


## REVIEW ESSAY

## Prospects &amp; Overviews

# Early life exposure to air pollution impacts neuronal and glial cell function leading to impaired neurodevelopment

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## Abstract

The World Health Organisation recently listed air pollution as the most significant threat to human health. Air pollution comprises particulate matter (PM), metals, black carbon and gases such as ozone (O<sub>3</sub>), nitrogen dioxide (NO<sub>2</sub>) and carbon monoxide (CO). In addition to respiratory and cardiovascular disease, PM exposure is linked with increased risk of neurodegeneration as well as neurodevelopmental impairments. Critically, studies suggest that PM crosses the placenta, making direct *in utero* exposure a reality. Rodent models reveal that neuroinflammation, neurotransmitter imbalance and oxidative stress are triggered following gestational/early life exposure to PM, and may be exacerbated by concomitant mitochondrial dysfunction. Gestational PM exposure (potentiated by mitochondrial impairment in the metabolically active neonatal brain) not only impacts neurodevelopment but may sensitise the brain to subsequent cognitive impairment. Having reviewed this field, we conclude that strategies are urgently required to reduce exposure to PM during this sensitive developmental period.

## KEYWORDS

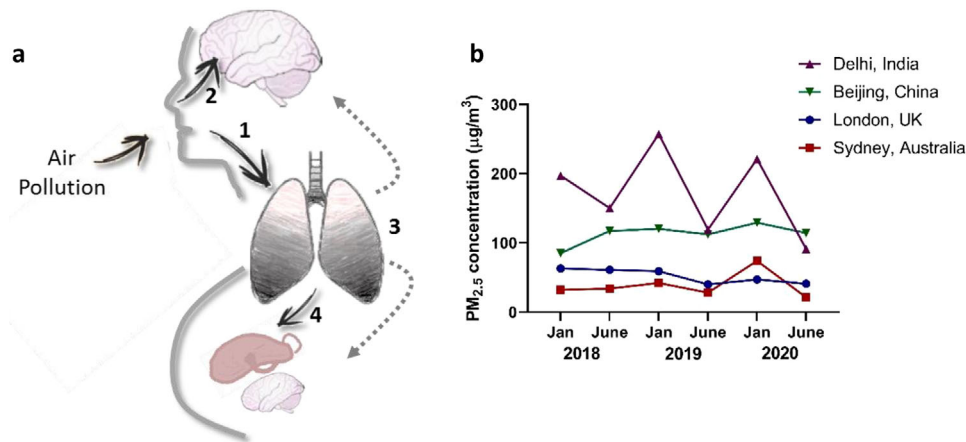
air pollution, gestational exposure, mitochondria, neurodevelopment, neuroinflammation, oxidative stress

## INTRODUCTION

Air pollution is one of the most pervasive and harmful environmental toxicants in the modern world. The World Health Organization estimate that 9 in 10 people are exposed to polluted air, contributing to an estimated 7 million deaths (World Health Organization, <https://www.who.int/news-room/air-pollution>. Press Release 02/05/18, accessed Oct 2020), ranging from respiratory complications such as pneumonia or chronic obstructive pulmonary disease, to stroke and cardiovascular conditions.<sup>[1,2]</sup>

**Abbreviations:** ASD, autism spectrum disorder; CO, carbon monoxide; CNS, central nervous system; DE, diesel exhaust; DEP, diesel exhaust particles; DA, dopamine; E, embryonic; MDPs, mitochondrial derived peptides; mtDNA, mitochondrial DNA; n, nano-particle sized; NDR, neurodevelopmental regression; NO<sub>2</sub>, Nitrogen dioxide; O<sub>3</sub>, ozone; PM, particulate matter; P, postnatal; RNS, reactive nitrogen species; ROS, reactive oxygen species; TLR4, Toll-like receptor 4; TRAP, traffic related air pollution; UFPM, ultrafine particulate matter

The main components of air pollution include gases such as nitrogen dioxide (NO<sub>2</sub>), ozone (O<sub>3</sub>), sulphur dioxide (SO<sub>2</sub>) and carbon monoxide (CO) as well as particulate matter (PM) composed of metals, soil/dust and organic chemicals.<sup>[3]</sup> PM is classified by its aerodynamic diameter, ranging from 10 μm particles (PM<sub>10</sub>), through to smaller PM<sub>2.5</sub> and ultrafine particles (UFPM), and chemical composition; these alter its biological and pathological effects. Approximately 25% of urban ambient particulate matter (PM<sub>2.5</sub>) is derived from Traffic Related Air Pollution (TRAP) with additional contributions from domestic fuel burning, industry, human and natural sources<sup>[4]</sup> The predominant route of exposure to TRAP is inhalation, and the smaller PM particles penetrate further into the lungs (Figure 1A). However, evidence suggests that PM<sub>2.5</sub> and UFPM can enter the brain through the olfactory system, eventually reaching the olfactory cortex and migrating to other brain regions. Furthermore, peripheral exposure



**FIGURE 1** Routes and levels of exposure to air pollution. (A) The predominant direct route of exposure to air pollution is inhalation (1). However, the brain can be exposed directly to smaller PM through the olfactory system (2), bypassing the blood brain barrier and migrating to other brain regions. Indirect exposure results from a systemic inflammatory response affecting maternal and developing brain (3). Critically recent research suggests that the foetal brain is also at risk from direct exposure from PM passing through the placenta (4). (B) Particulate matter exposure varies both seasonally and by location. Mean PM<sub>2.5</sub> concentrations ( $\mu\text{g m}^{-3}$ ) are shown in different cities from 2018–2020. Data were obtained and analyzed from the open data framework, World Air Quality Index Project, providing an average daily air quality index collected by relevant centers (UK-AIR, air quality information resource, Defra, UK, London Air Quality Network-Environmental Research Group, King's College London; Office of Environment and Heritage, New South Wales, Delhi Pollution Control Committee (Government of NCT of Delhi), India Central Pollution Control Board, Beijing Environmental Protection Monitoring center)

to PM triggers systemic inflammation that is known to contribute to brain pathology (Figure 1A). Therefore, although PM exposure is strongly associated with cardiovascular and respiratory disease,<sup>[5–7]</sup> increasing evidence suggests that air pollution not only contributes to, but drives long-term adverse effects in the central nervous system (CNS;<sup>[8]</sup>). Large scale studies carried out in diverse global populations have identified links between PM exposure and an increased risk of neurodegenerative diseases such as Alzheimer's disease and related dementias<sup>[9–12]</sup> as well as amyotrophic lateral sclerosis,<sup>[13]</sup> and Parkinson's disease.<sup>[12,14]</sup> As the majority of neurodegenerative conditions occur spontaneously, risk factors such as environmental toxicants may play a significant (yet preventable) role in triggering such pathologies. In the case of air pollution, risk due to exposure can vary widely depending on population density, industry and season; PM<sub>2.5</sub> concentrations can differ as much as ten-fold between major cities (Figure 1B).

Numerous epidemiological studies have provided well-documented links between early life PM exposures (during pregnancy and early childhood) and altered brain function, performance and behavior in both humans and in equivalent animal models. Data suggest that early-life exposure is associated with a plethora of altered behavior and performance measures such as reduced cognition, working memory, learning,<sup>[15–18]</sup> attentiveness, reaction times,<sup>[19]</sup> communication,<sup>[20]</sup> and behavioral regulation.<sup>[21]</sup> Mood disorders were also observed in rodent models where anxiety-like symptoms and increased depressive behaviors were displayed following PM exposure.<sup>[22,23]</sup> These behavioral data indicate the neurodevelopment trajectory is vulnerable to the harmful effects of exposure to air pollution and studies link early-

life PM exposure with an increased risk of autism spectrum disorder (ASD).<sup>[24–34]</sup>

Recently black carbon particles were identified on the foetal side of all human placentae examined in the study and significantly, particle load correlated with maternal exposure levels.<sup>[35]</sup> These data, combined with previous observations in rabbits where DE-derived nanoparticles were observed in placental trophoblasts,<sup>[36]</sup> provide disturbing evidence that direct exposure of the developing foetus to PM is likely (Figure 1A). Furthermore, evidence is accumulating that the third trimester represents the most susceptible window for PM exposure to impact pregnancy.<sup>[19,29]</sup> Exposure to PM therefore affects the CNS directly and indirectly with potentially life-long consequences. PM exposure can also result in long lasting epigenetic modifications with reduced histone methylation observed in both rodent and human brain.<sup>[37–40]</sup> Furthermore, and most relevantly, in utero PM<sub>2.5</sub> exposure has been associated with global hypomethylation in human placental tissue.<sup>[41]</sup> Chronic exposure of the developing brain to PM (postnatally and in utero) may therefore result in neurodevelopmental delay/disorders and sensitise infants to longer term neurodegenerative disease.

Here we review cellular mechanisms triggered in response to early life PM exposure and highlight available evidence from in vitro studies and in vivo rodent models exposed to PM during gestation (Table 1) and in early life. These reveal neuroinflammation, neurotransmitter imbalance and oxidative stress are crucial, interlinked players in the impairment of the developing brain after PM exposure (Figure 2) and that mitochondrial dysfunction may act to amplify these pathological mechanisms.

**TABLE 1** Rodent studies of gestational exposure to air pollution

Species	Type of air pollution exposure	Exposure concentration	Control exposure	Analyses	Key findings in offspring	Age/sex of offspring	Reference
Mice	DEP (oropharyngeal aspiration)	50–1000 $\mu\text{g mL}^{-1}$	Vehicle	Molecular pathway Immunohistochemistry/ Immunofluorescence	<ul style="list-style-type: none"> <li>Increased proinflammatory cytokines, increased TLR4 signaling/caspase-1 activation</li> <li>“Activated” microglial morphology, gross changes in cortical volume, increased microglial-neuronal interactions</li> </ul>	Adult, M	Bolton et al., 2013, 2017
Mice	Diluted Diesel exhaust (DE)	250–300 $\mu\text{g m}^{-3}$	Filtered air	Molecular pathway Immunohistochemistry/ Immunofluorescence	<ul style="list-style-type: none"> <li>Increased proinflammatory cytokines, increased DMNT1/decreased Reelin</li> <li>Disorganization of cortical lamina in the somatosensory cortex, neuroinflammation</li> </ul>	P3, P21 and P60, M&F	Chang et al., 2019
Mice	Concentrated ambient UFPM	92.69 $\mu\text{g m}^{-3}$ (average exposure value)	Filtered air	Immunohistochemistry/ Immunofluorescence	<ul style="list-style-type: none"> <li>Hypermyelination in corpus callosum, ventriculomegaly, reduced hippocampal area. Hypermyelination in cerebellum</li> </ul>	P11–15, M&F P11–15, M	Klocke et al., 2017, 2018
Mice	PM <sub>2.5</sub> (during gestation and early life)	600 $\mu\text{g m}^{-3}$	Filtered air	Behavior Immunohistochemistry/ Immunofluorescence Molecular pathway	<ul style="list-style-type: none"> <li>Increased anxiety-like behaviors (P90)</li> <li>Reduced corpus callosum volume (P90)</li> <li>Increased glial activation in numerous brain regions</li> <li>Decreased hippocampal BDNF expression</li> </ul>	P30 and P90, M	Di Domenico et al., 2020
Mice	PM <sub>2.5</sub> (oropharyngeal aspiration)	25–250 $\mu\text{g mL}^{-1}$ (max equiv to 1000 $\mu\text{g m}^{-3}$ )	Vehicle	Molecular Pathway Immunohistochemistry/ Immunofluorescence Behavior	<ul style="list-style-type: none"> <li>B-vitamin alleviated oxidative stress, neuroinflammation, mitochondrial damage, and synaptic dysfunction.</li> <li>B-vitamin restored hippocampal neurogenesis</li> <li>Gestational B-vitamin intervention corrected ASD-like behaviors (P40)</li> </ul>	P14 and P40, M&F	Wang et al., 2019
Mice	PM <sub>2.5</sub>	75–1000 $\mu\text{g m}^{-3}$	Filtered air	Behavior Electron microscopy Molecular pathway	<ul style="list-style-type: none"> <li>Impaired spatial memory (P30)</li> <li>Mitochondrial damage in hippocampal ultrastructure</li> <li>Increased levels of c-Fos, proapoptotic genes. Increased secretions of pro-inflammatory proteins, TNF-<math>\alpha</math> and IL-1<math>\beta</math></li> </ul>	P14 and P30, M&F	Zheng et al., 2018
Mice	Nano-sized and sub-fraction PM (n/sPM)	100–300 $\mu\text{g m}^{-3}$	Filtered air	Behavior Molecular pathway	<ul style="list-style-type: none"> <li>Increased depressive behaviors (M&gt;F)</li> <li>Body fat greater and glucose intolerance impaired (M). sPM altered inflammatory and glutamate associated genes in cerebral cortex (F)</li> </ul>	Adults, M&F	Haghani et al., 2020
Mice	Diluted Diesel exhaust (DE)	171 $\mu\text{g m}^{-3}$ (average exposure value)	Filtered air	Behavior Molecular pathway	<ul style="list-style-type: none"> <li>Reduced spontaneous locomotor activity</li> <li>Increased neurotransmitter levels (dopamine and noradrenaline) in the prefrontal cortex. Decreased neurotransmitter turnover in striatum, hippocampus, midbrain and cerebellum</li> </ul>	Adults, M	Suzuki et al., 2010

(Continues)

TABLE 1 (Continued)

Species	Type of air pollution exposure	Exposure concentration	Control exposure	Analyses	Key findings in offspring	Age/sex of offspring	Reference
Mice	Diluted Diesel exhaust (DE)	1000 $\mu\text{g m}^{-3}$	Filtered air	Behavior Molecular pathway	<ul style="list-style-type: none"> <li>Reduced spontaneous locomotor motor activity</li> <li>Decreased DA signaling</li> </ul>	Adults, M	Yokota et al., 2008
Rats	Ambient Nano-sized TRAP (gestation to adulthood)	340 $\mu\text{g m}^{-3}$	Filtered air	Immunohistochemistry/ Immunofluorescence Molecular pathway Behavior	<ul style="list-style-type: none"> <li>Impaired neurogenesis (M).</li> <li>'Activated' microglia in the dentate gyrus and CA1</li> <li>Altered BBB, with a 75% decrease of tight junction proteins</li> <li>Impaired memory and increased depressive behaviors</li> </ul>	Adult, M&F	Woodward et al., 2018
Rats	PM <sub>2.5</sub> (during gestation and early life)	43.8 $\mu\text{g m}^{-3}$ (average exposure value)	Filtered air	Molecular pathway Behavior	<ul style="list-style-type: none"> <li>Reduced OXTR protein, CAT and GSH concentrations (P22)</li> <li>Repetitive/restricted behavior (P29 and P43) and poor social interactions (P32)</li> </ul>	P22, P29, P32 and P43, M	Emam et al., 2020

## CHRONIC NEUROINFLAMMATION FOLLOWING EXPOSURE TO PM IS MEDIATED BY GLIAL CELL ACTIVATION

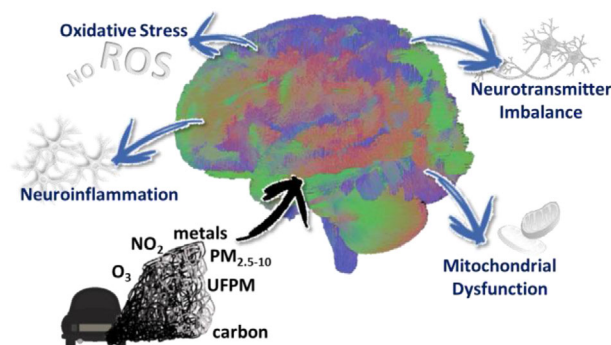
Glial cells (including astrocytes, microglia, and oligodendrocytes) comprise over 80% of the human brain.<sup>[42]</sup> It is therefore unsurprising that glial cell activation is a well-established outcome of PM exposure, and increased numbers of astrocytes and microglia are evident in the cortex, hippocampus, and dentate gyrus in rodents after exposure to PM.<sup>[43]</sup> Considering their cardinal role in brain health, chronic neuroinflammation may have significant adverse consequences in the immature brain during development.

### PM exposure results in microglial activation

Microglia are the resident innate immune cells of the brain; they are pivotal to normal brain development, contributing to neurogenesis, synaptic pruning, myelination and angiogenesis.<sup>[44]</sup>

Exposure of primary mouse microglia to diesel exhaust particles (DEP) or PM<sub>2.5</sub> for 4–24 h resulted in increased production of proinflammatory cytokines (IL1- $\beta$ , IL6) accompanied by a characteristic morphological change indicative of activation and increased reactive oxygen species (ROS) production.<sup>[45,46]</sup> Similar effects were observed in studies of primary rat microglia using a smaller particle size after both acute (<24 h) and chronic (<7 days) treatments<sup>[47,48]</sup> and in a microglial cell line with acute treatment using PM<sub>2.5</sub>.<sup>[49–51]</sup> Primary mixed glial cultures and microglial cell line experiments suggest that PM activate proinflammatory gene expression via toll-like receptor 4/NF- $\kappa$ B signaling.<sup>[52–55]</sup>

Co-cultures of primary microglia with rodent cerebellar granule<sup>[45]</sup> or cortical<sup>[46,56,57]</sup> neurons have established that exposure to DEP and/or PM<sub>2.5</sub> (and subsequent proinflammatory cytokine production) reduced neurite outgrowth and increased neurotoxicity by



**FIGURE 2** Overview of PM exposure in the immature brain. Direct or indirect exposure of the immature brain to air pollution (comprising gases e.g., nitrogen dioxide [NO<sub>2</sub>], ozone [O<sub>3</sub>], particulate matter [PM<sub>10</sub>, PM<sub>2.5</sub>, UFPM] and other compounds [e.g., black carbon, metals]) can result in perturbation of cellular mechanisms in neurons and glia, causing neurodevelopmental delay and long term impairment

2–3-fold. Co-treatment with drugs that blocked microglial activation and reduced the expression of proinflammatory cytokines (pioglitazone, minocycline) had neuroprotective roles.<sup>[45]</sup> A significant increase in extracellular glutamate levels was recorded following PM<sub>2.5</sub> exposure, which exacerbated neuronal cell death.<sup>[57]</sup> Interestingly, neurotoxicity was substantially increased in co-cultures pre-exposed to amyloid  $\beta$  oligomers.<sup>[46]</sup> These observations were reproduced in hippocampal organotypic culture generated from a triple transgenic model of Alzheimer's disease where microglial activation and amyloid  $\beta$  production were increased after exposure to UFPM (<4  $\mu\text{m}$ ).

In vivo, exposure to TRAP in rodent models of neurodevelopment and ageing have identified increased microglial activation across multiple brain regions. Offspring of rats were exposed to maternally-inhaled nanoparticle-sized PM throughout gestation and for 25 further weeks. Following this exposure, increased Iba1 staining (a marker for

activated microglia) was observed in specific hippocampal subregions, potentially representing the influence of neuroinflammation on neurogenesis in these regions.<sup>[58]</sup> Both chronic and intermittent gestational PM exposure (TRAP or DEP) in rodent models showed increased expression of TLR4, increased formation of the NLRP3 inflammasome and its downstream effector, caspase-1, in their offspring.<sup>[43,58-61]</sup> Interestingly, DEP-mediated neuroinflammation resulted in increased expression of DNA methyl transferase 1 and subsequent inhibition of Reelin expression; polymorphisms of Reelin have been identified in ASD.<sup>[60]</sup> Oropharyngeal exposure of pregnant mice to DEP from embryonic day (E)2, resulted in activated microglia and increased cortical volume in the foetal brain at E18; DEP-driven alteration in cortical volume was absent in TLR4<sup>-/-</sup> brains.<sup>[43]</sup> Significantly, the increase in volume was reversed by postnatal day (P)30 and in males, cortical volume was decreased, but microglia-neuron interactions increased suggesting that microglia were actively phagocytosing neurons.

### Astrocyte activation and oligodendrocyte dysfunction follows PM exposure

Although microglial activation is well documented following PM exposure, there is evidence implicating aberrant functioning of astrocytes in PM-induced neurotoxicity through mixed glial experiments.<sup>[52,53]</sup> However, in primary astrocytes exposed to PM<sub>2.5</sub>, there was a notable increase in GFAP expression, indicative of activation, which was accompanied by increased oxidative stress markers.<sup>[62]</sup> Examination of a rat astrocyte C6 astrogloma cell line exposed to < PM<sub>2.5</sub> for up to 24 h also found increased markers of activation including iNOS and IL-1 $\beta$ .<sup>[63]</sup> In parallel, activation of JAK2/STAT3 and p38/JNK/ERK MAPK-mediated signaling was increased, promoting proinflammatory gene expression. In utero and in the early postnatal period, exposure to PM<sub>2.5</sub> in mice resulted in dose-dependent increases in GFAP expression.<sup>[43,64-67]</sup>

Oligodendrocytes mediate the myelination of axons, a process that predominantly occurs in the third trimester of pregnancy and into the first years of life. Gestational exposure to ambient PM (UFPM to < PM<sub>2.5</sub>) resulted in increased ventricle size and increased myelin-positive staining of the corpus callosum in mice of both sexes at P11-15.<sup>[68,69]</sup> Hypermyelination was also observed in the cerebellum of male mice at the same age following gestational exposure.<sup>[69]</sup> Male mouse pups exposed to the same UFPM < PM<sub>2.5</sub> postnatally (at P4-7 and P10-13, equivalent to human third trimester) retained increased lateral ventricle size until adulthood.<sup>[70]</sup> This postnatal exposure to PM at a critical developmental period resulted in a reduction in the size of the corpus callosum at P14 in both sexes although hypomyelination was only observed in males. Astrocyte/microglial activation was evident throughout the time course,<sup>[68-70]</sup> suggesting impaired oligodendrocyte maturation leading to an altered myelination trajectory as observed in preterm brain,<sup>[71]</sup> although oxidative stress-mediated oligodendrocyte cell death has not been ruled out.

### PM EXPOSURE RESULTS IN NEUROTRANSMITTER IMBALANCE LINKED TO NEUROINFLAMMATION

Glutamate is the major excitatory neurotransmitter in the brain and impairments in its extracellular clearance by glial cells can result in oligodendrocyte damage, excitotoxic neuronal cell death and microglial activation. Exposure of mouse pups to ambient UFPM at P4-7 and P10-13 resulted in excitotoxicity that persisted until early adulthood, through a sustained increase in hippocampal glutamate.<sup>[72]</sup> Chronic ambient DEP exposure (4 weeks) increased glutamatergic NMDA receptor subunit expression especially in combination with systemic inflammation, which also led to impaired learning and memory.<sup>[73]</sup> Increased NMDA receptor subunit expression was also observed in mouse hippocampal slices following nano-particle sized (n)PM exposure, accompanied by increased glutamatergic AMPA receptor subunit expression.<sup>[74]</sup> However, electrophysiological (EPSC) measurement of synaptic function was decreased, suggesting impaired localization of AMPA and NMDA receptors, potentially due to increased S-nitrosylation and impaired insertion of AMPA receptors into the synaptic membrane.<sup>[57,74]</sup> In vitro, in primary rat hippocampal neurons exposed to nPM, co-treatment with AP5, a NMDA receptor antagonist blocked S-nitrosylation, neurotoxicity and attenuated the inhibition of neurite outgrowth.<sup>[74-76]</sup> An in vitro comparison of human primary neurons and rat cortical neurons exposed to PM<sub>2.5</sub> (24 h) found that human neuronal viability was significantly decreased compared with rat neurons.<sup>[57]</sup> Addition of conditioned medium from PM<sub>2.5</sub>-exposed microglia produced a further decrease in viability in rat primary neurons. This neurotoxicity was blocked by pretreatment with MK801, an NMDA receptor antagonist suggesting that glial cell production of glutamate contributes to the excitotoxic environment evoked by exposure to PM<sup>[57]</sup> and further cementing the link between neuroinflammation and excitotoxicity.<sup>[72]</sup>

Dopaminergic neurotransmitter signaling is also targeted by exposure to PM, significant in light of human population studies linking exposure to air pollution with the incidence of Parkinson's disease.<sup>[14]</sup> In co-cultures of primary rat mesencephalic neurons and microglia, exposure to DEP resulted in loss of neuronal viability and reduced dopamine (DA) uptake.<sup>[77]</sup> This functional impairment in DA neurons was prevented by CD11b/MAC1 inhibitor treatment again linking neuroinflammation with neurotransmitter dysfunction.<sup>[47]</sup> In vivo, gestational exposure to DEP led to decreased DA turnover in the striatum and nucleus accumbens, and a reduction of spontaneous locomotor activity in male offspring.<sup>[78,79]</sup>

### PM EXPOSURE INCREASES OXIDATIVE STRESS, OVERPOWERING ANTI-OXIDANT RESPONSES

Oxidative stress, an imbalance of antioxidants and free radicals leading to tissue damage, is a cardinal pathway associated with PM exposure and PM-induced neurotoxicity and is a pathological hallmark of both in vitro cellular and in vivo brain experiments. Moreover, pathway

enrichment analysis in pregnant women indicates that maternal oxidative stress-related pathways are altered, ultimately resulting in an increased risk of pregnancy complications.<sup>[80]</sup> PM exposure results in a dramatic increase in ROS and reactive nitrogen species (RNS) in primary rodent microglia<sup>[46,50,55,57]</sup> and neurons<sup>[51,74]</sup> resulting in significant damage to cellular components including proteins, lipids and DNA. Both acute and chronic exposure of adult rodents to inhaled PM<sub>2.5</sub> result in increased oxidative stress markers such as SOD1 and NRF2 in the brain accompanied by lipid peroxidation.<sup>[81-84]</sup> NRF2 is a transcription factor that can counteract ROS production and is vital for maintaining redox balances in the cell. Male rats chronically exposed to coarse, fine and UFPM increased antioxidant HO-1 and SOD-2 gene expression in the presence of NRF2 activation in the striatum.<sup>[81]</sup> However, Sotty and colleagues found that despite its activation, NRF2 failed to achieve sufficient protection against UFPM < PM<sub>2.5</sub>-mediated-oxidative burst in vitro in epithelial BEAS-2B cells.<sup>[85]</sup>

The immature CNS is particularly susceptible to oxidative stress injury due to its high metabolic activity, low level of antioxidants, and high cellular content of proteins and lipids.<sup>[86]</sup> Chronic exposure to stress-inducing toxins such as PM during early life, resulting in increased production of reactive oxygen or reactive nitrogen species (ROS/RNS), therefore have the potential of overwhelming the limited antioxidant system.<sup>[59]</sup> Gestational exposure of pregnant rodents to inhaled PM<sub>2.5</sub> increased the expression of markers of oxidative stress in their pups.<sup>[65,87,88]</sup> Such exposure led to increased intracellular ROS production accompanied by GSH depletion and the activation of NF- $\kappa$ B,<sup>[64]</sup> lipid peroxidation, signal changes in transport expression and function, a decrease of various tight junction proteins, blood brain barrier compromise and matrix metalloproteinase activation in their offspring.<sup>[65,87]</sup> Male mouse pups gestationally exposed to PM<sub>2.5</sub> had an aberrant expression of oxidative stress biomarkers CAT, OK-1, and SOD, which ultimately led to the downregulation of the oxytocin receptor, known to modulate key neurodevelopmental outcomes and associated with development of autism spectrum disorder.<sup>[87,89]</sup> It is noteworthy that many of these outcomes are due to air pollution inducing systemic oxidative stress and inflammation in non-neuronal tissues. Although systemic exposure to air pollution has shown to induce oxidative stress in mouse brain, the exacerbating and direct effect of DEP-induced systemic inflammation on neurotoxicity requires further elucidation.<sup>[82]</sup> However, antioxidant agents such as vitamin B6, B12, and folic acid blocked PM<sub>2.5</sub>-induced oxidative stress, abnormal DNA methylation and the aberrant expression of synapse-related genes in the hippocampus in offspring as well as normalizing the autism-like behavioral outcomes observed.<sup>[64]</sup>

## MITOCHONDRIAL DYSFUNCTION IS TRIGGERED BY MULTIPLE ROUTES FOLLOWING PM EXPOSURE AND MAY OFFER A NEW THERAPEUTIC TARGET

Mitochondria reside in all cells in the brain, a tissue known for its high metabolic demand, and brain mitochondria are some of the most long lived in the body.<sup>[90]</sup> Therefore, efficient mitochondrial function and

quality control is vital to preserve the resilience of brain to injury. Mitochondria are well known for their ability to generate ATP through oxidative phosphorylation, their calcium buffering and steroid hormone synthesis. In addition, they play key roles in cell death, regulating neurite outgrowth and synaptic function and mediating the inflammatory response.<sup>[91,92]</sup> Mitochondria therefore have the capacity to play a significant role in the brain's response to PM exposure.

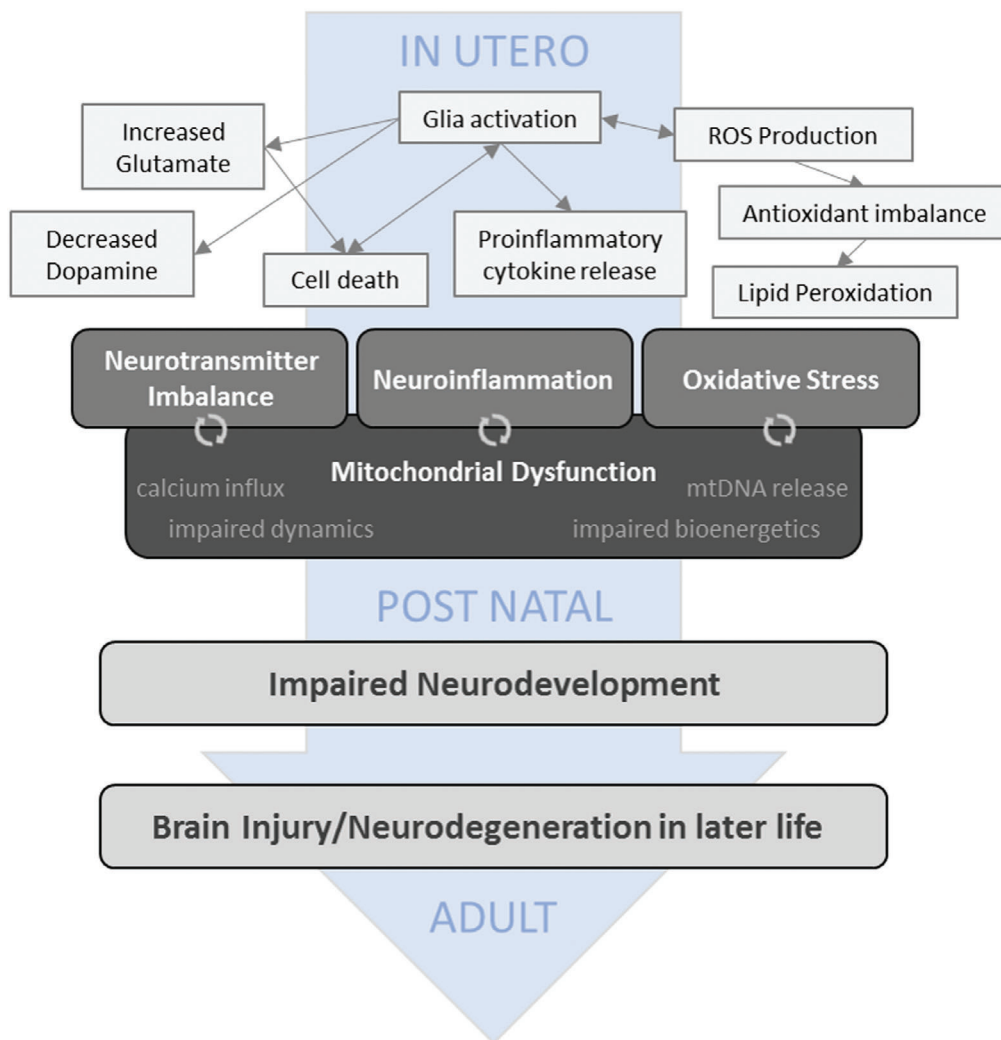
## Mitochondrial dysfunction following PM exposure

Mitochondrial dysfunction can be initiated via a variety of mechanisms including calcium influx, metabolic imbalance, impaired electron transport chain/ATP depletion, and mitochondrial permeabilization; the consequences are cellular energy crises, ROS production, and subsequent oxidative stress. In line with studies in respiratory and cardiovascular models of air pollution, mitochondria in the CNS cell population are increasingly being investigated as a target of air pollution.<sup>[93]</sup> In vitro, PM<sub>2.5</sub> exposure induced mitochondrial fragmentation in the neuron-like SH-SY5Y cell line, impaired ATP production, induced mitochondrial ROS production, and decreased mitochondrial membrane potential.<sup>[88]</sup>

Metabolic activity in the olfactory system, a primary route of entry for PM to the brain, may also be vulnerable to air pollution. Following exposure to PM<sub>1</sub>, PM<sub>2.5</sub> or PM<sub>10</sub> for up to 24h, primary human olfactory mucosal cells displayed increased oxidative stress (most significant for smaller PM) accompanied by decreased mitochondrial membrane potential and impaired basal and maximal respiration.<sup>[94]</sup> ATP production was also significantly impaired in cells treated with PM<sub>2.5</sub> or PM<sub>10</sub>. In addition to reduced antioxidant expression, alterations in mitochondrial fission and fusion genes were also observed in the nasal mucosa of rats exposed to PM<sub>2.5</sub>.<sup>[95]</sup> Mice exposed to UFPM via intranasal inhalation had decreased learning and memory, increased inflammation and ROS generation, and a diminished mitochondrial gene expression in the hippocampus.<sup>[59]</sup> This inhibition of mitochondrial genes was also separately observed in mouse lung tissue following intratracheal inhalation of DEPs.<sup>[96]</sup>

The energy demands of the brain are normally high, but during periods of neurodevelopment, sustained energy provision is critical. Few studies have examined the effect of gestational exposure to PM on mitochondria in the brain. Offspring of mice exposed to PM<sub>2.5</sub> throughout gestation showed evidence of increased neuronal apoptosis, mitochondrial matrix swelling, and structural damage in the hippocampus of mouse pups following gestational exposure to PM.<sup>[66,97]</sup> Similarly to rodents exposed to PM postnatally,<sup>[59,97]</sup> gestationally-exposed offspring, tested at P30, demonstrated impaired spatial learning.<sup>[66]</sup> Increased ROS and NO production and decreased neurite outgrowth were observed in wild-type mouse hippocampal cultures treated with UFPM, altering NMDA receptor function and decreasing evoked post-synaptic currents of CA1 neurons.<sup>[74]</sup>

Recently, a comprehensive analysis was conducted to determine changes in the rat brain proteome when exposed to coarse, PM<sub>2.5</sub> or UFPM particles for 1, 3, and 10 months.<sup>[98]</sup> At all time points



**FIGURE 3** Cellular mechanisms altered in the brain following PM exposure. Exposure in utero and in early life can trigger a wide variety of mechanisms broadly categorized into neurotransmitter imbalance, neuroinflammation and oxidative stress. Mitochondrial dysfunction, also triggered by exposure to PM, amplifies and potentiates these pathological effects. Unless this pathway integration is fully understood, therapeutic intervention will be only partially successful and the risks to the developing and ageing brain from exposure to air pollution are significant

and following exposure to both PM<sub>2.5</sub> and UFPM, mitochondrial activity was impaired through dysregulated expression of proteins involved in electron transport chain complexes. Additionally, down-regulation of NADPH was observed, implying a reduction in antioxidant capability.<sup>[98]</sup> These changes were accompanied by impairment of cellular processes required for development of dopaminergic neurons within the substantia nigra perhaps contributing to the potential vulnerability to Parkinson's disease after PM exposure.

### The potential of mitochondria-targeted treatments to mitigate exposure to PM

The multifaceted nature of their function may place mitochondria at the crossroads of the injury responses described earlier. In addition to being a major contributor of ROS and RNS, mitochondria can support a sustained inflammatory response, both through formation of the

inflammasome<sup>[99]</sup> as well as by release of pro-inflammatory mitochondrial DNA (mtDNA), which can act as a damage-associated molecular pattern.<sup>[100]</sup> Glutamate-mediated excitotoxicity results in calcium influx, mitochondrial swelling, loss of mitochondrial membrane potential and instigation of cell death mechanisms.<sup>[101]</sup> These functions may become more significant in a metabolically active environment such as the developing brain. Further support for this hypothesis comes from a very recent investigation of mitochondrial oxygen consumption rate of peripheral blood mononuclear cells of children with ASD (with and without neurodevelopmental regression (NDR);<sup>[102]</sup>). This pivotal study identified that prenatal exposure to air pollution resulted in prolonged alterations in mitochondrial respiration. In addition, these alterations impacted neurodevelopment and behavior with differences linked to direct (energy producing) and indirect (oxidative stress) effects of mitochondrial impairment. Finally, mitochondrial respiration was increased in children with ASD+NDR but decreased in children with ASD alone.<sup>[102]</sup> Therefore, therapies targeted at

maintaining mitochondrial homeostasis may provide a new avenue for pharmacological intervention with the potential for lifelong impact.

Pre-treatment of mice with Metformin (targeting complex I of the ETC) decreased mitochondrial ROS production, prevented release of proinflammatory IL6 and reduced vulnerability to arterial thrombosis (a model of stroke) in response to PM<sub>2.5</sub> exposure.<sup>[103]</sup> In vitro studies have also provided evidence that mitochondrial derived peptides (MDPs, e.g., humanin) may protect mitochondrial function following PM exposure. In vitro studies using SH-SY5Y cells following 24 h exposure to UFPM showed an increased oxidation of mtDNA and decreased mitochondrial oxygen consumption rate, which was rescued by pre-treatment with MDPs.<sup>[104]</sup> In vivo, the mitochondrially-targeted antioxidant MitoQ was administered to rats prior to chronic exposure to simulated vehicle exhaust (5 h/day, 2 weeks;<sup>[105]</sup>). MitoQ pre-treatment prevented mitochondrial ROS production, normalized measures of mitochondrial dynamics, and mitochondrial antioxidant defences, and reduced evidence of neuropathological behavior phenotypes (e.g., anxiety/depression;<sup>[105]</sup>). Mitochondrial-targeted therapies (e.g., SS peptides;<sup>[106]</sup>) may therefore warrant more extensive investigation in the context of neuroprotection in both the adult and developing brain following chronic exposure to air pollution.

## CONCLUSION AND OUTLOOK

This review highlights that early-life exposure to air pollution is associated with a wide range of interlinked direct and indirect cellular pathways leading to neurodevelopmental impairments with consequences for later life (Figure 3). Exposure to PM evokes a complex system of glial cell activation, neuroinflammation, neurotransmitter imbalance, and oxidative stress; mitochondrial dysfunction is emerging as a convergence point for the propagation of these pathological signals. Glia and neuronal cell culture models provide a simplified environment in which to analyse and manipulate cell-specific signaling pathways. Rodent models have proved to be a critical tool in studying the early life effects of PM exposure on brain function through pre- and post-natal exposure supporting the observations made in vitro (Table 1). Such studies in rodents have highlighted the consequences of glia activation and development of a proinflammatory environment for which the immature brain is ill-equipped to compensate. Subsequent damage to oligodendrocytes, white matter tracts and brain regions specializing in learning and memory act to impair neurodevelopment, leading to lifelong behavioral consequences. These rodent models are also critical if we are to understand the sex-specific differences in neurodevelopmental disorders such as ASD and the impact of differential gene expression regulating processes such as antioxidant defences. Indeed, many of the cellular pathways described here have been reported to be dysfunctional in people with ASD. Questions persist regarding effect size, critical susceptible windows, direct versus indirect exposure and the impact on the ageing brain. However, there remains an urgent and unmet need for strategies to decrease exposure to PM during pregnancy and early life, in order to reduce the risk of subsequent neurological impairment.

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## CONFLICT OF INTEREST

The authors declare no conflict of interest.

## DATA AVAILABILITY STATEMENT

Data sharing not applicable to this article as no datasets were generated or analysed during the current study.

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## REFERENCES

- Huangfu, P., & Atkinson, R. (2020). Long-term exposure to NO<sub>2</sub> and O<sub>3</sub> and all-cause and respiratory mortality: A systematic review and meta-analysis. *Environment International* 144, 105998.
- Cohen, A. J., Brauer, M., Burnett, R., Anderson, H. R., Frostad, J., Estep, K., Balakrishnan, K., Brunekreef, B., Dandona, L., Dandona, R., Feigin, V., Freedman, G., Hubbell, B., Jobling, A., Kan, H., Knibbs, L., Liu, Y., Martin, R., Morawska, L., & Forouzanfar, M. H. (2017). Estimates and 25-year trends of the global burden of disease attributable to ambient air pollution: An analysis of data from the Global Burden of Diseases Study 2015. *Lancet* 389, 1907–1918.
- Monks, P. S., Granier, C., Fuzzi, S., Stohl, A., Williams, M. L., Akimoto, H., Amann, M., Baklanov, A., Baltensperger, U., Bey, I., Blake, N., Blake, R. S., Carslaw, K., Cooper, O. R., Dentener, F., Fowler, D., Fragkou, E., Frost, G. J., Generoso, S., & Von Glasow, R. (2009). Atmospheric composition change – Global and regional air quality. *Atmospheric Environment* 43, 5268–5350.
- Karagulian, F., Belis, C. A., Dora, C. F. C., Prüss-Ustün, A. M., Bonjour, S., Adair-Rohani, H., & Amann, M. (2015). Contributions to cities' ambient particulate matter (PM): A systematic review of local source contributions at global level. *Atmospheric Environment* 120, 475–483.
- Kim, D., Chen, Z., Zhou, L. F., & Huang, S. X. (2018). Air pollutants and early origins of respiratory diseases. *Chronic Diseases and Translational Medicine*. 4, 75–94.
- Miller, M. R. (2020). Oxidative stress and the cardiovascular effects of air pollution. *Free Radical Biology and Medicine* 151, 69–87.
- Arias-Pérez, R. D., Tabora, N. A., Gómez, D. M., Narvaez, J. F., Porras, J., & Hernandez, J. C. (2020). Inflammatory effects of particulate matter air pollution. *Environmental Science and Pollution Research International*. 27(34), 42390–42404.
- Costa, L. G., Cole, T. B., Dao, K., Chang, Y.-C., Coburn, J., & Garrick, J. M. (2020). Effects of air pollution on the nervous system and its possible role in neurodevelopmental and neurodegenerative disorders. *Pharmacology & Therapeutics* 210, 107523.
- He, F., Tang, J. J., Zhang, T., Lin, J., Li, F., Gu, X., & Chen, R. (2020). Impact of air pollution on cognitive impairment in older people: A cohort study in rural and suburban China. *Journal of Alzheimers Disease*. 77(4), 1671–1679.
- Cacciottolo, M., Wang, X., Driscoll, I., Woodward, N., Saffari, A., Reyes, J., Serre, M. L., Vizuete, W., Sioutas, C., Morgan, T. E., Gatz, M., Chui, H. C., Shumaker, S. A., Resnick, S. M., Espeland, M. A., Finch, C. E., & Chen, J. C. (2017). Particulate air pollutants, APOE alleles and their contributions to cognitive impairment in older women and to amyloidogenesis in experimental models. *Translational Psychiatry* 7, e1022.
- Lee, J. H., Byun, M. S., Yi, D., Ko, K., Jeon, S. Y., Sohn, B. K., Lee, J.-Y., Lee, Y., Joung, H., & Lee, D. Y. (2020). Long-term exposure to PM10 and



- in vivo Alzheimer's disease pathologies. *Journal of Alzheimers Disease* 78(2), 745–756.
12. Fu, P., Guo, X., Cheung, F. M. H., & Yung, K. K. L. (2019). The association between PM2.5 exposure and neurological disorders: A systematic review and meta-analysis. *The Science of the Total Environment* 655, 1240–1248.
  13. Seelen, M., Toro Campos, R. A., Veldink, J. H., Visser, A. E., Hoek, G., Brunekreef, B., Van Der Kooij, A. J., De Visser, M., Raaphorst, J., Van Den Berg, L. H., & Vermeulen, R. C. H. (2017). Long-term air pollution exposure and amyotrophic lateral sclerosis in Netherlands: A population-based case-control study. *Environmental Health Perspectives* 125, 097023.
  14. Shi, L., Wu, X., Danesh Yazdi, M., Braun, D., Abu Awad, Y., Wei, Y., Liu, P., Di, Q., Wang, Y., Schwartz, J., Dominici, F., Kioumourtzoglou, M. A., & Zanobetti, A. (2020). Long-term effects of PM2.5 on neurological disorders in the American Medicare population: A longitudinal cohort study. *Lancet Planet Health*, 4(12), e557–e565.
  15. Basagaña, X., Esnaola, M., Rivas, I., Amato, F., Alvarez-Pedrerol, M., Forn, J., López-Vicente, M., Pujol, J., Nieuwenhuijsen, M., Querol, X., & Sunyer, J. (2016). Neurodevelopmental deceleration by urban fine particles from different emission sources: A longitudinal observational study. *Environmental Health Perspectives* 124, 1630–1636.
  16. Chiu, Y.-H. M., Hsu, H.-H. L., Coull, B. A., Bellinger, D. C., Kloog, I., Schwartz, J., Wright, R. O., & Wright, R. J. (2016). Prenatal particulate air pollution and neurodevelopment in urban children: Examining sensitive windows and sex-specific associations. *Environment International* 87, 56–65.
  17. Rivas, I., Querol, X., Wright, J., & Sunyer, J. (2018). How to protect school children from the neurodevelopmental harms of air pollution by interventions in the school environment in the urban context. *Environment International* 121, 199–206.
  18. Shehab, M. A., & Pope, F. D. (2019). Effects of short-term exposure to particulate matter air pollution on cognitive performance. *Scientific Reports* 9, 8237.
  19. Chiu, Y.-H. M., Bellinger, D. C., Coull, B. A., Anderson, S., Barber, R., Wright, R. O., & Wright, R. J. (2013). Associations between traffic-related black carbon exposure and attention in a prospective birth cohort of urban children. *Environmental Health Perspectives* 121, 859–864.
  20. Harris, M. H., Gold, D. R., Rifas-Shiman, S. L., Melly, S. J., Zanobetti, A., Coull, B. A., Schwartz, J. D., Gryparis, A., Kloog, I., Koutrakis, P., Bellinger, D. C., White, R. F., Sagiv, S. K., & Oken, E. (2015). Prenatal and childhood traffic-related pollution exposure and childhood cognition in the project viva cohort (Massachusetts, USA). *Environmental Health Perspectives* 123, 1072–1078.
  21. Harris, M. H., Gold, D. R., Rifas-Shiman, S. L., Melly, S. J., Zanobetti, A., Coull, B. A., Schwartz, J. D., Gryparis, A., Kloog, I., Koutrakis, P., Bellinger, D. C., Belfort, M. B., Webster, T. F., White, R. F., Sagiv, S. K., & Oken, E. (2016). Prenatal and childhood traffic-related air pollution exposure and childhood executive function and behavior. *Neurotoxicology and Teratology* 57, 60–70.
  22. Jia, Z., Wei, Y., Li, X., Yang, L., Liu, H., Guo, C., Zhang, L., Li, N., Guo, S., Qian, Y., & Li, Z. (2018). Exposure to ambient air particles increases the risk of mental disorder: findings from a natural experiment in Beijing. *International Journal of Environmental Research and Public Health* 15, 160.
  23. Khan, A., Plana-Ripoll, O., Antonsen, S., Brandt, J., Geels, C., Landecker, H., Sullivan, P. F., Pedersen, C. B., & Rzhetsky, A. (2019). Environmental pollution is associated with increased risk of psychiatric disorders in the US and Denmark. *PLoS Biology* 17, e3000353.
  24. Becerra, T. A., Wilhelm, M., Olsen, J., Cockburn, M., & Ritz, B. (2013). Ambient air pollution and autism in Los Angeles county, California. *Environmental Health Perspectives* 121, 380–386.
  25. Gong, T., Dalman, C., Wicks, S., Dal, H., Magnusson, C., Lundholm, C., Almqvist, C., & Pershagen, G. (2017). Perinatal exposure to traffic-related air pollution and autism spectrum disorders. *Environmental Health Perspectives* 125, 119–126.
  26. Chen, G., Jin, Z., Li, S., Jin, X., Tong, S., Liu, S., Yang, Y., Huang, H., & Guo, Y. (2018). Early life exposure to particulate matter air pollution (PM(1), PM(2.5) and PM(10)) and autism in Shanghai, China: A case-control study. *Environment International* 121, 1121–1127.
  27. Guxens, M., Ghassabian, A., Gong, T., Garcia-Esteban, R., Porta, D., Giorgis-Allemand, L., Almqvist, C., Aranbarri, A., Beelen, R., Badaloni, C., Cesaroni, G., De Nazelle, A., Estarlich, M., Forastiere, F., Forn, J., Gehring, U., Ibarluzea, J., Jaddoe, V. W. V., Korek, M., ..., Sunyer, J. (2016). Air pollution exposure during pregnancy and childhood autistic traits in four European population-based cohort studies: The ESCAPE project. *Environmental Health Perspectives* 124, 133–140.
  28. Jo, H., Eckel, S. P., Chen, J.-C., Cockburn, M., Martinez, M. P., Chow, T., Lurmann, F. W., Funk, W. E., Xiang, A. H., & McConnell, R. (2019). Gestational diabetes mellitus, prenatal air pollution exposure, and autism spectrum disorder. *Environment International* 133, 105110.
  29. Kaufman, J. A., Wright, J. M., Rice, G., Connolly, N., Bowers, K., & Anixt, J. (2019). Ambient ozone and fine particulate matter exposures and autism spectrum disorder in metropolitan Cincinnati, Ohio. *Environmental Research* 171, 218–227.
  30. Lyall, K., Schmidt, R. J., & Hertz-Picciotto, I. (2014). Maternal lifestyle and environmental risk factors for autism spectrum disorders. *International Journal of Epidemiology* 43, 443–464.
  31. Raz, R., Roberts, A. L., Lyall, K., Hart, J. E., Just, A. C., Laden, F., & Weiskopf, M. G. (2015). Autism spectrum disorder and particulate matter air pollution before, during, and after pregnancy: A nested case-control analysis within the Nurses' Health Study II Cohort. *Environmental Health Perspectives* 123, 264–270.
  32. Ritz, B., Liew, Z., Yan, Q., Cuia, X., Virk, J., Ketzler, M., & Raaschou-Nielsen, O. (2018). Air pollution and Autism in Denmark. *Environmental Epidemiology*, 2, e028.
  33. Volk, H. E., Lurmann, F., Penfold, B., Hertz-Picciotto, I., & McConnell, R. (2013). Traffic-related air pollution, particulate matter, and autism. *JAMA Psychiatry* 70, 71–7.
  34. Wang, P., Tuvblad, C., Younan, D., Franklin, M., Lurmann, F., Wu, J., Baker, L. A., & Chen, J.-C. (2017). Socioeconomic disparities and sexual dimorphism in neurotoxic effects of ambient fine particles on youth IQ: A longitudinal analysis. *PLoS One* 12, e0188731.
  35. Bové, H., Bongaerts, E., Slenders, E., Bijnens, E. M., Saenen, N. D., Gyselaers, W., Van Eyken, P., Plusquin, M., Roefsaers, M. B. J., Ameloot, M., & Nawrot, T. S. (2019). Ambient black carbon particles reach the fetal side of human placenta. *Nature Communications* 10, 3866.
  36. Valentino, S. A., Tarrade, A., Aioun, J., Mourier, E., Richard, C., Dahirel, M., Rousseau-Ralliard, D., Fournier, N., Aubrière, M.-C., Lallemand, M.-S., Camous, S., Guinot, M., Charlier, M., Aujean, E., Al Adhami, H., Fokkens, P. H., Agier, L., Boere, J. A., Cassee, F. R., ..., Chavatte-Palmer, P. (2016). Maternal exposure to diluted diesel engine exhaust alters placental function and induces intergenerational effects in rabbits. *Particle and Fibre Toxicology* 13, 39.
  37. Calderón-Garcidueñas, L., Herrera-Soto, A., Jury, N., Maher, B. A., González-Maciel, A., Reynoso-Robles, R., Ruiz-Rudolph, P., Van Zundert, B., & Varela-Nallar, L. (2020). Reduced repressive epigenetic marks, increased DNA damage and Alzheimer's disease hallmarks in the brain of humans and mice exposed to particulate urban air pollution. *Environmental Research* 183, 109226.
  38. Nwanaji-Enwerem, J. C., Colicino, E., Trevisi, L., Kloog, I., Just, A. C., Shen, J., Brennan, K., Dereix, A., Hou, L., Vokonas, P., Schwartz, J., & Baccarelli, A. A. (2016). Long-term ambient particle exposures and blood DNA methylation age: Findings from the VA normative aging study. *Environ Epigenet*, 2, dww006.
  39. Carmona, J. J., Sofer, T., Hutchinson, J., Cantone, L., Coull, B., Maity, A., Vokonas, P., Lin, X., Schwartz, J., & Baccarelli, A. A. (2014). Short-term airborne particulate matter exposure alters the epigenetic landscape of human genes associated with the mitogen-activated

- protein kinase network: A cross-sectional study. *Environmental Health* 13, 94.
40. White, A. J., Kresovich, J. K., Keller, J. P., Xu, Z., Kaufman, J. D., Weinberg, C. R., Taylor, J. A., & Sandler, D. P. (2019). Air pollution, particulate matter composition and methylation-based biologic age. *Environment International* 132, 105071.
  41. Janssen, B. G., Godderis, L., Pieters, N., Poels, K., Kiciński, M., Cuypers, A., Fierens, F., Penders, J., Plusquin, M., Gyselaers, W., & Nawrot, T. S. (2013). Placental DNA hypomethylation in association with particulate air pollution in early life. *Particle and Fibre Toxicology* 10, 22.
  42. Jäkel, S., & Dimou, L. (2017). Glial cells and their function in the adult brain: A journey through the history of their ablation. *Frontiers in Cellular Neuroscience* 11, 24.
  43. Bolton, J. L., Marinero, S., Hassanzadeh, T., Natesan, D., Le, D., Belliveau, C., Mason, S. N., Auten, R. L., & Bilbo, S. D. (2017). Gestational exposure to air pollution alters cortical volume, microglial morphology, and microglia-neuron interactions in a sex-specific manner. *Frontiers in Synaptic Neuroscience* 9, 10.
  44. Harry, G. J., & Kraft, A. D. (2012). Microglia in the developing brain: A potential target with lifetime effects. *Neurotoxicology* 33, 191–206.
  45. Roqué, P. J., Dao, K., & Costa, L. G. (2016). Microglia mediate diesel exhaust particle-induced cerebellar neuronal toxicity through neuroinflammatory mechanisms. *Neurotoxicology* 56, 204–214.
  46. Wang, B.-R., Shi, J.-Q., Ge, N.-N., Ou, Z., Tian, Y.-Y., Jiang, T., Zhou, J.-S., Xu, J., & Zhang, Y.-D. (2018). PM2.5 exposure aggravates oligomeric amyloid beta-induced neuronal injury and promotes NLRP3 inflammasome activation in an in vitro model of Alzheimer's disease. *Journal of Neuroinflammation* 15, 132.
  47. Levesque, S., Taetzsch, T., Lull, M. E., Kodavanti, U., Stadler, K., Wagner, A., Johnson, J. A., Duke, L., Kodavanti, P., Surace, M. J., & Block, M. L. (2011). Diesel exhaust activates and primes microglia: air pollution, neuroinflammation, and regulation of dopaminergic neurotoxicity. *Environmental Health Perspectives* 119, 1149–1155.
  48. Levesque, S., Taetzsch, T., Lull, M. E., Johnson, J. A., Mcgraw, C., & Block, M. L. (2013). The role of MAC1 in diesel exhaust particle-induced microglial activation and loss of dopaminergic neuron function. *Journal of Neurochemistry* 125, 756–765.
  49. Zhang, H., Haghani, A., Mousavi, A. H., Cacciottolo, M., D'agostino, C., Safi, N., Sowlat, M. H., Sioutas, C., Morgan, T. E., Finch, C. E., & Forman, H. J. (2019). Cell-based assays that predict in vivo neurotoxicity of urban ambient nano-sized particulate matter. *Free Radical Biology and Medicine* 145, 33–41.
  50. Lovett, C., Cacciottolo, M., Shirmohammadi, F., Haghani, A., Morgan, T. E., Sioutas, C., & Finch, C. E. (2018). Diurnal variation in the proinflammatory activity of urban fine particulate matter (PM 2.5) by in vitro assays. *F1000Res* 7, 596.
  51. Chen, X., Liu, S., Zhang, W., Wu, C., Liu, H., Zhang, F., Lu, Z., & Ding, W. (2018). Nrf2 deficiency exacerbates PM2.5-induced olfactory bulb injury. *Biochemical and Biophysical Research Communications* 505, 1154–1160.
  52. Woodward, N. C., Levine, M. C., Haghani, A., Shirmohammadi, F., Saffari, A., Sioutas, C., Morgan, T. E., & Finch, C. E. (2017). Toll-like receptor 4 in glial inflammatory responses to air pollution in vitro and in vivo. *Journal of Neuroinflammation* 14, 84.
  53. Haghani, A., Johnson, R., Safi, N., Zhang, H., Thorwald, M., Mousavi, A., Woodward, N. C., Shirmohammadi, F., Coussa, V., Wise, J. P., Forman, H. J., Sioutas, C., Allayee, H., Morgan, T. E., & Finch, C. E. (2020). Toxicity of urban air pollution particulate matter in developing and adult mouse brain: Comparison of total and filter-eluted nanoparticles. *Environment International* 136, 105510.
  54. Sama, P., Long, T. C., Hester, S., Tajuba, J., Parker, J., Chen, L.-C., & Veronesi, B. (2007). The cellular and genomic response of an immortalized microglia cell line (BV2) to concentrated ambient particulate matter. *Inhalation Toxicology* 19, 1079–1087.
  55. Bai, K.-J., Chuang, K.-J., Chen, C.-L., Jhan, M.-K., Hsiao, T.-C., Cheng, T.-J., Chang, L.-T., Chang, T.-Y., & Chuang, H.-C. (2019). Microglial activation and inflammation caused by traffic-related particulate matter. *Chemico-Biological Interactions* 311, 108762.
  56. Cheng, H., Davis, D. A., Hasheminassab, S., Sioutas, C., Morgan, T. E., & Finch, C. E. (2016). Urban traffic-derived nanoparticulate matter reduces neurite outgrowth via TNFalpha in vitro. *Journal of Neuroinflammation* 13, 19.
  57. Liu, F., Huang, Y., Zhang, F., Chen, Q., Wu, B., Rui, W., Zheng, J. C., & Ding, W. (2015). Macrophages treated with particulate matter PM2.5 induce selective neurotoxicity through glutaminase-mediated glutamate generation. *Journal of Neurochemistry* 134, 315–326.
  58. Woodward, N. C., Haghani, A., Johnson, R. G., Hsu, T. M., Saffari, A., Sioutas, C., Kanoski, S. E., Finch, C. E., & Morgan, T. E. (2018). Prenatal and early life exposure to air pollution induced hippocampal vascular leakage and impaired neurogenesis in association with behavioral deficits. *Translational Psychiatry* 8, 261.
  59. Li, X., Zhang, Y., Li, B., Yang, H., Cui, J., Li, X., Zhang, X., Sun, H., Meng, Q., Wu, S., Li, S., Wang, J., Aschner, M., & Chen, R. (2020). Activation of NLRP3 in microglia exacerbates diesel exhaust particles-induced impairment in learning and memory in mice. *Environment International* 136, 105487.
  60. Chang, Y.-C., Daza, R., Hevner, R., Costa, L. G., & Cole, T. B. (2019). Prenatal and early life diesel exhaust exposure disrupts cortical lamina organization: Evidence for a reelin-related pathogenic pathway induced by interleukin-6. *Brain, Behavior, and Immunity* 78, 105–115.
  61. Bolton, J. L., Huff, N. C., Smith, S. H., Mason, S. N., Foster, W. M., Auten, R. L., & Bilbo, S. D. (2013). Maternal stress and effects of prenatal air pollution on offspring mental health outcomes in mice. *Environmental Health Perspectives* 121, 1075–1082.
  62. Xu, M.-X., Zhu, Y.-F., Chang, H.-F., & Liang, Y. (2016). Nanoceeria restrains PM2.5-induced metabolic disorder and hypothalamus inflammation by inhibition of astrocytes activation related NF-kappaB pathway in Nrf2 deficient mice. *Free Radical Biology and Medicine* 99, 259–272.
  63. Li, T., Zhao, J., Ge, J., Yang, J., Song, X., Wang, C., Mao, J., Zhang, Y., Zou, Y., Liu, Y., & Chen, G. (2016). Particulate matter facilitates C6 glioma cells activation and the release of inflammatory factors through MAPK and JAK2/STAT3 pathways. *Neurochemical Research* 41, 1969–1981.
  64. Wang, T., Zhang, T., Sun, L., Li, W., Zhang, C., Yu, L., & Guan, Y. (2019). Gestational B-vitamin supplementation alleviates PM2.5-induced autism-like behavior and hippocampal neurodevelopmental impairment in mice offspring. *Ecotoxicology and Environmental Safety* 185, 109686.
  65. Di Domenico, M., Benevenuto, S. G. D. M., Tomasini, P. P., Yariwake, V. Y., De Oliveira Alves, N., Rahmeier, F. L., Da Cruz Fernandes, M., Moura, D. J., Nascimento Saldiva, P. H., & Veras, M. M. (2020). Concentrated ambient fine particulate matter (PM2.5) exposure induce brain damage in pre and postnatal exposed mice. *Neurotoxicology* 79, 127–141.
  66. Zheng, X., Wang, X., Wang, T., Zhang, H., Wu, H., Zhang, C., Yu, L., & Guan, Y. (2018). Gestational exposure to particulate matter 2.5 (PM2.5) leads to spatial memory dysfunction and neurodevelopmental impairment in hippocampus of mice offspring. *Frontiers in Neuroscience* 12, 1000.
  67. Block, M. L., & Calderón-Garcidueñas, L. (2009). Air pollution: mechanisms of neuroinflammation and CNS disease. *Trends in Neurosciences* 32, 506–516.
  68. Klocke, C., Allen, J. L., Sobolewski, M., Mayer-Proschel, M., Blum, J. L., Lauterstein, D., Zelikoff, J. T., & Cory-Slechta, D. A. (2017). Neuropathological consequences of gestational exposure to concentrated ambient fine and ultrafine particles in the mouse. *Toxicological Sciences: An Official Journal of the Society of Toxicology* 156, 492–508.

69. Klocke, C., Sherina, V., Graham, U. M., Gunderson, J., Allen, J. L., Sobolewski, M., Blum, J. L., Zelikoff, J. T., & Cory-Slechta, D. A. (2018). Enhanced cerebellar myelination with concomitant iron elevation and ultrastructural irregularities following prenatal exposure to ambient particulate matter in the mouse. *Inhalation Toxicology* 30, 381–396.
70. Allen, J. L., Oberdorster, G., Morris-Schaffer, K., Wong, C., Klocke, C., Sobolewski, M., Conrad, K., Mayer-Proschel, M., & Cory-Slechta, D. A. (2017). Developmental neurotoxicity of inhaled ambient ultrafine particle air pollution: Parallels with neuropathological and behavioral features of autism and other neurodevelopmental disorders. *Neurotoxicology* 59, 140–154.
71. Van Tilborg, E., Heijnen, C. J., Benders, M. J., Van Bel, F., Fleiss, B., Gressens, P., & Nijboer, C. H. (2016). Impaired oligodendrocyte maturation in preterm infants: Potential therapeutic targets. *Progress in Neurobiology* 136, 28–49.
72. Allen, J. L., Liu, X., Pelkowski, S., Palmer, B., Conrad, K., Oberdorster, G., Weston, D., Mayer-Pröschel, M., & Cory-Slechta, D. A. (2014). Early postnatal exposure to ultrafine particulate matter air pollution: persistent ventriculomegaly, neurochemical disruption, and glial activation preferentially in male mice. *Environmental Health Perspectives* 122, 939–945.
73. Win-Shwe, T.-T., Yamamoto, S., Fujitani, Y., Hirano, S., & Fujimaki, H. (2008). Spatial learning and memory function-related gene expression in the hippocampus of mouse exposed to nanoparticle-rich diesel exhaust. *Neurotoxicology* 29, 940–947.
74. Davis, D. A., Akopian, G., Walsh, J. P., Sioutas, C., Morgan, T. E., & Finch, C. E. (2013). Urban air pollutants reduce synaptic function of CA1 neurons via an NMDA/NO pathway in vitro. *Journal of Neurochemistry* 127, 509–519.
75. Morgan, T. E., Davis, D. A., Iwata, N., Tanner, J. A., Snyder, D., Ning, Z., Kam, W., Hsu, Y.-T., Winkler, J. W., Chen, J.-C., Petasis, N. A., Baudry, M., Sioutas, C., & Finch, C. E. (2011). Glutamatergic neurons in rodent models respond to nanoscale particulate urban air pollutants in vivo and in vitro. *Environmental Health Perspectives* 119, 1003–1009.
76. Woodward, N. C., Pakbin, P., Saffari, A., Shirmohammadi, F., Haghani, A., Sioutas, C., Cacciottolo, M., Morgan, T. E., & Finch, C. E. (2017). Traffic-related air pollution impact on mouse brain accelerates myelin and neuritic aging changes with specificity for CA1 neurons. *Neurobiology of Aging* 53, 48–58.
77. Block, M. L., Wu, X., Pei, Z., Li, G., Wang, T., Qin, L., Wilson, B., Yang, J., Hong, J. S., & Veronesi, B. (2004). Nanometer size diesel exhaust particles are selectively toxic to dopaminergic neurons: The role of microglia, phagocytosis, and NADPH oxidase. *FASEB Journal* 18, 1618–1620.
78. Yokota, S., Mizuo, K., Moriya, N., Oshio, S., Sugawara, I., & Takeda, K. (2009). Effect of prenatal exposure to diesel exhaust on dopaminergic system in mice. *Neuroscience Letters* 449, 38–41.
79. Suzuki, T., Oshio, S., Iwata, M., Saburi, H., Odagiri, T., Udagawa, T., Sugawara, I., Umezawa, M., & Takeda, K. (2010). In utero exposure to a low concentration of diesel exhaust affects spontaneous locomotor activity and monoaminergic system in male mice. *Particle and Fibre Toxicology* 7, 7.
80. Yan, Q., Liew, Z., Uppal, K., Cui, X., Ling, C., Heck, J. E., Von Ehrenstein, O. S., Wu, J., Walker, D. I., Jones, D. P., & Ritz, B. (2019). Maternal serum metabolome and traffic-related air pollution exposure in pregnancy. *Environment International* 130, 104872.
81. Guerra, R., Vera-Aguilar, E., Uribe-Ramirez, M., Gookin, G., Camacho, J., Osornio-Vargas, A. R., Mugica-Alvarez, V., Angulo-Olavis, R., Campbell, A., Froines, J., Kleinman, T. M., & De Vizcaya-Ruiz, A. (2013). Exposure to inhaled particulate matter activates early markers of oxidative stress, inflammation and unfolded protein response in rat striatum. *Toxicology Letters* 222, 146–154.
82. Milani, C., Farina, F., Botto, L., Massimino, L., Lonati, E., Donzelli, E., Ballarini, E., Crippa, L., Marmiroli, P., Bulbarelli, A., & Palestini, P. (2020). Systemic exposure to air pollution induces oxidative stress and inflammation in mouse brain, contributing to neurodegeneration onset. *International Journal of Molecular Sciences*, 21, 3699.
83. Cole, T. B., Coburn, J., Dao, K., Roqué, P., Chang, Y.-C., Kalia, V., Guilarte, T. R., Dziedzic, J., & Costa, L. G. (2016). Sex and genetic differences in the effects of acute diesel exhaust exposure on inflammation and oxidative stress in mouse brain. *Toxicology* 374, 1–9.
84. Chu, C., Zhang, H., Cui, S., Han, B., Zhou, L., Zhang, N., Su, X., Niu, Y., Chen, W., Chen, R., Zhang, R., & Zheng, Y. (2019). Ambient PM2.5 caused depressive-like responses through Nrf2/NLRP3 signaling pathway modulating inflammation. *Journal of Hazardous Materials* 369, 180–190.
85. Sotty, J., Kluz, J., De Sousa, C., Tardivel, M., Anthérieu, S., Alleman, L.-Y., Canivet, L., Perdrix, E., Loyens, A., Marchetti, P., Lo Guidice, J.-M., & Garçon, G. (2020). Mitochondrial alterations triggered by repeated exposure to fine (PM2.5-0.18) and quasi-ultrafine (PM0.18) fractions of ambient particulate matter. *Environment International* 142, 105830.
86. Blomgren, K., & Hagberg, H. (2006). Free radicals, mitochondria, and hypoxia-ischemia in the developing brain. *Free Radical Biology and Medicine* 40, 388–397.
87. Emam, B., Shahsavani, A., Khodaghali, F., Zarandi, S. M., Hopke, P. K., Hadei, M., Behbahani, H., & Yarahmadi, M. (2020). Effects of PM2.5 and gases exposure during prenatal and early-life on autism-like phenotypes in male rat offspring. *Particle and Fibre Toxicology* 17, 8.
88. Wang, Y., Zhang, M., Li, Z., Yue, J., Xu, M., Zhang, Y., Yung, K. K. L., & Li, R. (2019). Fine particulate matter induces mitochondrial dysfunction and oxidative stress in human SH-SY5Y cells. *Chemosphere* 218, 577–588.
89. Loparo, D., & Waldman, I. D. (2015). The oxytocin receptor gene (OXTR) is associated with autism spectrum disorder: a meta-analysis. *Molecular Psychiatry* 20, 640–646.
90. Navarro, A., & Boveris, A. (2004). Rat brain and liver mitochondria develop oxidative stress and lose enzymatic activities on aging. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* 287, R1244–R1249.
91. Misgeld, T., & Schwarz, T. L. (2017). Mitostasis in neurons: Maintaining mitochondria in an extended cellular architecture. *Neuron* 96, 651–666.
92. Culmsee, C., Michels, S., Scheu, S., Arolt, V., Dannlowski, U., & Alferink, J. (2018). Mitochondria, microglia, and the immune system—how are they linked in affective disorders? *Front Psychiatry* 9, 739.
93. Chew, S., Kolosowska, N., Saveleva, L., Malm, T., & Kanninen, K. M. (2020). Impairment of mitochondrial function by particulate matter: Implications for the brain. *Neurochemistry International* 135, 104694.
94. Chew, S., Lampinen, R., Saveleva, L., Korhonen, P., Mikhailov, N., Grubman, A., Polo, J. M., Wilson, T., Komppula, M., Rönkkö, T., Gu, C., Mackay-Sim, A., Malm, T., White, A. R., Jalava, P., & Kanninen, K. M. (2020). Urban air particulate matter induces mitochondrial dysfunction in human olfactory mucosal cells. *Particle and Fibre Toxicology* 17, 18.
95. Guo, Z., Hong, Z., Dong, W., Deng, C., Zhao, R., Xu, J., Zhuang, G., & Zhang, R. (2017). PM2.5-induced oxidative stress and mitochondrial damage in the nasal mucosa of rats. *International Journal of Environmental Research and Public Health*, 14, 134.
96. Li, X., Yang, H., Sun, H., Lu, R., Zhang, C., Gao, N., Meng, Q., Wu, S., Wang, S., Aschner, M., Wu, J., Tang, B., Gu, A., Kay, S. A., & Chen, R. (2017). Taurine ameliorates particulate matter-induced emphysema by switching on mitochondrial NADH dehydrogenase genes. *PNAS* 114, E9655–E9664.
97. Li, Q., Zheng, J., Xu, S., Zhang, J., Cao, Y., Qin, Z., Liu, X., & Jiang, C. (2018). The neurotoxicity induced by PM<sub>2.5</sub> might be strongly related to changes of the hippocampal tissue structure and neurotransmitter levels. *Toxicology Research* 7, 1144–1152.

98. Araújo, J. E., Jorge, S., Santos, H. M., Chiechi, A., Galstyan, A., Lodeiro, C., Diniz, M., Kleinman, M. T., Ljubimova, J. Y., & Capelo, J. L. (2019). Proteomic changes driven by urban pollution suggest particulate matter as a deregulator of energy metabolism, mitochondrial activity, and oxidative pathways in the rat brain. *The Science of the Total Environment* 687, 839–848.
99. Zhong, Z., Liang, S., Sanchez-Lopez, E., He, F., Shalpour, S., Lin, X.-J., Wong, J., Ding, S., Seki, E., Schnabl, B., Hevener, A. L., Greenberg, H. B., Kisseleva, T., & Karin, M. (2018). New mitochondrial DNA synthesis enables NLRP3 inflammasome activation. *Nature* 560, 198–203.
100. Grazioli, S., & Pugin, J. (2018). Mitochondrial damage-associated molecular patterns: From inflammatory signaling to human diseases. *Frontiers in Immunology* 9, 832.
101. Mira, R. G., & Cerpa, W. (2020). Building a bridge between NMDAR-mediated excitotoxicity and mitochondrial dysfunction in chronic and acute diseases. *Cellular and Molecular Neurobiology*.
102. Frye, R. E., Cakir, J., Rose, S., Delhey, L., Bennuri, S. C., Tippett, M., Melnyk, S., James, S. J., Palmer, R. F., Austin, C., Curtin, P., & Arora, M. (2020). Prenatal air pollution influences neurodevelopment and behavior in autism spectrum disorder by modulating mitochondrial physiology. *Molecular Psychiatry*.
103. Soberanes, S., Misharin, A. V., Jairaman, A., Morales-Nebreda, L., McQuattie-Pimentel, A. C., Cho, T., Hamanaka, R. B., Meliton, A. Y., Reyfman, P. A., Walter, J. M., Chen, C.-I., Chi, M., Chiu, S., Gonzalez-Gonzalez, F. J., Antalek, M., Abdala-Valencia, H., Chiarella, S. E., Sun, K. A., Woods, P. S., ..., Budinger, G. R. S. (2019). Metformin targets mitochondrial electron transport to reduce air-pollution-induced thrombosis. *Cell Metabolism* 29, 335–347.e5.
104. Breton, C. V., Song, A. Y., Xiao, J., Kim, S.-J., Mehta, H. H., Wan, J., Yen, K., Sioutas, C., Lurmann, F., Xue, S., Morgan, T. E., Zhang, J., & Cohen, P. (2019). Effects of air pollution on mitochondrial function, mitochondrial DNA methylation, and mitochondrial peptide expression. *Mitochondrion* 46, 22–29.
105. Salvi, A., Liu, H., & Salim, S. (2020). Involvement of oxidative stress and mitochondrial mechanisms in air pollution-related neurobiological impairments. *Neurobiol Stress* 12, 100205.
106. Szeto, H. H., & Liu, S. (2018). Cardiolipin-targeted peptides rejuvenate mitochondrial function, remodel mitochondria, and promote tissue regeneration during aging. *Archives of Biochemistry and Biophysics* 660, 137–148.

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