22267949

Shen H, Barakat AI, Anastasio C (2011). Generation of hydrogen peroxide from San Joaquin Valley particles in a cell-free solution. *Atmos Chem Phys*, 11(2):753–65. doi:10.5194/acp-11-753-2011

Shen RR, Sha LY, Wang ZS,, Deng YH, Yang SS (2009). A toxicological study based on DNA damage of inhalable particulates collected in Macao during winter. *China Environ Sci*, 29:991–6.

Shi T, Duffin R, Borm PJ, Li H, Weishaupt C, Schins RP (2006). Hydroxyl-radical-dependent DNA damage by ambient particulate matter from contrasting sampling locations. *Environ Res*, 101(1):18–24. doi:10.1016/j.envres.2005.09.005 PMID:16298360

Shi ZB, Shao LY, Jones TP (2004). Oxidative DNA damage of urban airborne particulates in plasmid DNA assay. *Chin Sci Bull*, 49:673–8.

Shimizu H, Suzuki Y, Hayashi K (1982). Mutagenic activities of urban air pollutants: (part 1) diurnal and seasonal variations of mutagenicity of nitromethane extracts. *Tokyo Jikeikai Med J*, 97:785–95.

Shukla A, Timblin C, BeruBe K, Gordon T, McKinney W, Driscoll K et al. (2000). Inhaled particulate matter causes expression of nuclear factor (NF)-kappaB-related genes and oxidant-dependent NF-kappaB activation in vitro. *Am J Respir Cell Mol Biol*, 23(2):182–7. doi:10.1165/ajrcmb.23.2.4035 PMID:10919984

Sigaud S, Goldsmith CA, Zhou H, Yang Z, Fedulov A, Imrich A et al. (2007). Air pollution particles diminish bacterial clearance in the primed lungs of mice. *Toxicol Appl Pharmacol*, 223(1):1–9. doi:10.1016/j.taap.2007.04.014 PMID:17561223

Singh R, Kaur B, Kalina I, Popov TA, Georgieva T, Garte S et al. (2007b). Effects of environmental air pollution on endogenous oxidative DNA damage in humans. *Mutat Res*, 620(1–2):71–82. doi:10.1016/j.mrfmmm.2007.02.024 PMID:17434188

Singh R, Sram RJ, Binkova B, Kalina I, Popov TA, Georgieva T et al. (2007a). The relationship between biomarkers of oxidative DNA damage, polycyclic aromatic hydrocarbon DNA adducts, antioxidant status and genetic susceptibility following exposure to environmental air pollution in humans. *Mutat Res*, 620(1–2):83–92. doi:10.1016/j.mrfmmm.2007.02.025 PMID:17445838

Škarek M, Cupr P, Bartos T, Kohoutek J, Klánová J, Holoubek I (2007). A combined approach to the evaluation of organic air pollution – a case study of urban air in Sarajevo and Tuzla (Bosnia and Herzegovina). *Sci Total Environ*, 384(1–3):182–93. doi:10.1016/j.scitotenv.2007.06.040 PMID:17675217

Skillrud DM, Offord KP, Miller RD (1986). Higher risk of lung cancer in chronic obstructive pulmonary disease. A prospective, matched, controlled study. *Ann Intern Med*, 105(4):503–7. doi:10.7326/0003-4819-105-4-503 PMID:3752756

Slama R, Darrow L, Parker J, Woodruff TJ, Strickland M, Nieuwenhuijsen M et al. (2008). Meeting report: atmospheric pollution and human reproduction. *Environ Health Perspect*, 116(6):791–8. doi:10.1289/ehp.11074 PMID:18560536

Smith KR, Aust AE (1997). Mobilization of iron from urban particulates leads to generation of reactive oxygen species in vitro and induction of ferritin synthesis

in human lung epithelial cells. *Chem Res Toxicol*, 10(7):828–34. doi:10.1021/tx960164m PMID:9250418

Soares SRC, Bueno-Guimarães HM, Ferreira CM, Rivero DH, De Castro I, Garcia ML et al. (2003). Urban air pollution induces micronuclei in peripheral erythrocytes of mice in vivo. *Environ Res*, 92(3):191–6. doi:10.1016/S0013-9351(02)00061-0 PMID:12804515

Soberanes S, Gonzalez A, Urich D, Chiarella SE, Radigan KA, Osornio-Vargas A et al. (2012). Particulate matter air pollution induces hypermethylation of the p16 promoter via a mitochondrial ROS-JNK-DNMT1 pathway. *Sci Rep*, 2:275 doi:10.1038/srep00275 PMID:22355787

Somers CM (2011). Ambient air pollution exposure and damage to male gametes: human studies and in situ 'sentinel' animal experiments. *Syst Biol Reprod Med*, 57(1–2):63–71. doi:10.3109/19396368.2010.500440 PMID:21208146

Somers CM, Cooper DN (2009). Air pollution and mutations in the germline: are humans at risk? *Hum Genet*, 125(2):119–30. doi:10.1007/s00439-008-0613-6 PMID:19112582

Somers CM, McCarry BE, Malek F, Quinn JS (2004). Reduction of particulate air pollution lowers the risk of heritable mutations in mice. *Science*, 304(5673):1008–10. doi:10.1126/science.1095815 PMID:15143280

Somers CM, Yauk CL, White PA, Parfett CL, Quinn JS (2002). Air pollution induces heritable DNA mutations. *Proc Natl Acad Sci U S A*, 99(25):15904–7. doi:10.1073/pnas.252499499 PMID:12473746

Soogarun S, Suwansaksri J, Wiwanitkit V (2006). High sister chromatid exchange among a sample of traffic policemen in Bangkok, Thailand. *Southeast Asian J Trop Med Public Health*, 37(3):578–80. PMID:17120983

Sørensen M, Autrup H, Hertel O, Wallin H, Knudsen LE, Loft S (2003a). Personal exposure to $PM_{2.5}$ and biomarkers of DNA damage. *Cancer Epidemiol Biomarkers Prev*, 12(3):191–6. PMID:12646506

Sørensen M, Daneshvar B, Hansen M, Dragsted LO, Hertel O, Knudsen L et al. (2003b). Personal $PM_{2.5}$ exposure and markers of oxidative stress in blood. *Environ Health Perspect*, 111(2):161–6. doi:10.1289/ehp.5646 PMID:12573899

Sørensen M, Schins RP, Hertel O, Loft S (2005). Transition metals in personal samples of $PM_{2.5}$ and oxidative stress in human volunteers. *Cancer Epidemiol Biomarkers Prev*, 14(5):1340–3. doi:10.1158/1055-9965.EPI-04-0899 PMID:15894700

Sørensen M, Skov H, Autrup H, Hertel O, Loft S (2003c). Urban benzene exposure and oxidative DNA damage: influence of genetic polymorphisms in metabolism genes. *Sci Total Environ*, 309(1–3):69–80. doi:10.1016/S0048-9697(03)00054-8 PMID:12798093

Srám RJ, Benes I, Binková B, Dejmek J, Horstman D, Kotěsovec F et al. (1996). Teplice program – the impact of air pollution on human health. *Environ Health Perspect*, 104:Suppl 4: 699–714. PMID:8879999

Srám RJ, Beskid O, Rössnerova A, Rössner P, Lnenickova Z, Milcova A et al. (2007). Environmental exposure to carcinogenic polycyclic aromatic hydrocarbons – the interpretation of cytogenetic analysis by FISH. *Toxicol Lett*, 172(1–2):12–20. doi:10.1016/j.toxlet.2007.05.019 PMID:17604575

Šrám RJ, Binkova B, Dostal M, Merkerova-Dostalova M, Libalova H, Milcova A et al. (2013). Health impact of air pollution to children. *Int J Hyg Environ Health*, 216(5):533–40. doi:10.1016/j.ijheh.2012.12.001 PMID:23312845

Srám RJ, Binková B, Rössner P, Rubes J, Topinka J, Dejmek J (1999). Adverse reproductive outcomes from exposure to environmental mutagens. *Mutat Res*, 428(1–2):203–15. doi:10.1016/S1383-5742(99)00048-4 PMID:10517994

Srám RJ, Podrazilová K, Dejmek J, Mracková G, Pilcík T (1998). Single cell gel electrophoresis assay: sensitivity of peripheral white blood cells in human population studies. *Mutagenesis*, 13(1):99–103. doi:10.1093/mutage/13.1.99 PMID:9491403

Sree Devi V, Durga Rao V, Hara Gopal VV, Siva Prasad B, Sandhya Devi G, Jyothy A et al. (2009). Cytogenetic evaluation of traffic policemen occupationally exposed to vehicular exhaust. *Indian J Med Res*, 130(5):520–5. PMID:20090099

Sreedevi V, Hemaprasad M, Sandhyadevi G, Reddy PP (2006). Induction of sister chromatid exchanges in traffic policemen exposed to vehicular exhaust. *Mutat Res*, 606(1–2):80–4. doi:10.1016/j.mrgentox.2006.03.004 PMID:16697248

Staessen JA, Nawrot T, Hond ED, Thijs L, Fagard R, Hoppenbrouwers K et al. (2001). Renal function, cytogenetic measurements, and sexual development in adolescents in relation to environmental pollutants: a feasibility study of biomarkers. *Lancet*, 357(9269):1660–9. doi:10.1016/S0140-6736(00)04822-4 PMID:11425371

Stahlhofen W, Scheuch G, Bailey MR (1995). Investigations of retention of inhaled particles in the human bronchial tree. *Radiat Prot Dosimetry*, 60:311–9.

Steerenberg PA, Nierkens S, Fischer PH, van Loveren H, Opperhuizen A, Vos JG et al. (2001). Traffic-related air pollution affects peak expiratory flow, exhaled nitric oxide, and inflammatory nasal markers. *Arch Environ Health*, 56(2):167–74. doi:10.1080/00039890109604069 PMID:11339681

Stieb DM, Chen L, Eshoul M, Judek S (2012). Ambient air pollution, birth weight and preterm birth: a systematic review and meta-analysis. *Environ Res*, 117:100–11. doi:10.1016/j.envres.2012.05.007 PMID:22726801

Stoeger T, Reinhard C, Takenaka S, Schroeppel A, Karg E, Ritter B et al. (2006). Instillation of six different ultrafine carbon particles indicates a surface area

threshold dose for acute lung inflammation in mice. *Environ Health Perspect*, 114(3):328–33. doi:10.1289/ehp.8266 PMID:16507453

Strak M, Hoek G, Godri KJ, Gosens I, Mudway IS, van Oerle R et al. (2013). Composition of PM affects acute vascular inflammatory and coagulative markers – the RAPTES project. *PLoS ONE*, 8(3):e58944 doi:10.1371/journal.pone.0058944 PMID:23516583

Strak M, Janssen NA, Godri KJ, Gosens I, Mudway IS, Cassee FR et al. (2012). Respiratory health effects of airborne particulate matter: the role of particle size, composition, and oxidative potential – the RAPTES project. *Environ Health Perspect*, 120(8):1183–9. doi:10.1289/ehp.1104389 PMID:22552951

Strandell M, Zakrisson S, Alsberg T, Westerholm R, Winquist L, Rannug U (1994). Chemical analysis and biological testing of a polar fraction of ambient air, diesel engine, and gasoline engine particulate extracts. *Environ Health Perspect*, 102:Suppl 4: 85–92. doi:10.1289/ehp.94102s485 PMID:7529708

Sun TY, Li YF, Gao HN, Yang SL (1995). Mutagenic effect of extracts of ambient air fallout particulates on germ cells of male mice. *Carcinog Teratog Mutagen*, 7:180–4.

Tablin F, den Hartigh LJ, Aung HH, Lame MW, Kleeman MJ, Ham W et al. (2012). Seasonal influences on CAPs exposures: differential responses in platelet activation, serum cytokines and xenobiotic gene expression. *Inhal Toxicol*, 24(8):506–17. doi:10.3109/08958378.2012.695815 PMID:22746400

Taioli E, Sram RJ, Garte S, Kalina I, Popov TA, Farmer PB (2007). Effects of polycyclic aromatic hydrocarbons (PAHs) in environmental pollution on exogenous and oxidative DNA damage (EXPAH project): description of the population under study. *Mutat Res*, 620(1–2):1–6. doi:10.1016/j.mrfmmm.2007.02.016 PMID:17420032

Takagi Y, Sakiya K, Endoh O, Goto S, Kohzaki K, Murata M et al. (1992). Mutagenicity of airborne particulates in Sagamihara City. *J Vet Med Sci*, 54(2):193–9. doi:10.1292/jvms.54.193 PMID:1606249

Takeda N, Teranishi K, Hamada K (1983). Mutagenicity in *Salmonella typhimurium* TA98 of the serum extract of the organic matter derived from airborne particulates. *Mutat Res*, 117(1–2):41–6. doi:10.1016/0165-1218(83)90151-9 PMID:6339909

Tan MG, Wu YF, Li HY, Li Y, Zhang G, Shao S et al. (2004). Cytotoxicities of PM_{10} airborne particulates on human blood lymphocytes [in Chinese]. *Stud Trace Elem Health*, 21:1–4.

Tan MG, Wu YF, Shao SS, Li HY, Zou MJ, Dong M et al. (2002). Determination of TSP in the Da Pu tunnel of Shanghai and study of its chromosome aberration effect for human peripheral blood lymphocytes [in Chinese]. *Stud Trace Elem Health*, 19:51–4.

Tang D, Li TY, Liu JJ, Chen YH, Qu L, Perera F (2006). PAH-DNA adducts in cord blood and fetal and child development in a Chinese cohort. *Environ Health Perspect*, 114(8):1297–300. doi:10.1289/ehp.8939 PMID:16882543

Tang D, Li TY, Liu JJ, Zhou ZJ, Yuan T, Chen YH et al. (2008). Effects of prenatal exposure to coal-burning pollutants on children's development in China. *Environ Health Perspect*, 116(5):674–9. doi:10.1289/ehp.10471 PMID:18470301

Tao F, Gonzalez-Flecha B, Kobzik L (2003). Reactive oxygen species in pulmonary inflammation by ambient particulates. *Free Radic Biol Med*, 35(4):327–40. doi:10.1016/S0891-5849(03)00280-6 PMID:12899936

Tao F, Kobzik L (2002). Lung macrophage-epithelial cell interactions amplify particle-mediated cytokine release. *Am J Respir Cell Mol Biol*, 26(4):499–505. doi:10.1165/ajrcmb.26.4.4749 PMID:11919087

Tarantini A, Maitre A, Lefebvre E, Marques M, Marie C, Ravanat JL et al. (2009). Relative contribution of DNA strand breaks and DNA adducts to the genotoxicity of benzo[*a*]pyrene as a pure compound and in complex mixtures. *Mutat Res*, 671(1–2):67–75. doi:10.1016/j.mrfmmm.2009.08.014 PMID:19733579

Tokiwa H, Kitamori S, Nakagawa R, Horikawa K, Matamala L (1983). Demonstration of a powerful mutagenic dinitropyrene in airborne particulate matter. *Mutat Res*, 121(2):107–16. doi:10.1016/0165-7992(83)90108-2 PMID:6348530

Topinka J, Schwarz LR, Wiebel FJ, Cerná M, Wolff T (2000). Genotoxicity of urban air pollutants in the Czech Republic. Part II. DNA adduct formation in mammalian cells by extractable organic matter. *Mutat Res*, 469(1):83–93. doi:10.1016/S1383-5718(00)00061-9 PMID:10946245

Topinka J, Sevastyanova O, Binkova B, Chvatalova I, Milcova A, Lnenickova Z et al. (2007). Biomarkers of air pollution exposure – a study of policemen in Prague. *Mutat Res*, 624(1–2):9–17. doi:10.1016/j.mrfmmm.2007.02.032 PMID:17493640

Török G, Csik M, Kertesz M, Fay E, Somorjay T, Börzsönyi M et al. (1989). Mutagenicity and PAH content of airborn particulates and of fallen dusts from two Hungarian towns and emission samples from aluminum reduction and power plants. *Ann Ist Super Sanita*, 25(4):595–9. PMID:2631626

Toyokuni S, Sagripanti JL (1992). Iron-mediated DNA damage: sensitive detection of DNA strand breakage catalyzed by iron. *J Inorg Biochem*, 47(3–4):241–8. doi:10.1016/0162-0134(92)84069-Y PMID:1431883

Toyokuni S, Sagripanti JL (1994). Increased 8-hydroxydeoxyguanosine in kidney and liver of rats continuously exposed to copper. *Toxicol Appl Pharmacol*, 126(1):91–7. doi:10.1006/taap.1994.1094 PMID:8184438

Traversi D, Schilirò T, Degan R, Pignata C, Alessandria L, Gilli G (2011). Involvement of nitro-compounds in the mutagenicity of urban $PM_{2.5}$ and PM_{10} in Turin. *Mutat Res*, 726(1):54–9. doi:10.1016/j.mrgentox.2011.09.002 PMID:21920459

Tsai DH, Amyai N, Marques-Vidal P, Wang JL, Riediker M, Mooser V et al. (2012). Effects of particulate matter on inflammatory markers in the general adult population. *Part Fibre Toxicol*, 9(1):24 doi:10.1186/1743-8977-9-24 PMID:22769230

Tsou TC, Chen CL, Liu TY, Yang JL (1996). Induction of 8-hydroxydeoxyguanosine in DNA by chromium(III) plus hydrogen peroxide and its prevention by scavengers. *Carcinogenesis*, 17(1):103–8. doi:10.1093/carcin/17.1.103 PMID:8565117

Tuntawiroon J, Mahidol C, Navasumrit P, Autrup H, Ruchirawat M (2007). Increased health risk in Bangkok children exposed to polycyclic aromatic hydrocarbons from traffic-related sources. *Carcinogenesis*, 28(4):816–22. doi:10.1093/carcin/bgl175 PMID:17071945

Tuominen J, Salomaa S, Pyysalo H, Skytta E, Tikkanen L, Nurmela T et al. (1988). Polynuclear aromatic compounds and genotoxicity in particulate and vapor phases of ambient air: effect of traffic, season, and meteorological conditions. *Environ Sci Technol*, 22(10):1228–34. doi:10.1021/es00175a017 PMID:22148620

Turner MC, Chen Y, Krewski D, Calle EE, Thun MJ (2007). Chronic obstructive pulmonary disease is associated with lung cancer mortality in a prospective study of never smokers. *Am J Respir Crit Care Med*, 176(3):285–90. doi:10.1164/rccm.200612-1792OC PMID:17478615

Ulrich MM, Alink GM, Kumarathasan P, Vincent R, Boere AJ, Cassee FR (2002). Health effects and time course of particulate matter on the cardiopulmonary system in rats with lung inflammation. *J Toxicol Environ Health A*, 65(20):1571–95. doi:10.1080/00984100290071676 PMID:12396869

Umbuzeiro GA, Franco A, Martins MH, Kummrow F, Carvalho L, Schmeiser HH et al. (2008). Mutagenicity and DNA adduct formation of PAH, nitro-PAH, and oxy-PAH fractions of atmospheric particulate matter from São Paulo, Brazil. *Mutat Res*, 652(1):72–80. doi:10.1016/j.mrgentox.2007.12.007 PMID:18294902

UNEP (1999). Phasing lead out of gasoline: an analysis of policy approaches in different countries. Paris, France: United Nations Environment Programme. Available from: www.un.org/esa/gite/iandm/unep-lead.pdf.

Upadhyay D, Panduri V, Ghio A, Kamp DW (2003). Particulate matter induces alveolar epithelial cell DNA damage and apoptosis: role of free radicals and the mitochondria. *Am J Respir Cell Mol Biol*, 29(2):180–7. doi:10.1165/rcmb.2002-0269OC PMID:12600817

Valavanidis A, Fiotakis K, Bakeas E, Vlahogianni T (2005). Electron paramagnetic resonance study of the generation of reactive oxygen species catalysed by transition metals and quinoid redox cycling by inhalable ambient particulate matter. *Redox Rep*, 10(1):37–51. doi:10.1179/135100005X21606 PMID:15829110

Valavanidis A, Salika A, Theodoropoulou A (2000). Generation of hydroxyl radicals by urban suspended particulate air matter. The role of iron ions. *Atmos Environ*, 34(15):2379–86. doi:10.1016/S1352-2310(99)00435-5

van Eeden SF, Tan WC, Suwa T, Mukae H, Terashima T, Fujii T et al. (2001). Cytokines involved in the systemic inflammatory response induced by exposure to particulate matter air pollutants (PM_{10}). *Am J Respir Crit Care Med*, 164(5):826–30. doi:10.1164/ajrccm.164.5.2010160 PMID:11549540

van Houdt JJ, Alink GM, Boleij JSM (1987). Mutagenicity of airborne particles related to meteorological and air pollution parameters. *Sci Total Environ*, 61:23–36. doi:10.1016/0048-9697(87)90353-6

van Leeuwen DM, Pedersen M, Hendriksen PJ, Boorsma A, van Herwijnen MH, Gottschalk RW et al. (2008). Genomic analysis suggests higher susceptibility of children to air pollution. *Carcinogenesis*, 29(5):977–83. doi:10.1093/carcin/bgn065 PMID:18332047

van Leeuwen DM, van Herwijnen MH, Pedersen M, Knudsen LE, Kirsch-Volders M, Sram RJ et al. (2006). Genome-wide differential gene expression in children exposed to air pollution in the Czech Republic. *Mutat Res*, 600(1-2):12–22. doi:10.1016/j.mrfmmm.2006.05.032 PMID:16814814

van Maanen JMS, Borm PJA, Knaapen A, van Herwijnen M, Schilderman PA, Smith KR et al. (1999). In vitro effects of coal fly ashes: hydroxyl radical generation, iron release, and DNA damage and toxicity in rat lung epithelial cells. *Inhal Toxicol*, 11(12):1123–41. doi:10.1080/089583799196628 PMID:10562700

Vattanasit U, Navasumrit P, Khadka MB, Kanitwithayanun J, Promvijit J, Autrup H et al. (2014). Oxidative DNA damage and inflammatory responses in cultured human cells and in humans exposed to traffic-related particles. *Int J Hyg Environ Health*, 217(1):23–33. doi:10.1016/j.ijheh.2013.03.002 PMID:23567252

Vellosi R, Vannucchi C, Bianchi F, Fiorio R, Rosellini D, Ciacchini G et al. (1994). Mutagenic activity and chemical analysis of airborne particulates collected in Pisa (Italy). *Bull Environ Contam Toxicol*, 52(3):465–73. doi:10.1007/BF00197838 PMID:7511445

Vesterdal LK, Jantzen K, Sheykhzade M, Roursgaard M, Folkmann JK, Loft S et al. (2014). Pulmonary exposure to particles from diesel exhaust, urban dust or single-walled carbon nanotubes and oxidatively damaged DNA and vascular function in *apoE*$^{-/-}$ mice. *Nanotoxicology*, 8(1):61–71. doi:10.3109/17435390.2012.750385 PMID:23148895

Viau CJ, Lockard JM, Enoch HG, Sabharwal PS (1982). Comparison of the genotoxic activities of extracts from ambient and forest fire polluted air. *Environ Mutagen*, 4(1):37–43. doi:10.1002/em.2860040106 PMID:7040069

Victorin K (1994). Review of the genotoxicity of nitrogen oxides. *Mutat Res*, 317(1):43–55. doi:10.1016/0165-1110(94)90011-6 PMID:7507572

Victorin K (1996). Genotoxicity and carcinogenicity of ozone. *Scand J Work Environ Health*, 22:Suppl 3: 42–51. PMID:9122655

Villalobos-Pietrini R, Amador-Muñoz O, Waliszewski S, Hernández-Mena L, Munive-Colín Z, Gómez-Arroyo S et al. (2006). Mutagenicity and polycyclic aromatic hydrocarbons associated with extractable organic matter from airborne particles ≤10 µm in south-west Mexico City. *Atmos Environ*, 40(30):5845–57. doi:10.1016/j.atmosenv.2006.05.009

Villarini M, Moretti M, Fatigoni C, Agea E, Dominici L, Mattioli A et al. (2008). Evaluation of primary DNA damage, cytogenetic biomarkers and genetic polymorphisms for *CYP1A1* and *GSTM1* in road tunnel construction workers. *J Toxicol Environ Health A*, 71(21):1430–9. doi:10.1080/15287390802328580 PMID:18800292

Vinitketkumnuen U, Kalayanamitra K, Chewonarin T, Kamens R (2002). Particulate matter, PM_{10} & $PM_{2.5}$ levels, and airborne mutagenicity in Chiang Mai, Thailand. *Mutat Res*, 519(1–2):121–31. doi:10.1016/S1383-5718(02)00130-4 PMID:12160897

Vinzents PS, Møller P, Sørensen M, Knudsen LE, Hertel O, Jensen FP et al. (2005). Personal exposure to ultrafine particles and oxidative DNA damage. *Environ Health Perspect*, 113(11):1485–90. doi:10.1289/ehp.7562 PMID:16263500

Viras LG, Athanasiou K, Siskos PA (1990). Determination of mutagenic activity of airborne particulates and of the benzo[α]pyrene concentrations in Athens atmosphere. *Atmos Environ Part B Urban Atmos*, 24(2):267–74. doi:10.1016/0957-1272(90)90032-P

Wang CY, Lee MS, King CM, Warner PO (1980). Evidence for nitroaromatics as direct-acting mutagens of airborne particulates. *Chemosphere*, 9(2):83–7. doi:10.1016/0045-6535(80)90093-4

Wang XY, Ding GW (1998). Induction of SCE in human peripheral blood lymphocytes by TSP in Lanzhou region. *China Pub Health*, 14:417–8.

Wang XZ, Zhang YE (1984). The mutation test research on the indoor air pollutants and the outdoor ones. *J Harbin Med Univ*, 1:56–61.

Wang Y, Long DH, Zhu HG, Zhou DH (1991). Mutagenicity of total suspended particulate in the air of Beijing detected by micronucleus test in mice. *China Environ Sci*, 11:401–4.

Watanabe N (2000). Urinary 17-KS and 17-OHCS as markers of endocrine disruption by air pollution in primary school children. *Environ Health Prev Med*, 4(4):221–3. doi:10.1007/BF02931262 PMID:21432489

Watterson TL, Sorensen J, Martin R, Coulombe RA Jr (2007). Effects of $PM_{2.5}$ collected from Cache Valley Utah on genes associated with the inflammatory response in human lung cells. *J Toxicol Environ Health A*, 70(20):1731–44. doi:10.1080/15287390701457746 PMID:17885930

Watts R, Fitzgerald B, Heil G, Garabedian H, Williams R, Warren S et al. (1989). Use of bioassay methods to evaluate mutagenicity of ambient air collected near a municipal waste combustor. *JAPCA*, 39(11):1436–9. doi:10.1080/08940630.1989.10466636 PMID:2607359

Wei A, Meng Z (2006a). Evaluation of micronucleus induction of sand dust storm fine particles ($PM_{2.5}$) in human blood lymphocytes. *Environ Toxicol Pharmacol*, 22(3):292–7. doi:10.1016/j.etap.2006.04.003 PMID:21783723

Wei A, Meng Z (2006b). Induction of chromosome aberrations in cultured human lymphocytes treated with sand dust storm fine particles ($PM_{2.5}$). *Toxicol Lett*, 166(1):37–43. doi:10.1016/j.toxlet.2006.05.010 PMID:16814966

Wei AL, Meng ZQ (2006c). Genetic damage of dust storm fine particles on human blood lymphocytes. *J Environ Health*, 23:291–3.

Wei AL, Meng ZQ, Niu RF (2006). Effects of dust storm fine particles ($PM_{2.5}$) on micronuclei formation in human blood lymphocytes. *Acta Scientiae Circumstaniae*, 26:509–14.

Wei YH, Chang KT, Chiang PC, Chang SC (1991). Analysis and tracing of polycyclic aromatic hydrocarbons and mutagenicity of airborne particulates from the Taipei area. *Proc Natl Sci Counc Repub China B*, 15(1):53–62. PMID:1946812

White PA, Claxton LD (2004). Mutagens in contaminated soil: a review. *Mutat Res*, 567(2–3):227–345. doi:10.1016/j.mrrev.2004.09.003 PMID:15572286

Whittaker A, BéruBé K, Jones T, Maynard R, Richards R (2004). Killer smog of London, 50 years on: particle properties and oxidative capacity. *Sci Total Environ*, 334–335:435–45. doi:10.1016/j.scitotenv.2004.04.047 PMID:15504529

WHO (2006). Air quality guidelines, global update 2005. Copenhagen: World Health Organization Regional Office for Europe. Available from: http://www.who.int/phe/health_topics/outdoorair/outdoorair_aqg/en/.

Whyatt RM, Santella RM, Jedrychowski W, Garte SJ, Bell DA, Ottman R et al. (1998). Relationship between ambient air pollution and DNA damage in Polish mothers and newborns. *Environ Health Perspect*, 106:Suppl 3: 821–6. doi:10.1289/ehp.98106821 PMID:9646044

Wigle DT, Arbuckle TE, Turner MC, Bérubé A, Yang Q, Liu S et al. (2008). Epidemiologic evidence of relationships between reproductive and child health outcomes and environmental chemical contaminants. *J Toxicol Environ Health B Crit Rev*, 11(5–6):373–517. doi:10.1080/10937400801921320 PMID:18470797

Wise H, Balharry D, Reynolds LJ, Sexton K, Richards RJ (2006). Conventional and toxicogenomic assessment of the acute pulmonary damage induced by the instillation of Cardiff PM_{10} into the rat lung. *Sci Total Environ*,

360(1–3):60–7. doi:10.1016/j.scitotenv.2005.08.056 PMID:16597461

Wolff GT, Siak JS, Chan TL, Korsog PE (1986). Multivariate statistical analyses of air quality data and bacterial mutagenicity data from ambient aerosols. *Atmos Environ Part A Gen Top*, 20(11):2231–41. doi:10.1016/0004-6981(86)90314-8

Wu S, Deng F, Wei H, Huang J, Wang H, Shima M et al. (2012). Chemical constituents of ambient particulate air pollution and biomarkers of inflammation, coagulation and homocysteine in healthy adults: a prospective panel study. *Part Fibre Toxicol*, 9(1):49 doi:10.1186/1743-8977-9-49 PMID:23231781

Wullenweber M, Ketseridis G, Xander L, Ruden H (1982). Seasonal variations in the mutagenicity of urban aerosols, sampled in Berlin (West), with *Salmonella typhimurium* TA 98 (Ames test). *Staub Reinhalt Luft*, 42:411–5.

Xiao ZH, Shao LY, Zhang N (2009). A toxicological assessment of PM_{10} in Lanzhou: results from plasmid DNA assay. *China Environ Sci*, 29:561–6.

Xu DQ, Niu JP, Wan XZ, Wang YX, Chen XY, Xu J (2008b). DNA damages of fine particles from sandstorm in rat alveolar macrophages. *J Environ Health*, 25:67–8.

Xu DQ, Zhang WL (2004). Monitoring of pollution of air fine particles ($PM_{2.5}$) and study on their genetic toxicity. *Biomed Environ Sci*, 17(4):452–8. PMID:15745250

Xu H, Wang X, Pöschl U, Feng S, Wu D, Yang L et al. (2008a). Genotoxicity of total and fractionated extractable organic matter in fine air particulate matter from urban Guangzhou: comparison between haze and nonhaze episodes. *Environ Toxicol Chem*, 27(1):206–12. doi:10.1897/07-095.1 PMID:18092867

Xu HJ, Wang XM (2008). A comparison of comet assay and cell-blocked micronucleus for studying the genotoxicity of organic components in air particulate matters from urban area of Guangzhou. *China Trop Med*, 8:1321–3.

Yamaguchi T, Yamazaki H, Yamauchi A, Kakiuchi Y (1994). Mutagenicity of airborne particulates in a roadside atmosphere. *Jpn J Toxicol Environ Health*, 40(6):542–9. doi:10.1248/jhs1956.40.542

Yang K, Airoldi L, Pastorelli R, Restano J, Guanci M, Hemminki K (1996). Aromatic DNA adducts in lymphocytes of humans working at high and low traffic density areas. *Chem Biol Interact*, 101(2):127–36. doi:10.1016/0009-2797(96)03720-9 PMID:8760394

Yang WM, Wu BY (1984). Mutagenic study of airborne particulates: chromosomal aberrations in bone marrow cells in mice. *J Environ Health*, 1:11–4.

Yang WM, Wu BY, Ma YP (1994). Content of PAHs in different diameter airborne particles and relationship with their mutagenicity. *J Environ Health*, 11:10–3.

Yao JQ, Zhao XS, Zhu HG (1993). Mutagenicity of total suspended particulate in air by micronucleus test in mice. *Shanghai Environ Sci*, 12:18–20.

Yauk C, Polyzos A, Rowan-Carroll A, Somers CM, Godschalk RW, Van Schooten FJ et al. (2008). Germline mutations, DNA damage, and global hypermethylation in mice exposed to particulate air pollution in an urban/industrial location. *Proc Natl Acad Sci U S A*, 105(2):605–10. doi:10.1073/pnas.0705896105 PMID:18195365

Yauk CL (2004). Advances in the application of germline tandem repeat instability for in situ monitoring. *Mutat Res*, 566(2):169–82. doi:10.1016/j.mrrev.2003.08.001 PMID:15164979

Yauk CL, Fox GA, McCarry BE, Quinn JS (2000). Induced minisatellite germline mutations in herring gulls (*Larus argentatus*) living near steel mills. *Mutat Res*, 452(2):211–8. doi:10.1016/S0027-5107(00)00093-2 PMID:11024480

Yauk CL, Quinn JS (1996). Multilocus DNA fingerprinting reveals high rate of heritable genetic mutation in herring gulls nesting in an industrialized urban site. *Proc Natl Acad Sci U S A*, 93(22):12137–41. doi:10.1073/pnas.93.22.12137 PMID:8901546

Yi S, Zhang F, Qu F, Ding W (2014). Water-insoluble fraction of airborne particulate matter (PM_{10}) induces oxidative stress in human lung epithelial A549 cells. *Environ Toxicol*, 29(2):226–33. doi:10.1002/tox.21750 PMID:22331617

Yu ZY, Zhu HG, Jiang SH (1989). An investigation of mutagenicity of airborne particles in a gaswork in Shanghai. *J Health Toxicol*, 3:239–41.

Yuan FS, Ma YP, Wu ZC (1999a). Content of metals in different diameter airborne particles and effect on micronuclei formation in human lymphocytes [in Chinese]. *Wei Sheng Yan Jiu*, 28:21–2.

Yuan FS, Ma YP, Zhai WH (1999b). Effects of airborne particle sizes on micronuclei frequency in human binucleate lymphocytes. *J Health Toxicol*, 13:132–3.

Yuan FS, Xun Q (1994). Study of unscheduled DNA synthesis in human amnion cell FL induced atmospheric particles. *J Prev Med Chin People's Liberation Army*, 12:274–6.

Yuan FS, Xun Q, Ma YP (1994). Unscheduled DNA synthesis induced in human amnion cell FL by various-size atmospheric particles. *J Environ Health*, 11:49–51.

Zhang JH, Yang WM, Xu YW (2004). DNA damages in peripheral blood lymphocytes induced by organic extracts from ambient air particulates with various diameters [in Chinese]. *J Environ Health*, 21(2):88–90. Available from: http://www.cnki.net/kcms/detail/detail.aspx?filename=HJYJ200402012&dbcode=CJFQ&dbname=CJFD2004

Zhang M, Fu JL, He LY Wang Y, Hu M, Zhu T et al. (2003). Studies on the genetic and epigenetic toxicity of airborne fine particulate matter in Beijing. *China Environ Sci*, 23:337–40. Available from: http://en.cnki.

com.cn/Article_en/CJFDTOTAL-ZGHJ200304000.htm.

Zhang W, Lei TA, Lin ZQ, Zhang H-S, Yang D-F, Xi Z-G et al. (2011). Pulmonary toxicity study in rats with PM_{10} and $PM_{2.5}$: differential responses related to scale and composition. *Atmos Environ*, 45(4):1034–41. doi:10.1016/j.atmosenv.2010.10.043

Zhang XW, Li JW (1994). Effect of airborne particulate on sister chromatid exchange of lymphocytes of human peripheral blood. *J Lanzhou Med Coll*, 22:218–9.

Zhang XW, Zhang Z, Zhang XY, Yang WM (2002). Study on chromosomal damage induced by organic extracts of air particulates. *J Environ Health*, 19:200–1.

Zhang Y, Schauer JJ, Shafer MM, Hannigan MP, Dutton SJ (2008). Source apportionment of in vitro reactive oxygen species bioassay activity from atmospheric particulate matter. *Environ Sci Technol*, 42(19):7502–9. doi:10.1021/es800126y PMID:18939593

Zhao X, Niu J, Wang Y, Yan C, Wang X, Wang J (1998). Genotoxicity and chronic health effects of automobile exhaust: a study on the traffic policemen in the city of Lanzhou. *Mutat Res*, 415(3):185–90. doi:10.1016/S1383-5718(98)00066-7 PMID:9714799

Zhao X, Wan Z, Chen G, Zhu H, Jiang S, Yao J (2002). Genotoxic activity of extractable organic matter from urban airborne particles in Shanghai, China. *Mutat Res*, 514(1–2):177–92. doi:10.1016/S1383-5718(01)00338-2 PMID:11815256

Zhao X, Wan Z, Zhu H, Chen R (2003). The carcinogenic potential of extractable organic matter from urban airborne particles in Shanghai, China. *Mutat Res*, 540(1):107–17. doi:10.1016/S1383-5718(03)00178-5 PMID:12972063

Zhao XH, Wang XL, Niu JP, Wang J (2001). Pollution status and mutagenicity of particles in heavy traffic road of Lanzhou. *Acta Scientiae Circumstantiae*, 21:444–7.

Zhao XS, Zhu HG, Jiang SH, Yao JQ (1996). The mutagenic study of organic extract of suspended particulates in Shanghai. *Carcinog Teratog Mutagen*, 8:78–84.

Zhu CQ, Lam T, Jiang CQ, Wei BX, Chen YH, Xu QR (2003). A study on lymphocyte DNA damage in traffic policemen in Guangzhou [in Chinese]. *Zhonghua Lao Dong Wei Sheng Zhi Ye Bing Za Zhi*, 21(1):41–4. PMID:14761571

Zidzik J, Kalina I, Salagovic J, Sram RJ, Rössner P, Popov T et al. (2007). Influence of PAHs in ambient air on chromosomal aberrations in exposed subjects: international study – EXPAH. *Mutat Res*, 620(1–2):41–8. doi:10.1016/j.mrfmmm.2007.02.021 PMID:17391715

Zuurbier M, Hoek G, Oldenwening M, Meliefste K, Krop E, van den Hazel P et al. (2011). In-traffic air pollution exposure and CC16, blood coagulation, and inflammation markers in healthy adults. *Environ Health Perspect*, 119(10):1384–9. doi:10.1289/ehp.1003151 PMID:21665568

Zwanenburg TS (1988). Comparative analysis of the clastogenicity and cytotoxicity of airborne particulate matter generated during the fire at Schweizerhalle on November 1, 1986. *Mutat Res*, 206(3):395–409. doi:10.1016/0165-1218(88)90126-7 PMID:3200259