Aus dem Leibniz-Institut für Umweltmedizinische Forschung
Direktor: Univ.-Prof. Dr. med. Jean Krutmann

Role of odor identification on air pollution-induced cognitive impairment

Findings of the SALIA cohort

Dissertation
zur Erlangung des Grades einer Doktorin der Medizin
der Medizinischen Fakultät der
Heinrich-Heine-Universität Düsseldorf

vorgelegt von
Hannah Merz
2021

Als Inauguraldissertation gedruckt mit Genehmigung der Medizinischen Fakultät der Heinrich-Heine-Universität Düsseldorf

gez.:

Dekan:	Prof. Dr. med. Nikolaj Klöcker
Erstgutachterin:	Prof. Dr. med. Barbara Hoffmann
Zweitgutachter:	Prof. Dr. med. Christian Lange-Asschenfeldt

To all girls and women who dare to rise.

Zusammenfassung

Die Belastung mit Luftschadstoffen wurde zuvor bereits mit leichten kognitiven Störungen in Verbindung gebracht. Neurodegenerative Erkrankungen treten oft in Kombination mit Geruchsverlust auf. Unsere Studie bezog sich auf die Auswirkungen einer langfristigen Belastung durch Luftverschmutzung auf das olfaktorische System und auf leichte kognitive Störungen. Diese Beziehung könnte insbesondere im Hinblick auf den Geruchsverlust als Frühsymptom bei Alzheimer und Parkinson Patienten eine wichtige Rolle spielen.
Ziel dieser Studie war es, den Zusammenhang zwischen Luftverschmutzung, Geruchsidentifikation und leichten kognitiven Störungen zu analysieren.
Wir verwendeten Daten der SALIA-Kohorte (Study on the influence of Air pollution on Lung function, Inflammation, and Aging), in der die Auswirkungen von Luftverschmutzung auf die Gesundheit deutscher Frauen seit über 20 Jahren untersucht werden (Nachuntersuchungen 2008/2009 und 2012/13). Die Belastung mit Luftschadstoffen wurde durch Landnutzungsdaten und Regressionsmodellen der Wohnadresse jeder Probandin zugeordnet. Die Geruchsidentifikation wurde mit Sniffin‘ Sticks und leichte kognitive Störungen mit der neuropsychologischen CERAD-Plus Test Batterie untersucht.
Wir analysierten zunächst im Einzelnen die Zusammenhänge zwischen Luftschadstoffbelastung, Geruchsidentifikation und Kognition. Außerdem führten wir Mediations- und Interaktionsanalysen durch, mit der Fragestellung, ob luftverschmutzungsbedingte kognitive Störungen durch eine verminderte Geruchsidentifikation vermittelt wurden und ob sich der Zusammenhang zwischen Luftverschmutzung und leichten kognitiven Störungen in Abhängigkeit von Geruchsidentifikation veränderte.
Wir untersuchten 733 Frauen mit einem Durchschnittsalter von 73.6 Jahren bei den Nachuntersuchungen 2008/2009. Ein Anstieg von 1 Interquartilsabstand (IQR) von Feinstaub der Größe PM_{10} zeigte keine Assoziation mit einer verminderten Geruchsidentifikation (z.B. p-Wert = 0.731, β = 0.04 (95%-CI −0.19; 0.28)). Ein Anstieg von 1 Punkt im Geruchsidentifikationstest zeigte einen Zusammenhang mit einem Anstieg der CERAD-Plus Test Gesamtpunktzahl (p-Wert <0.001, β = 0.76 (95%-CI 0.47; 1.05)). Geruchsidentifikation vermittelte den Zusammenhang zwischen Luftverschmutzung und Kognition nicht. Die Interaktionsanalyse zeigte aber, dass sich der Zusammenhang zwischen langfristiger Luftverschmutzung und kognitiver Beeinträchtigung in Abhängigkeit von Geruchsidentifikation veränderte (z.B. NO_2: p-Wert = 0.028 für 2012/2013).
Unsere Studie lieferte keine Hinweise für einen Zusammenhang zwischen der Exposition gegenüber Luftverschmutzung und einer verminderten Geruchsidentifikation, aber es zeigte sich ein Zusammenhang zwischen verminderter Geruchsidentifikation und leichten kognitiven Störungen. Die Mediationsanalyse wurde durchgeführt, um die A-Priori-Hypothese zu beantworten. In der Interaktionsanalyse veränderte sich der Zusammenhang zwischen Luftverschmutzung und leichten kognitiven Störungen in Abhängigkeit von Geruchsidentifikation. Unsere Studie liefert daher erste Hinweise für eine Interaktion zwischen langfristiger Belastung durch Luftverschmutzung und verminderter Geruchsidentifikation in Bezug auf leichte kognitive Störungen in einer Kohorte älterer Frauen, die seit über 20 Jahren untersucht werden.

Abstract

Air pollution exposure has been associated with mild cognitive impairment (MCI). Neurodegenerative diseases often occur in combination with olfactory loss. Our study investigates the effects of long-term air pollution exposure on the olfactory system and cognition. This association could play an important role with respect to olfactory loss as an early symptom in Alzheimer's and Parkinson's patients.
The goal of this study was to investigate possible associations between long-term air pollution exposure, reduced odor identification, and MCI.
Our study was carried out using data of the SALIA (Study on the influence of Air pollution on Lung function, Inflammation, and Aging) cohort. The SALIA cohort has been used to investigate air pollution and its health effects on older women in Germany for over 20 years. Long-term exposure to air pollution was assessed and assigned individually to the home addresses of each study participant by land use regression models. Odor identification was assessed using the Sniffin' Sticks test and the assessment of cognition was carried out by using the neuropsychological CERAD-Plus test battery.
First, we examined the association between air pollution exposure and reduced odor identification. Second, we analyzed the association between reduced odor identification and cognitive impairment. Third, we investigated the association between exposure to air pollution and cognitive impairment. Subsequently, we performed a mediation analysis to examine whether odor identification functioned as a mediator on the pathway between air pollution exposure and cognitive impairment. We also carried out an interaction analysis to investigate whether this association between air pollution exposure and cognitive impairment changed depending on odor identification as a moderator.
We assessed 733 women with a mean age of 73.6 years in 2008/2009. An increase of 1 interquartile range (IQR) of particulate matter (PM) revealed no association with a reduced odor identification (e.g. PM_{10}: p-value = 0.731, β = 0.04 (95%-CI −0.19; 0.28)). An increase of 1 point in the odor identification test was significantly associated with an increase in the CERAD-Plus test total score for the follow-up investigations (p-value <0.001, β = 0.76 (95%-CI 0.47; 1.05). Odor identification was not a mediator on the pathway between long-term air pollution exposure and cognitive impairment in the mediation analysis. The results in the interaction analysis indicated that the association between air pollution exposure and cognitive impairment changed depending on odor identification as a moderator (e.g. NO_2: p-value = 0.028 in 2012/2013). Long-term exposure to air pollution and reduced odor identification also interacted with area-specific cognitive abilities in other CERAD subtests.
We were unable to provide evidence of an association between air pollution exposure and reduced odor identification. Then again, reduced odor identification was significantly associated with cognitive impairment. The mediation analysis was carried out to answer the a priori hypothesis. Air pollution-induced cognitive impairment changed depending on odor identification as a moderator in the interaction analysis. To the best of our knowledge, the evidence that long-term air pollution exposure and reduced odor identification interact when it comes to influencing cognition has not yet been researched in this particular setting, namely a population-based cohort of elderly women with a study period of more than 20 years.

Table of contents

List of abbreviations

AD	Alzheimer's disease
B-ADL	Bayer Activities of Daily Living scale
BBB	Blood-brain barrier
BC	Black carbon
BNT	Boston Naming Test
BMI	Body-Mass-Index
ADS	General Depression Scale, German: Allgemeine Depressionsskala
CERAD	Consortium to Establish a Registry for Alzheimer's Disease
95%-CI	95%-confidence interval
CNS	Central nervous system
CO	Carbon monoxide
COPD	Chronic obstructive pulmonary disease
EEA	European Environment Agency
ESCAPE	European Study of Cohorts for Air Pollution Effects
GBD	Global Burden of Disease
GIS	Geographic Information Systems
IARC	International Agency for Research on Cancer
ICD	International Classification of Diseases
IQR	Interquartile range
IUF	Leibniz-Research Institute for Environmental Medicine, German: Leibniz-Institut für Umweltmedizinische Forschung
LUR	Land use regression
MCI	Mild cognitive impairment
MMSE	Mini Mental State Examination
Mn	Manganese
N	Sample size
NH_3	Ammonia
NO	Nitrogen oxide
NO_2	Nitrogen dioxide
NO_x	Nitrogen oxides
NRW	North Rhine-Westphalia
PM	Particulate matter
$PM_{0.1}$	Particulate matter with an aerodynamic diameter of 0.1 µm or less
$PM_{2.5}$	Particulate matter with an aerodynamic diameter of 2.5 µm or less
$PM_{2.5}$abs	Filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less
PM_{10}	Particulate matter with an aerodynamic diameter of 10 µm or less
PNS	Peripheral nervous system
PD	Parkinson's disease

RR	Residuals of the regression model that best reflect the influences of the demographic variables
SALIA	Study on the influence of Air pollution on Lung function, Inflammation, and Aging
SD	Standard deviation
SLCP	Short-lived climate pollutants
SO_2	Sulfur dioxide
TSP	Total suspended particles up to a diameter of 30 µm
UFP	Ultrafine particles with a diameter of <0.1 µm
U.S.A.	United States of America
VCP	Visuo-construction performance
VOCs	Volatile organic compounds
WHO	World Health Organization

List of figures

List of tables

1 Introduction

1.1 Motivation

We breathe as long as we live and most of the time without even thinking about it. Breathing is life-sustaining and not optional for anyone. Consequently, air is the one substance that is absorbed the most and circulates constantly in our body (Künzli, Perez and Rapp, 2010).

However, the air we breathe today has changed drastically. We are exposed to a wide range of different air pollutants which are of natural or of anthropogenic origin. The proportion of the latter exceeded particularly in recent years. Anthropogenic air pollutants are caused by the expanding use of fossil energy sources in traffic and energy generation for domestic and industrial purposes. In the past, air pollution has often been seen as a serious, but localized problem in highly industrialized and densely populated cities. The “Great Smog of 1952” in London was one of the first major events raising public awareness on the medical danger for humans to breath in heavily contaminated air. During the incident, London was trapped under thick smoke for days and several thousands died as a consequence of the acute and persistent effects of air pollution (Logan, 1953). Since then, the concentrations of air pollutants in the atmosphere of our latitude have improved significantly due to political initiatives and strict legislations (World Health Organization, 2005, 2016; European Environment Agency, 2019). However, the most polluting industries have also been relocated to countries with less stringent environmental and air pollution regulations. Anthropogenic air pollution is a global issue. The proportion of air pollutants increases due to an ever greater up-take of motorized traffic, factory farming, high-intensity agriculture, and growing population. Living near major roads or big cities implies higher exposure to harmful concentrations of air pollutants. However, it has to be noted, that the distinction between “dirty” cities and “clean” country life is not clear, because air pollutants are transported over long distances in the atmosphere and secondary pollutants, e.g. ground-level ozone is generated everywhere by photochemical reactions. It has also been shown that even lower air pollution levels are associated with harmful consequences.

Air pollution is often less visible nowadays, which is why understanding and communicating the associated health risks has become a greater challenge. For several years, studies have been investigating the association between air pollution and health. Air pollution is considered a significant environmental risk to health entailing a threat to both, individuals and public health. According to the WHO (World Health Organization) and the GBD (Global Burden of Disease) project, ambient (outdoor) air pollution in urban and rural areas is one of the most important risk factor for morbidity and mortality (World Health Organization, 2016; Gakidou *et al.*, 2017). Today, the association between air pollution and cardiopulmonary diseases is widely acknowledged and research has also linked air pollution to many other health aspects (World Health Organization, 2005). Despite the increasing incidence of cardiopulmonary diseases in industrialized countries and a growing body of evidence indicating the harmful effects of air pollution on the brain, further research is required in order to better understand the association between air pollution and human health. The health effects of air pollution appear to be particularly dangerous to the youngest and oldest members of our society. In view of the demographic change of the population in industrialized countries towards a higher proportion of elderly, a special focus should be placed on the effects of air pollution after lifelong exposure and in relation to age-specific diseases (European Environment Agency, 2019).

The incidence of neurodegenerative diseases increases in the elderly, and such diseases often occur in combination with olfactory loss. Our study investigated the association between long-term air pollution exposure, reduced odor identification, and cognitive impairment in a cohort of older women. When breathing through the nose, air pollutants come into direct contact with the olfactory system. Anatomically, the outer components of the olfactory system are in particularly close contact with the brain. This might open up a possible transport route for air pollutants to gain access to the brain via the olfactory nerve. Cohort studies, such as ours, are particularly useful for studying chronic changes in health related to long-term air pollution under real conditions and adjustment of multiple risk factors. The association between air pollution, odor identification, and cognition could be important, especially with respect to olfactory loss as an early symptom in both, Alzheimer's and Parkinson's patients.

1.2 Theoretical background

1.2.1 Air pollution

The term air pollution refers to contamination of the indoor and outdoor environment by chemical, physical or biological agents that change the natural characteristics of the atmosphere. Air pollution is an ubiquitous phenomenon of complex origin. In order to better understand this phenomenon and its effects on health, it is necessary to understand air pollution as a mixture of different pollutants that interact with each other. In our study, we mainly focused on pollution of the ambient air.

When it comes to air pollution, climate change must also be taken into account because these are closely related. Air pollutant emissions can change the climate, e.g. ozone and black carbon (BC) contribute to warm, while particle sulfates contribute to cool the earth's atmosphere. Conversely, climate change can severely affect local air quality, and the warming of the atmosphere associated with climate change can lead to an increase in ground-level ozone in many regions. Thus, air quality affects climate change and vice versa (Loft, 2009; Edenhofer *et al.*, 2014).

A better understanding of the different sources and processes responsible for pollutant emissions is of great importance. In particular, natural processes and emissions caused by human activities are involved. The contribution of the latter has increased rapidly and exceeds natural sources by far, especially since industrialization. Primary pollutants are directly generated and released into the atmosphere immediately afterwards without taking part in other chemical reactions. This group includes gases, e.g. carbon monoxide (CO) which arises from burning fossil fuels. Secondary pollutants, on the other hand, are not generated directly. They arise from existing primary pollutants that interact in the atmosphere, such as ground-level ozone which is generated by chemical reactions of primary pollutants with heat and sunlight. Some pollutants, such as particulate matter (PM) can be both, primary and secondary. Visible air pollution is often referred to as smog and is made up of a wide variety of these compounds (World Health Organization, 2005).

1.2.1.1 Types of air pollutants

The WHO lists the following pollutants from ambient air pollution: soot, BC, CO, ground-level ozone, nitrogen dioxide (NO_2), PM, and sulfur dioxide (SO_2). This list is supplemented by ammonia (NH_3), volatile organic compounds (VOCs), and heavy metal emissions (World Health Organization, 2005).

Soot is a generic term for carbon particles that form at high temperatures through gas phase processes (e.g. diesel engines). BC is a dark residue that results from the incomplete combustion of hydrocarbon fuels. The term refers to the light-absorbing components of aerosols that contain more elemental carbon. It is a major part of $PM_{2.5}$ and belongs to the group of short-lived climate pollutants (SLCP) which are one of the largest contributors to global warming after CO_2. Due to the close physical and chemical similarity to soot, BC is also associated with various health risks (Janssen *et al.*, 2012).

CO is a colorless, odorless, and tasteless gas with a density slightly below that of air. It can be dangerous for humans in high concentrations. However, long-term exposure to lower concentrations might also be harmful (Allred *et al.*, 1989). One of the largest CO sources is incomplete combustion of gasoline in road traffic. Thus, CO is measured frequently in the atmosphere of urban areas. The incineration of different other carbon-containing fuels lead to a reaction in which some of the carbon contained oxidizes to CO_2 (World Health Organization, 2005).

Ground-level ozone is a component of photochemical smog and considered an important health risk. Associations with breathing problems and respiratory diseases are being researched (Mitis, Iavarone and Martuzzi, 2007; National Research Council, 2008). In addition to its impacts on health, ground-level ozone is also one of the most important greenhouse gases. It belongs to the group of SLCP and is generated secondarily when CO, methane, or VOCs oxidize in the presence of nitrogen oxides (NO_x) and sunlight. Thus, the highest ozone levels occur in sunny weather. NO_x and VOCs are produced by motor traffic exhaust, industrial plants, and chemical solvents while the main methane sources include waste, fossil fuels, and agriculture (Künzli, Perez and Rapp, 2010).

NH_3 is mainly an animal by-product that arises from the inefficient conversion of nitrogenous feed. The main source of emissions is factory farming (Künzli, Perez and Rapp, 2010).

Nitrogen monoxide (NO) and NO_2 belong to the NO_x family. There is a growing body of evidence indicating an association between NO_2 and bronchitis, asthma, respiratory infections, general lung function and growth. Evidence also points to an association with premature mortality due to cardiovascular and respiratory diseases (Gauderman *et al.*, 2005; Gehring *et al.*, 2006; Brunekreef, 2007). NO and NO_2 contribute to the formation of PM, smog, and ground-level ozone. Therefore, these gases are considered highly relevant when it comes to air pollution. NO_2 emissions arise when nitrogen is converted into NO_x during combustion. Coal contains a lot of nitrogen compared to oil and gas and is particularly important in this context. Additionally, an important reaction between nitrogen and oxygen takes place in high-temperature combustion. This explains why power generation, industry, and transport are predominant sources of emissions (World Health Organization, 2005; European Environment Agency, 2019).

Solid and liquid particles of organic and inorganic substances suspended in the air form a complex mixture which is collectively referred to as PM. The various components include water, sulfate, nitrate, NH_3, and other organic chemicals, metals, soil and dust particles. The basis for the classification of PM is the aerodynamic diameter of these particles. A distinction is made between total suspended particles (= TSP, for all particles up to a diameter of 30 µm), PM_{10} (with a diameter of ≤10 µm), $PM_{2.5}$ (with a diameter of ≤2.5 µm), and ultrafine particles or $PM_{0.1}$ (UFP, with a diameter of <0.1 µm). A large number of toxicological and human medical studies indicate that PM contribute to the development of health problems (Ackermann-Liebrich *et al.*, 1997; Braun-Fahrländer *et al.*, 1997; Atkinson *et al.*, 1999; Dominici *et al.*, 2003; Mar *et al.*, 2005; Schikowski *et al.*, 2005; Analitis *et al.*, 2006; Hoffmann *et al.*, 2007; Yitshak-Sade *et al.*, 2019). The different size fractions can absorb chemical substances on their surface. PM also remain in the atmosphere longer than other pollutants and are transported over much greater distances. Recent studies suggest that particles may migrate into the small airways and alveoli, penetrate the blood-air barrier in the alveoli, and enter the bloodstream. The categories also help identify the sources and processes involved in the formation of these particles. The coarser fraction (TSP) mainly arises when dust or soil is whirled up by roads, agriculture, mining, storms, or volcanic activities and suspended in the ambient

air. Sea salt, pollen, mold, spores, and other biological material are also assigned to this category. Fine particles (mainly PM_{10} and $PM_{2.5}$) arise as direct emissions. The main sources are combustion processes, such as the burning of gasoline and diesel by motor vehicles, of solid fuels, such as coal, lignite, heavy oil, and biomass to generate electricity, as well as other industrial processes, such as building, mining, melting, and the production of cement, steel, ceramics, and bricks. UFP generally arise as direct emissions from combustion processes and photochemical reactions in the air. Primary UFP have a very limited lifespan (minutes to hours). Larger, more complex aggregates of up to 1 µm form quickly in coagulation and condensation reactions (Künzli, Perez and Rapp, 2010; European Environment Agency, 2019).

SO_2 is a colorless gas with a pungent odor. Exposure to SO_2 has been associated with impaired respiratory function and contributes to an increased risk of infection (Hedley *et al.*, 2002). Sulfur particles are a component of natural raw materials. On combustion, the contained sulfur is converted to SO_2 and released. Heating, power generation, and motor vehicles are the main sources of emissions (World Health Organization, 2005).

VOCs is a collective term for mixtures of different compositions. Basically, it includes all organic substances that evaporate easily in ambient air and escape into the atmosphere. VOCs also include different carbon molecules, such as aldehydes, ketones, or light hydrocarbons. Therefore, the main sources of emissions are motor vehicles, solvents, and industry in general (Künzli, Perez and Rapp, 2010).

Emissions of heavy metals (e.g. lead) are another source of air pollution associated with serious health effects. Lead is a natural raw material that can be found in industrially manufactured products. Even after the introduction of cleaner fuels and technologies for emission control, the main sources of lead emissions have always been motor vehicles and industrial plants (Tan *et al.*, 2016).

Air pollution levels are generally expressed in terms of mass in µg x m^{-3}. For this study, we focused mainly on PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2. Nonetheless, it should be emphasized again that air pollution is always a mixture of different interacting pollutants.

1.2.1.2 Sources of air pollution

Many gases and particles detected in the atmosphere arise in natural processes. Trees and other plants release biogenic VOCs, volcanic eruptions dissolve ashes, ocean foam and wind swirl up soil particles, and dust storms lead to increased PM concentrations in arid areas, but also wherever dust is transported due to climatic conditions. Other natural emission sources, like forest fires, occur occasionally. As a result of climate change, such natural emissions and weather conditions are becoming increasingly extreme and occur with a higher frequency (Künzli, Perez and Rapp, 2010).

Human sources of air pollution are particularly important. According to the European Environment Agency (EEA), the main sources of air pollution emissions are transport, commercial, institutional and households, energy production and distribution, industry, agriculture, and waste (European Environment Agency, 2019).

Road traffic is a major source of emissions of many anthropogenic air pollutants. The vast majority of motor vehicles are driven by internal combustion engines in which gasoline or diesel is burned. Most modern vehicle engines have a catalytic converter through which exhaust gases pass prior to emission, but air pollutants are still released. The EEA calculated that around 40% of primary $PM_{2.5}$ emissions come from transport (European Environment Agency, 2020). However, traffic-related air pollution is not only caused by exhaust gases. Particles are also whirled up by the brakes, the abrasion of the road surface, and the mechanical wear of the tires. In contrast to other sources of air pollutants, emissions from motor traffic are mobile and occur in the immediate vicinity of the places where people live and work.

Fossil fuels are burned in refineries, industrial and power plants, as well as for heating and cooking in private households. These form another important emission source and take place stationary. Emissions arise not only during production, but also during the distribution of energy. Other sources of emission include the burning of biomass, waste incineration, and combustion processes with high temperatures. These processes produce air pollutants when materials contain components, such as sulfur, coal, and oil (World Health Organization, 2005; Künzli, Perez and Rapp, 2010; European Environment Agency, 2019).

In view of a growing population with all its implied consequences and impacts, these anthropological sources of air pollutants continue to come into focus. In our study, we took the immediate vicinity of the home address of our study participants into account when assessing air pollution. As already mentioned above, anthropologically caused sources of air pollutants are of particular importance in this direct environment.

1.2.2 Health effects

Many studies have confirmed that exposure to air pollution is associated with a variety of adverse health effects (WHO, Henschel and Chan, 2013). Research initially focused on acute effects attributed to short-term exposure to air pollution. A variety of outcomes, such as hospitalization due to respiratory, cardiovascular, and other problems are associated with short-term exposure to air pollution. Long-term exposure to air pollution and its association with chronic health effects has only recently been investigated as it usually requires large study populations and lots of time. However, long-term exposure to moderate levels of air pollution is associated with adverse health effects, such as incidence and prevalence of chronic cardiopulmonary diseases (Schulz *et al.*, 2019).

In particular, epidemiological studies provide evidence on the effects of air pollution on health. Furthermore, the studies can be designed in such a way that acute, subacute and chronic effects can be investigated in the general population, but also in selected groups with an increased or decreased susceptibility. None of the health effects associated with air pollution are specific to air pollution exposure. Many other factors may cause or contribute to identical or similar health problems. Short-term fluctuations in air pollution and daily changing health problems must be distinguished from the long-term effects of air pollution. As a result, epidemiological studies need to adjust for other important risk factors. Meanwhile, experimental studies provide key information to improve our knowledge of the mechanisms underlying the association between air pollutants and health effects.

It should be mentioned that the adverse health effects of air pollution are probably due to a mixture of various air pollutants. Chronic health effects are likely to be caused by long-term cumulative exposure to a mixture of air pollutants (World Health Organization, 2005; Künzli, Perez and Rapp, 2010). We examined the association between air pollution and reduced odor identification and cognitive impairment as chronic health effects.

1.2.2.1 Public health

The variety of adverse health effects associated with exposure to air pollution affects not only individuals, but also threatens public health.

Unlike many preventable risk factors, exposure to air pollution is difficult for individuals to avoid. Basically, there are no unexposed people and especially those living in urban areas are constantly exposed to high concentrations of air pollutants. From a public health perspective, this is the main reason for a paradox in relation to air pollution: the individual carries only a small relative risk – while the risk to public health is much greater. Thus, the risk of being affected by the most common health effects of air pollution is relatively small. Nevertheless, the absolute number of people affected remains substantial due to widespread exposure and our inability to selectively protect vulnerable groups. These vulnerable groups are at higher risk of suffering serious health damage from air pollutants and include small children, the poor, the sick, and those with low levels of education. Vulnerability is determined not only by personal characteristics, such as age, health, diet, and genetic makeup, but also by environmental factors, such as exposure characteristics, living conditions, and environmental background (Künzli *et al.*, 2000; Cohen *et al.*, 2004; Gauderman *et al.*, 2005; Mitis, Iavarone and Martuzzi, 2007; Medina *et al.*, 2009; Jacobs *et al.*, 2010).

The overall impact of air pollution on public health is less likely to be dominated by serious health damages of individuals, but rather by mild health effects of many others, such as subclinical and subtle symptomatic events. However, many epidemiological studies and risk analyses examine the serious health consequences (including an increased mortality risk and reduced life expectancy), since these data are often already routinely categorized and generally available (World Health Organization, 2005).

1.2.2.2 Overview of adverse health effects

Since the respiratory tract is the primary entry point for air pollutants into the human body, investigations were first carried out on respiratory health. With further insight into the systemic effects of air pollution, the influence of air pollutants on the cardiovascular system was recognized. Today's research area covers many more health sectors associated with exposure to air pollution, including inflammatory and oxidative stress-related processes, as well as cancer, reproduction, mortality, and cognition.

The association between exposure to air pollution and respiratory effects is well investigated because intuitively the lung seemed to be an obvious organ to be affected by air pollution. A large number of published studies indicate associations between air pollution and respiratory health in children. They seem to be particularly vulnerable to harmful effects of air pollution (Schwartz, 2004; Gauderman *et al.*, 2005). Evidence from studies with adults point in the same direction, namely that deficits in lung function and increased symptoms of obstructive airway diseases correlate with air pollution exposure (Pope, 1989; Atkinson *et al.*, 1999; Schikowski *et al.*, 2005; Analitis *et al.*, 2006; Götschi *et al.*, 2008; Adam *et al.*, 2015).

Exposure to air pollutants has been associated with different cardiovascular effects. The main pathology in the context of cardiovascular diseases is atherosclerosis which develops over a longer period of time. Air pollution has been associated with chronic calcification of the coronary arteries. It is currently suspected that it is also associated with increased inflammation and coagulation factors. However, further steps on the pathophysiological pathway to heart disease have yet to be identified (Peters, 2005; Gehring *et al.*, 2006; Hoffmann *et al.*, 2007; Miller *et al.*, 2007; Brook, 2008; Mills *et al.*, 2009).

Peters *et al.* (1997) were among the first to investigate the association between air pollution and blood markers by comparing measurements of plasma viscosity in humans exposed to different levels of air pollution. A large variety of studies followed, all focusing on different outcomes, such as inflammation, coagulation, and endothelial function. It is difficult to draw a single conclusion. However, this also indicates that the associations between air pollution and blood markers are

manifold (Seaton *et al.*, 1999; Peters, 2001; Brook *et al.*, 2002; Rückerl *et al.*, 2006, 2007; Briet *et al.*, 2007; Delfino *et al.*, 2009; Hildebrandt *et al.*, 2009).

The International Agency for Research on Cancer (IARC) divides air pollutants into different categories based on different studies. For adults, death from lung cancer is of primary concern in relation to air pollution. In order to make statements in this context, population-based studies with large population samples and long follow-up periods are used. The risk of lung cancer is associated with exposure to air pollution in most of these studies (Pope *et al.*, 2002; Nafstad *et al.*, 2003; Edwards *et al.*, 2006; Chen *et al.*, 2015).

The pollution of the ambient air during pregnancy seems to be associated with disadvantages for mother and child. There is evidence that air pollution is associated with intrauterine growth restrictions, low birth weight, and preterm delivery (Maisonet *et al.*, 2004; Wilhelm and Ritz, 2005).

Mortality is one of the most commonly investigated outcomes related to air pollution over time, as it is routinely catalogued and generally available. Death is an acute event, whereas life expectancy or time to death is the result of both, acute and chronic pathologies. It is particularly difficult to make a clear distinction between short-term and long-term consequences of air pollution in terms of mortality since acute and chronic diseases are interrelated. However, many studies have shown that the overall impact of long-term pollution on mortality is far greater than the proportion attributed to acute pollution (Logan, 1953; Waller, Brooks and Adler, 1973; Dockery *et al.*, 1993; Nemery, Hoet and Nemmar, 2001; Brunekreef, 2007; Beelen *et al.*, 2008; Yitshak-Sade *et al.*, 2019).

1.2.2.3 Neurological effects

Recently, evidence suggesting an association between air pollution and neurological effects is increasing (Peters *et al.*, 2006; Guxens and Sunyer, 2012). Early approaches from the 1980s detected an increase in strokes in people exposed to indoor coal fumes, leading to the initial hypothesis of the deleterious effects of air pollution on the brain (Zhang, Yu and Zhou, 1988). Associations between air pollution and stroke have later been reproduced (Wellenius, Schwartz and Mittleman, 2005; Chen, 2010).

The first histological evidence for the association between exposure to complex mixtures of air pollution and neurotoxicity was provided by a Mexican research team. They found inflammatory processes in the tissue of the central nervous system (CNS) of mongrel dogs that were chronically exposed to the heavily polluted metropolitan air in Mexico City. They described not only early and significant inflammatory processes, but also the stimulation of stress protein reactions, the premature expression of diffuse amyloid plaques, a change in the blood-brain barrier (BBB), and increased DNA damage in the olfactory mucosa, olfactory bulb, frontal cortex, and hippocampus (Calderón-Garcidueñas *et al.*, 2002, 2003). These early approaches were confirmed and expanded. Animal studies with very different approaches showed an increase in pro-inflammatory cytokines and other markers of oxidative stress which indicated a generalized inflammation of the brain tissue following high levels of air pollutants (Campbell *et al.*, 2005; Veronesi *et al.*, 2005; Levesque *et al.*, 2011; Guxens and Sunyer, 2012; Fagundes *et al.*, 2015). Air pollution-induced neurotoxicity was also associated with changes in the BBB, activation of microglia, endothelial cell damage, and neuroinflammation as confirmed by in-vitro studies (Block *et al.*, 2004; Hartz *et al.*, 2008; Davis *et al.*, 2013; Costa *et al.*, 2014). The previously mentioned Mexican research team also investigated the neurotoxic effects caused by air pollutants in human brain tissue. The number of inflammatory processes and deposits of amyloid differed significantly in the brains of deceased who were exposed to high levels of air pollution during their lifetime compared to controls (Calderón-Garcidueñas *et al.*, 2004). Investigated pathologies were similar to those of neurodegenerative diseases, such as Alzheimer's disease (AD) and Parkinson's disease (PD), with olfactory loss being one of the first

symptoms of both (Peters *et al.*, 2006; Block *et al.*, 2012; Lucchini, Dorman, *et al.*, 2012).

The nasal cavity is a frequent entry portal for inhaled noxae, which is why the olfactory system appears to be a vulnerable target for toxicological damage. In animal studies, an association between smoking and changes in the olfactory epithelium was early established (Matulionis, 1974; Vent *et al.*, 2003, 2004). Epidemiological studies confirmed that both, smoking and passive smoking, reduced the ability to identify odors. 638 subjects with different smoking status were tested with 40 odors in order to better assess the effects of cigarette smoke on the sense of smell. Depending on the dose, current smokers and past smokers had a reduced ability to recognize odors, and smokers were also more likely to suffer from olfactory deficits (Frye, Schwartz and Doty, 1990). On the one hand, smoking seemed to be an important covariate in the context of research on olfactory loss, on the other hand, these results provided the first indications that inhaled pollutants may have an impact on the olfactory system. Since then, knowledge in this research field has increased and the influence of smoking on the olfactory system has been confirmed (Ahlström *et al.*, 1987; Ajmani *et al.*, 2017). These studies were crucial for further investigations into the effects of air pollutants on the sense of smell, but also on cognition.

Changes in the olfactory barrier could play a role in inflammation of the brain, as this may help air pollutants to get easier access to the brain tissue. Inhaled noxae are associated with various health risks. Research in this field focused also on chemically induced lesions of the human nose, olfactory nerve toxicity, nasal immunopathology, and carcinogenesis. Several studies have also shown an association between nasal cancer and different chemicals, e.g. formaldehyde (Halperin *et al.*, 1983; Holt, 1996). A study in mice even showed lesions of the nasal cavity after long-term ozone exposure (Herbert *et al.*, 1996). Another study examined the effects of atmospheric pollutants, such as acetone, benzene, ammonia, formaldehyde, and ozone on the olfactory epithelium of humans and animals using electroencephalography. The olfactory bulb turned out to be one of the most sensitive structures in this study (Bokina *et al.*, 1976). Chemoperception appears to represent a dominant window from the brain to the external environment. Therefore, nasal neurotoxicology is developing rapidly with

a special focus on the toxicity of the olfactory nerve (Feron *et al.*, 2001; Oberdörster *et al.*, 2004; Elder *et al.*, 2006).

Although some studies have already addressed the translocation of ambient air pollutants into brain tissue, the effects of air pollution on cognition have not yet been investigated as comprehensively. There are studies that point out the negative effects of traffic-related air pollution on children's cognitive abilities (Suglia *et al.*, 2007; Freire *et al.*, 2010; Calderón-Garcidueñas *et al.*, 2011). A growing number of studies have also highlighted the importance of air pollution for the cognitive function of adults. An exposure study suggested functional changes in electroencephalography in the human brain in response to diesel exhaust indicating a generalized cortical stress response (Crüts *et al.*, 2008). Ranft *et al.* (2009) were among the first to investigate the chronic effects of traffic-related air pollution on cognition in the elderly. Several other studies provided epidemiological data indicating the negative neurological effects of air pollutants in adults of different ages. Various air pollutants, such as ozone, BC, or the distance to the next main road, were associated with poorer performance of area-specific cognitive abilities, such as verbal learning, short-term memory, language and executive function (Chen and Schwartz, 2009; Power *et al.*, 2011, 2016; Guxens and Sunyer, 2012; Wellenius *et al.*, 2012; Weuve, 2012; Ailshire and Crimmins, 2014; Tonne *et al.*, 2014).

1.2.3 Possible transport routes for air pollutants

There are several approaches that provide important insights into how air pollutants gain access to the organism.

The association between air pollution and respiratory effects has been intensively researched. Particles appear to cause oxidative stress in the lungs which is related to local and systemic inflammation, as well as fever, fibrosis, and bronchitis. Interactions between air pollutants and respiratory neuronal afferents lead to an autonomous imbalance. Air pollutants also appear to be related to the formation of oxidative adducts in the epithelium which are associated with carcinogenesis (Brook, Brook and Rajagopalan, 2003; Donaldson *et al.*, 2003; Ailshire and Clarke, 2015). When epithelial cells in the lung tissue are damaged, the activity of intravascular macrophage-like cells is promoted. Macrophages and dendritic cells take up harmful particles through phagocytosis and initially transport them to the lymph nodes and into the venous blood circulation. PM and other particles may even penetrate directly from the respiratory tract into the systemic circulation via the blood-alveolar barrier (Guxens and Sunyer, 2012).

The mechanisms of how air pollution affects the cardiovascular system have been of interest for quite some time. Possible routes in connection with air pollution and the cardiovascular system appear to be directly or indirectly associated with oxidative stress and inflammation of the lung tissue. Air pollutants may stimulate pulmonary neural reflexes and lead to changes in the sympathetic and parasympathetic system. This may ultimately contribute to the instability of vascular plaques or initiating dysrhythmias (Hoffmann *et al.*, 2007; Brook *et al.*, 2010). Air pollution could irritate the lung tissue and trigger a systemic chain reaction (e.g. production of white blood cells, release of pro-oxidative and pro-inflammatory markers, including cytokines, acute phase proteins, and vasoactive hormones). As a result, effects on the heart rhythm, changes in endothelial function, activation and aggregation of blood platelets can be observed. Particles may even be transferred into the circulation, resulting in direct interaction with the cardiovascular system (Brook *et al.*, 2004, 2010; Mills *et al.*, 2009).

The olfactory mucosa is directly exposed to the external environment. Therefore, the olfactory neurons appear to be in direct contact with harmful air pollutants (Cullen and Leopold, 1999). This could serve as a possible transport route for air

pollutants between the external and the internal (e.g. the brain) along the olfactory nerve. Air pollutants may penetrate the protective BBB by damaging the olfactory barrier or by axonal transport to the olfactory bulb (Oberdörster *et al.*, 2004; Lucchini, Dorman, *et al.*, 2012). Experimental studies that examine this route along the olfactory nerve mostly focused on metals and often used radio-labeled isotypes. However, in recent years there have also been studies that focused on inhaled pollutants. Several studies indicate that manganese (Mn) nanoparticles or PM can be found in the olfactory bulb after instillation or inhalation (Tjälve and Henriksson, 1999; Sunderman, 2001; Elder *et al.*, 2006; Tin-Tin-Win-Shwe *et al.*, 2006). Other studies showed that PM applied to the olfactory mucosa was distributed throughout the brain after some time which was probably connected to the transport via the olfactory nerve (Oberdörster, Elder and Rinderknecht, 2009). A connection between reduced odor identification and PM became apparent on the basis of these mechanical approaches and was later supported by findings of epidemiological studies. These indicated a connection between occupational exposure and an impaired olfactory system (Doty, 2006; Gobba, 2006). Furthermore, a cross-sectional study of adults in cities in the U.S.A. (United States of America) recently showed an association between reduced odor identification and $PM_{2.5}$ in a general population (Ajmani *et al.*, 2016). At a cellular level, the production of cytokines could activate glia, apolipoprotein E, β-amyloid, reactive oxygen species, and nitric oxide synthase. This may be associated with pinocytosis, neuronal transport, direct neurotoxic effects, as well as oxidative and DNA damage which may ultimately lead to neuronal, synaptic loss, and cell death.

Air pollution is associated with a variety of negative CNS effects. These include cerebral vascular damage, neuroinflammation, and neurodegeneration. Various signaling transport routes and mechanisms underlying these effects are discussed (Brook *et al.*, 2004, 2010; Pope and Dockery, 2006; Block and Calderón-Garcidueñas, 2009; Mills *et al.*, 2009; Block *et al.*, 2012). Air pollutants and other particles enter the airways through mouth or nose by inhalation. Insoluble inhaled particles are enclosed in a secretory layer which forms a protective barrier against systemic absorption and further transport. The ciliated respiratory epithelium continuously transports this secretory layer towards the nasopharynx (Morgan and Monticello, 1990). If particles penetrate the blood-

alveolar barrier, they could reach the BBB with the bloodstream (Oberdörster *et al.*, 2004; Elder *et al.*, 2006). Epithelial cells of the lung tissue function as an important protective barrier. However, they could be damaged by air pollutants which subsequently leads to alveolar edema and infections of the respiratory tract (Calderón-Garcidueñas *et al.*, 2003; Block and Calderón-Garcidueñas, 2009). When the respiratory tract infection spreads to the whole organism, the immune system responds by producing inflammatory and oxidative stress mediators. It has been shown that circulating cytokines alter the cellular composition of innate immune cells in the brain and relate to various CNS effects (Rivest *et al.*, 2000; Campbell *et al.*, 2005, 2009; Veronesi *et al.*, 2005; D'Mello, Le and Swain, 2009).

Furthermore, cytokines come into contact with receptors of endothelial cells of the BBB. Here, they are associated with changes of tight junctions and transport proteins, resulting in dysfunction of the BBB. This allows particles to gain easier access to the brain and become mediators of oxidative stress. Oxidative stress in return has a direct negative impact on the DNA of the brain cells, neurons, and synapses (Calderón-Garcidueñas *et al.*, 2002; Block and Calderón-Garcidueñas, 2009).

1.2.4 Olfactory system

1.2.4.1 Anatomy

When inhaling particles, the olfactory system offers a hypothetical access to the brain. The anatomy should clarify whether this possible transport route to the brain is actually open to particles.

The olfactory system consists of 6 million peripheral bipolar receptor cells whose cell bodies, dendrites, and initial axon segments can be found within the olfactory mucosa. The olfactory mucosa is located in the roof of the nasal cavity and covers the cribriform plate. Thus, the olfactory mucosa is directly exposed to the external environment. When smelling, the air is mainly conducted from the nostrils to this region via a direct air flow connection anterior to the nasal concha. The olfactory mucosa contains specialized olfactory receptor cells, basal cells, supporting cells with apical microvilli, and glandular cells. The olfactory receptor cells with their axons are the primary afferent neurons of the olfactory tract. The lifespan of olfactory cells is a few weeks. After this time, they are regenerated by basal cells, which makes the olfactory mucosa one of the few places in the nervous system where new neurons are constantly emerging even in adulthood. Olfactory receptor cells belong to the bipolar cell type and are also referred to as primary sensory cells. This cell type serves as both, a receptor cell in the olfactory mucosa and a first-order neuron. This particular cell type projects an axon directly from the nasal cavity onto the olfactory bulb in the brain without any synapse intervening the aforementioned process. In contrast to receptor cells that are only sensor, bipolar cells can receive and transmit signals. Such cells express specific receptors that only respond to certain elements of different odors. The entire olfactory mucosa is covered by a film of mucus which mainly comes from Bowman’s glands. Odors diffuse in this mucus film and bind to specific receptor proteins in the cell membrane of olfactory cells (the first neuron of the olfactory system). Each olfactory cell has only one specialized receptor protein. Binding to these receptor proteins triggers a cascade of activations at the level of cell channels which ultimately induces an action potential. The induced action potential reaches the olfactory cortex via stations of the olfactory tract.

The central axons of the olfactory cells are combined in olfactory fibers. The olfactory fibers form the olfactory nerve (first cranial nerve), project through the

cribriform plate, and synapse within the olfactory bulb in the anterior cranial fossa. The olfactory bulb is a protuberance of the paleocortex and the first station in which the action potential is processed via synapses. Projecting cells (mitral and tuft cells) and interneurons (granule and periglomerular cells) are located there. The mitral cells are highly specialized and the second neuron of the olfactory tract. More than 1000 olfactory cell axons can form synapses with a single neuron. Dendrite plus synapse form the olfactory glomeruli. Axons of olfactory cells with the same receptor protein form olfactory glomeruli with only 1 or very few mitral cells. The basal axons of mitral and tuft cells form the olfactory tract and project the olfactory information onto different areas of the brain. They also form collaterals that project onto granule cells. Granule and periglomerular cells inhibit the activity of the mitral cells which leads to negative feedback within the olfactory bulb. As a result, less smell impressions are transmitted. Such inhibition processes are supposed to promote the formation of contrast which is important for precise odor identification. The olfactory tract is divided into a medial and lateral pathway in front of the anterior perforated substance. The medial pathway takes the medial olfactory tract to get to the septal area, the habenula of the epithalamus, the reticular formation, and the hippocampus. The lateral pathway uses the lateral olfactory tract to synapse on structures, like the anterior cortical nucleus of the amygdala, the thalamus, the piriform, the entorhinal and the orbitofrontal cortex. Additionally, the anterior olfactory nucleus is located in the olfactory tract. Its axons use the anterior commissure to reach the contralateral olfactory bulb and are part of the inhibiting efferences on the mitral and tuft cells.

The term olfactory cortex is not used uniformly. There is no specific olfactory area that can be clearly defined topographically, but rather a functional system that includes different brain areas that have various relationships with each other. The lateral pathway synapses on structures collectively termed as the primary olfactory cortex which are partly located in the basal cortex of the cerebrum. The lateral pathway projects onto the piriform cortex which encodes higher-order representations of odor quality, identity, and familiarity. This area is also associated with the learning and remembering of odors, as well as the coordination between olfactory information, vision, and taste. Connections to the amygdala may be associated with strong emotional responses to pleasant or unpleasant odors. The entorhinal cortex also receives olfactory information via

the lateral olfactory tract and is referred to as the main entrance to the hippocampus which is involved in learning skills and memory. There are also connections to the orbitofrontal cortex which is often referred to as the secondary olfactory cortex. The secondary olfactory cortex may also be related to the conscious perception and differentiation of odors. The limbic system and the insular cortex seem to be involved in the perception of odors. The medial pathway projects onto the septal region. Olfactory information transported via this pathway is unlikely to be involved in the conscious perception of odors. The septal region is located in the caudal medial side of the hemispheres, directly rostral of the third ventricle. There is also a connection to the habenula of the epithalamus and to the reticular formation which may be responsible for the awakening reaction of strong odors. A small part is connected to the hippocampus, which as part of the limbic system, is related to the creation of memories. The olfactory information reaches these areas via thalamus, hypothalamus, and brain stem (Doty, 2009; Aumüller *et al.*, 2014; Schünke *et al.*, 2018).

The olfactory system is connected to a large number of brain areas. This is also the reason why complex behavioral reactions can occur after odor stimuli, for example bad odors trigger nausea, whereas appetizing odors “make your mouth water” and why olfactory loss is plausible as an early symptom in neurodegenerative diseases, such as AD.

1.2.4.2 Olfactory loss

The nose, the nasal cavity, and the paranasal sinuses belong to the upper, air-conducting respiratory tract. The olfactory system fulfills important functions in connection with breathing and odor perception. Although often underestimated, olfaction is also crucial for quality of life, nutritional status, interpersonal communication, and safety. Many factors influence the olfactory system, like age, trauma, viral infections, exposure to toxic chemicals and nanoparticles, rhinosinusitis, nasal polyposis, and neurodegenerative diseases (Schubert *et al.*, 2009; Hüttenbrink *et al.*, 2013). In contrast to most animals, humans are microsmatic with a poor sense of smell. The low degree of odor perception and differentiation usually includes 7 basic odors: floral, ethereal, peppermint, musk, camphor, putrid, and pungent. A combination of these basic odors leads to the considerably larger number of all recognizable odors (approximately 10000) (Aumüller *et al.*, 2014).

Olfactory loss is common. Around 5% of the general population suffer from anosmia (complete lack of odor perception), with increasing prevalence in advancing age (Murphy *et al.*, 2002; Brämerson *et al.*, 2004). Reduced olfactory function is divided into quantitative and qualitative impairments. The quantitative classification includes complete (anosmia) or incomplete (partial anosmia, hyposmia, or microsmia) olfactory loss. In contrast, hyperosmia is a rare condition of abnormally hypersensitive olfactory function. In addition, distortions (dysosmia, e.g. a foul smell when smelling a rose) or spontaneous sensations (phantosmia, e.g. the presence of a smell when there is actually none) can also exist in the context of a qualitative impaired olfactory system. Reduced odor identification can occur regardless of a normal functioning olfactory system (olfactory agnosia). Subsequently, olfactory loss can be either bilateral or unilateral (Doty, 2009).

On the one hand, olfactory loss is a major burden on the quality of life (e.g. nutritional disorders or impaired flavor), on the other hand, there is also a reduced ability to detect dangers (e.g. gas leaks or fires) at an early stage. If the olfactory mucosa is markedly damaged, not only does the olfactory function disappear, but also the taste. Often, only simple somatosensory sensations and the perception of the primary taste qualities remain intact. Impairment of the olfactory system is also significant in connection with early detection and differential diagnosis of

neurodegenerative diseases (Santos *et al.*, 2004; Hummel and Nordin, 2005; Doty, 2009; Stevenson, 2009).

Reduced odor identification is clearly associated with mild cognitive impairment (MCI), a state in-between normal aging and neurodegenerative diseases. MCI is associated with a high risk of progression to AD or PD. Reduced olfactory function is also considered one of the earliest symptoms of neurodegenerative diseases, such as AD and PD. The deficit exists in 85–90% of the patients in the early stages of both diseases. Surprisingly, most AD and PD patients are unaware of their odor deficits before testing. Therefore, testing of olfactory function is used as an early detection tool, as an indicator of development, and as a predictor of mortality in these diseases. It should be noted that the extent of the reduced odor identification seems to be very weakly related to the disease stage, the severity of the other symptoms, and the results of neuropsychological tests (Doty, Deems and Stellar, 1988; Müller *et al.*, 2002; Hawkes, 2006; Doty, 2009; Wilson, Yu and Bennett, 2011; Devanand *et al.*, 2015; Zou *et al.*, 2016).

1.2.5 Cognitive changes in the elderly

The population development in Europe varies from country to country, but in general the continent's share of the world's population is decreasing. In addition, the average age of European citizens is the highest of all regions of the world, and this trend continues to increase even when considering migration (Bundeszentrale für politische Bildung, 2019; WHO Health Data, 2020).

There is no monocausal relationship between age and disease. Aging is not generally synonymous with disease, but the proportion of older citizens in society increases the likelihood of chronically degenerative and multiple diseases (multimorbidity). The most common diseases in the elderly include neurodegenerative diseases. In particular, the incidence of dementia increases exponentially with age. The increasing number of dementia patients is associated with increased economic costs. Not only the financial expenses for nursing and medical care should be mentioned, but also the enormous restrictions in quality and many other aspects of life for patients and their families in general (Kuhlmey and Schaeffer, 2008; Naegele *et al.*, 2013).

Early diagnosis is essential for the treatment success of dementia even though treatment options are generally limited. Therefore, the current state of research in this field also focuses intensely on the development of tools for early diagnosis. Neuropsychological testing of cognitive brain functions is of importance in this context, but also the knowledge and the detection of early symptoms. Neurodegenerative diseases often occur in combination with olfactory loss. Our study investigated the association between reduced odor identification and cognitive impairment after long-term exposure to air pollution. This association could play an important role, especially with respect to a further aging society and with olfactory loss as an early symptom in dementia patients (Doty *et al.*, 1991).

The term cognition is defined as any mental process, that involves dealing with knowledge through thought, experience, and the 5 senses: hearing, smell, taste, sight, and touch. When it comes to cognition, it is important to have a rough idea of the neuroanatomy and the cognitive changes associated with age. The following sections present common neurodegenerative diseases and the difference between accelerated and normal brain aging.

1.2.5.1 Brain anatomy

The brain serves as the sensory and nervous system for internal and external communication, as well as for perceiving processes inside or outside the organism (enteroception and exteroception). Due to these diverse and interlocking tasks, cognition is complex and can be classified in different ways. A brief overview of different aspects of neuroanatomy are given here, further details on this topic can be found elsewhere (Aumüller *et al.*, 2014; Rassow *et al.*, 2016; Schünke *et al.*, 2018).

The CNS macroscopically consists of 2 main tissue components. The white matter consists of glial cells, myelinated and unmyelinated nerve fibers. The action potential is transmitted there without synaptic processing of the neuronal information. The grey matter consists of glial cells, as well as the perikaryon, dendrites, and axons of neurons. The processing of neuronal information takes place there via synapses. The brain is divided into 2 hemispheres, which are in exchange with each other via the corpus callosum. Each hemisphere is further divided into several lobes: frontal, parietal, temporal, and occipital lobe. In addition, there is the island lobe on the lateral surface, the limbic lobe on the medial surface and the cerebellum on the lower surface of the brain. The brain stem consists of mesencephalon and rhombencephalon with pons and medulla oblongata. The surface of the mature brain is strongly contoured in the form of gyri and sulci. In the case of cerebral edema, the sulci become smaller, while in the case of brain atrophy (e.g. AD), they enlarge due to the loss of substance in the gyri. The surface of the brain is macroscopically divided into lobes, gyri, and sulci. However, differences were found between the macroscopically recognizable structures and the microscopic distribution of neurons. Sections of the cerebral cortex, that were microscopically structured in the same way, were designated as areas. The division into areas was initially based on the distribution of neurons in the different layers of the cortex and was hypothesized to represent the functional organization of the cortex. However, modern imaging methods have shown that certain functions can be assigned to many areas.

Since the CNS is very sensitive to external damage, it is surrounded by bones. There is an additional protective barrier between these bones and the CNS called meninges.

The meninges are divided into 3 layers from the outside in. The outer layer is called dura mater or hard meninges. It is a thick layer of tight collagenous connective tissue that can be divided in a periosteal and meningeal part. The middle layer is called the arachnoid mater and is thin, vascular-free, semi-transparent and has a stabilizing function. The inner layer is called pia mater and is in direct contact with the brain tissue. It consists of soft connective tissue and contains many blood vessels. The middle and inner layers are combined to form the soft meninges. The space between arachnoid and pia mater is called the subarachnoid space which is filled with cerebrospinal fluid and surrounds the brain for further protection. The subarachnoid space forms the outer cerebrospinal fluid space, but is connected to the inner cerebrospinal fluid space formed by the brain ventricles, via the paired lateral aperture and the unpaired median aperture of the fourth brain ventricle. The arterial and venous blood vessels of the brain tissue can also be found here.

At a cellular level, flat cells of the capillary endothelia are connected by “tight junctions” and represent the actual diffusion barrier. The paracellular passage of hydrophilic substances from the capillaries into the surrounding tissue and in the opposite direction is not possible in the CNS due to these “tight junctions”. Important hydrophilic substances that are essential for the CNS must be carried through the barrier using specific transport mechanisms. The BBB also prevents substances in the blood from accessing the brain tissue, such as toxins and bacteria that might disrupt the brain homeostasis. Capillaries without “tight junctions” can be found in the median eminence where hypothalamic control hormones are released into the portal vein of the anterior pituitary lobe. Other brain areas without “tight junctions” are the postremal area which is involved in triggering vomiting and must identify potentially toxic substances, or the neurohypophysis where oxytocin and aldehyde dehydrogenase are released into the blood via neurosecretion.

In summary, the neuroanatomy is complex and very different brain areas play a role in cognitive processes. Additionally, the brain is well protected from external influences, but there are also brain areas where substances from the blood can gain access to the brain tissue.

1.2.5.2 Normal brain aging

Everyone has a certain idea of aging which is culturally shaped and associated with stereotypes. Aging is often associated with increasing forgetfulness, physical and social loss, but also with wealth of experience and wisdom.

From an anatomical point of view, normal brain aging is associated with a decrease in brain weight, with signs of minimal to moderate atrophy of the subcortical and cortical cortex, with a decrease in the volume of the telencephalic lobes and white matter, and with a minimal to moderate enlargement of the ventricular system. Magnetic resonance imaging examinations of the brain of seniors often show random signal abnormalities in different areas, even when no symptoms appear clinically. However, minor lesions and changes in the neuroanatomy of seniors can also become clinically noticeable and affect very different cognitive functions (Creasey and Rapoport, 1985; Weis *et al.*, 2019).

Normal aging is related to changes in different cognitive areas. Thus, it is difficult to give an overview (Aebi, 2003; Harada, Natelson Love and Triebel, 2013).

Fluid intelligence refers to a person's innate ability to learn new information, while crystallized intelligence refers to skills and factual knowledge acquired over a lifespan. Only fluid intelligence seems to decrease with age. Seniors can often retain and expand their knowledge and perform routine actions without difficulty (crystallized intelligence), whereas new stimuli are processed more slowly and adaptation to new situations becomes increasingly difficult (fluid intelligence) (Horn and Cattell, 1967; Ryan, Sattler and Lopez, 2000; Lezak *et al.*, 2004).

Age-related problems in the working (short-term) memory usually appear when there is a rapid change between the management of old and the processing of new information. The long-term memory is split into declarative memory in which information is consciously stored and further divided into episodic (personal experiences in a spatial-temporal context) and semantic memory (context-independent knowledge). The non-declarative memory, on the other hand, stores experience-related behavior unrelated to consciousness, e.g. classic conditioning, cognitive and motor skills. Episodic memory impairment is one of the most common cognitive changes in the elderly. Seniors seem to encode new information less efficiently and have more problems retrieving learned information. Neither semantic memory, nor non-declarative memory seem to

decrease related to age (Salthouse, Babcock and Shaw, 1991; Luszcz and Bryana, 1999; Craik and Rose, 2012).

Selective attention is the ability to react to relevant stimuli and not to be distracted by irrelevant stimuli and is usually assessed with election-reaction tasks. Permanent attention is the ability to consciously maintain attention and to concentrate on a certain task over a longer period of time. Shared attention is the ability to focus on several things at the same time. Selective and permanent attention decrease with age if the relevant stimuli are unknown, difficult to distinguish from distractors, or if several characteristics appear at the same time. Neuropsychological tests evaluate shared attention with tasks in which several characteristics have to be taken into account simultaneously. Performance decreases with increasing complexity of such tasks in the elderly (Salthouse *et al.*, 1995; Humphrey and Kramer, 1999; Craik, Luo and Sakuta, 2010).

Language comprehension is complex and involves vocabulary, lexical knowledge, semantic networking, and the use of linguistic rules. Difficulties with comprehension are mostly associated with age if the length or syntactic complexity of a sentence increases. Age-related problems in language production often manifest in word-finding disorders and are tested with semantic fluency. The general language skills do not usually decrease with age (Snowdon *et al.*, 1996; Kemper, Herman and Lian, 2003; Singh-Manoux *et al.*, 2012).

Executive function summarizes higher-order cognitive processes, e.g. planning and execution of actions. Neuropsychological test methods for investigating executive function are widespread and seniors consistently perform worse than younger people. Impairment of executive function associated with age can manifest in increased routine action, reluctance to change, reduced cognitive flexibility, lack of initiative and activity (Rabbitt and Lowe, 2000; Oosterman *et al.*, 2010; Bock, Haeger and Voelcker-Rehage, 2019).

Visuo-constructive function is a generic term and summarizes skills that combine the reception and execution of information. These skills are tested using visual perception tasks in which fragmented images are to be recognized, or visuo-constructive performance (VCP) in which figures are to be drawn. Age-related impairments of the performance have been demonstrated in several studies (Capitani *et al.*, 1988; La Rue *et al.*, 1992; Howieson *et al.*, 1993).

1.2.5.3 Accelerated brain aging

Neurodegenerative diseases often develop very slowly which makes the diagnosis considerably more difficult. Additionally, those affected by memory loss, attention and concentration difficulties often do not notice the cognitive impairment themselves.

The diagnosis of MCI is made if the cognitive impairment exceeds the deficits that occur due to normal age changes. MCI is encoded with the ICD-10-GM-Code F06.7 and is called “Mild Cognitive Disorder” according to the International Statistical Classification of Diseases and Related Health Problems, 10th Revision, German Modification, Version 2020 (*DIMDI - ICD-10-GM Version 2020*, 2020). It is characterized by memory disorders associated with learning difficulties and mental performance losses which are more pronounced compared to healthy peers. Deficits are also clearly evident in the context of a lower ability to concentrate. Especially in stressful situations, new information is processed more slowly and there is a decrease in abstraction and discrimination. The distinction between MCI and an actual AD is based on a certain severity of cognitive impairment. However, MCI can also be seen as a preliminary stage of different forms of dementia (Burns and Zaudig, 2002; Sarazin and Dubois, 2002; Dubois and Albert, 2004; Petersen, 2004; Hödl, Bonelli and Kapfhammer, 2005; Visser and Verhey, 2008).

Dementia in AD is the most common form of dementia and contributes to 60–70% of the cases. Worldwide, around 50 million people currently suffer from dementia and almost 10 million cases are newly diagnosed every year. Dementia is a syndrome that mostly develops from a chronic or progressive brain disease and is encoded with the ICD-10-GM-Code F00–F03 (*DIMDI - ICD-10-GM Version 2020*, 2020). It is further differentiated into dementia of the Alzheimer type, vascular dementia, dementia in other diseases classified elsewhere (e.g. dementia in primary Parkinson’s syndrome), and unspecified dementia. Although dementia mainly affects seniors, it is not a normal part of aging. Dementia is defined as a disorder that affects various cortical functions, such as memory, thinking, behavior, orientation, perception, learning ability, language, judgment, and the executive. The cognitive impairment leads to noticeable and disruptive changes of professional, social, and private everyday life of those affected but

also has an impact on society as a whole (Hayden and Welsh-Bohmer, 2011; WHO, 2019).

AD was first described clinically and histopathologically by Alois Alzheimer in 1906 (Alzheimer, 1906) and is now one of the most common primary neurodegenerative diseases. Clinically, there is a progressive breakdown of higher cognitive functions. AD changes brain structures long before the onset of clinical symptoms. Macroscopically, pronounced brain atrophy is characteristic in areas of the brain associated with long-term memory, especially in the hippocampal formations, the temporo-parietal, entorhinal, and frontal cortex. Histopathologically, there are extracellular senile plaques from β-amyloid deposits and intracellular so-called neurofibrillary tangles from tau protein deposits. Only the postmortem detection of these neuropathological changes provides the final diagnosis of dementia of the Alzheimer type (Horn *et al.*, 1996; Zaudig, 1996; A. Delacourte *et al.*, 1999; Kretzschmar and Neumann, 2000; Lautenbacher and Gauggel, 2010). The preliminary AD diagnosis is often based on neuropathological tests that examine the cognitive impairment. However, the symptoms of Alzheimer's patients develop slowly and only worsen over time. The fully developed symptoms in Alzheimer's patients are often preceded by a significantly milder symptomatic phase. Patients in this mild symptomatic phase run the risk of being diagnosed later – with the development of full symptoms. However, early diagnosis is important for the success of treatment. Therefore, the diagnosis of MCI is particularly useful as it indicates a deviation from normal aging and helps to identify those patients who are at risk of developing dementia in the future. MCI is also often used in clinical research and large epidemiological studies to assess risk factors for the later development of dementia (Dubois and Albert, 2004).

1.3 Objectives

The goal of this study was to investigate associations between long-term exposure to air pollution, reduced odor identification, and cognition (Figure 1).

Air pollution exposure has been associated with MCI which is a transitional state in-between normal aging and neurodegenerative diseases. Neurodegenerative diseases often occur in combination with olfactory loss. Our findings refer to the effects of long-term air pollution exposure on the olfactory system. We investigated whether there was evidence of an association between reduced odor identification and cognitive impairment. We also examined the role of odor identification regarding the association between air pollution and cognitive impairment. This association could be important, especially with respect to olfactory loss as an early symptom in both, Alzheimer's and Parkinson's patients.

The present study used data of the SALIA (Study on the influence of Air pollution on Lung function, Inflammation, and Aging) cohort of the follow-up investigations in 2008/2009 and 2012/2013. First, we analyzed a possible association between long-term exposure to air pollution and reduced odor identification. Second, we investigated the relationship between reduced odor identification and cognitive impairment. Third, we examined the direct association between air pollution exposure and cognition. We performed a mediation analysis to investigate the role of odor identification as a mediator on the pathway between air pollution exposure and cognitive impairment. Subsequently, we conducted an interaction analysis to investigate if the association between air pollution exposure and cognitive impairment changes depending on odor identification as a moderator.

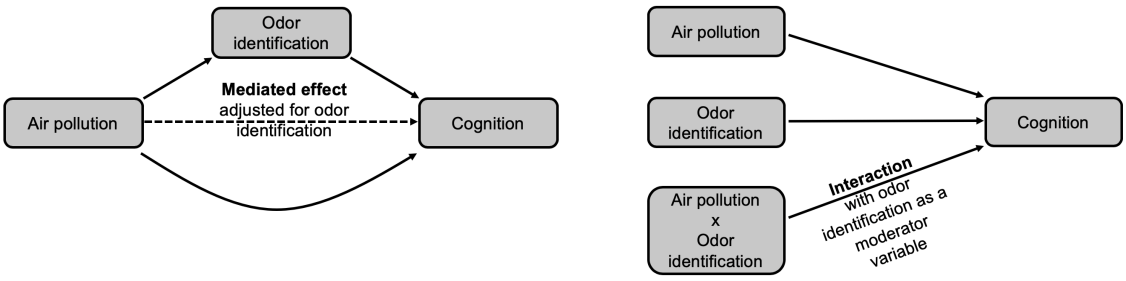


Figure 1: Associations between air pollution, odor identification, and cognition in the mediation (left) and interaction (right) analysis.

2 Materials and methods

2.1 Study population

The SALIA cohort study was initiated as part of the Environmental Health Survey which was an element of the “Clean Air Plan” introduced by the State Government of North Rhine-Westphalia (NRW) in the early 1980s (Schikowski *et al.*, 2005). The cohort was drawn from a random population of women in their mid-50s living in the study area. Members of the IUF (Leibniz-Research Institute for Environmental Medicine, German: Leibniz-Institut für Umweltmedizinische Forschung) performed the baseline investigations consecutively between 1985 and 1994 to examine the health effects of air pollution exposure. The study areas included non-industrialized rural regions north of the Ruhr area (Borken, Dülmen) as reference areas with lower pollution levels and industrialized regions (Dortmund, Duisburg, Essen, Gelsenkirchen, Herne) to represent different levels of exposure to ambient air pollution from high traffic load, steel and coal industries (Figure 2).

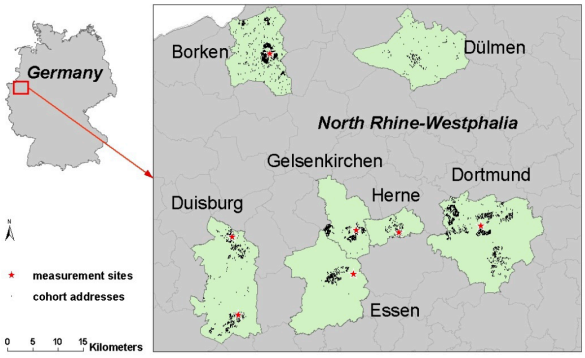


Figure 2: Map of study areas consisting of the regions of Borken, Dülmen, Dortmund, Duisburg, Essen, Gelsenkirchen, and Herne in North Rhine-Westphalia, Germany (by courtesy of IUF).

The study design was restricted to women only as the main focus was on the effects of exposure to ambient air pollutants. At the time of the baseline investigation, most women were housewives. Therefore, it was assumed that women spent most of their time at home as a result and were primarily exposed to air pollution near their home addresses. In contrast, men were excluded in order to avoid bias caused by high occupational exposure to air pollution from

working in the mining and steel industries, since these were predominant sources of income for men in the study area prior to the baseline investigation.

All women aged 54 to 55 years who were living in the predefined area were asked to participate in the study between 1985 and 1994. About seventy percent (N = 4874) responded positively. The baseline investigation consisted of a questionnaire on symptoms, risk factors, and already diagnosed respiratory diseases. The analysis was limited to 4757 women whose addresses were available and could be merged with geographic coordinates. Due to capacity reasons, only a subgroup, about every second responder (N = 2593), was additionally invited for lung function measurements.

Between 2002 and 2003, a mortality follow-up showed that 595 (approximately 12%) of the former study participants had died. The causes of death were obtained from official death certificates and coded according to the International Classification of Diseases (ICD). In addition, 252 women had moved during the follow-up period. These were lost for the follow-up after moving and were classified as censored at the time of movement (Schikowski *et al.*, 2005; Gehring *et al.*, 2006).

After a strong decline in concentrations of ambient air pollutants in the Ruhr region, it was decided to perform a follow-up questionnaire to investigate changes in pulmonary symptoms and general health in 2006. Due to organizational restrictions in local health departments, women from Dülmen and Herne did not participate in this questionnaire. Out of 4027 surviving participants with available addresses, 2116 answered a self-administered postal version of the baseline questionnaire and a total of 1639 women agreed to participate in further clinical examinations.

A follow-up investigation was performed in 2008/2009. All surviving women from this group who had participated in the baseline investigation and who had lung function measurements at the time of the baseline investigation were invited in a randomized manner. In total, 834 women underwent a clinical follow-up examination and further tests at local study centers (Vossoughi *et al.*, 2014; Schikowski *et al.*, 2015).

In 2012/2013, 624 women were asked to take part in a second follow-up investigation. Of these, 542 participated in further clinical examinations as well as a second cognitive assessment. A flow-chart of the study population is shown in Figure 3.

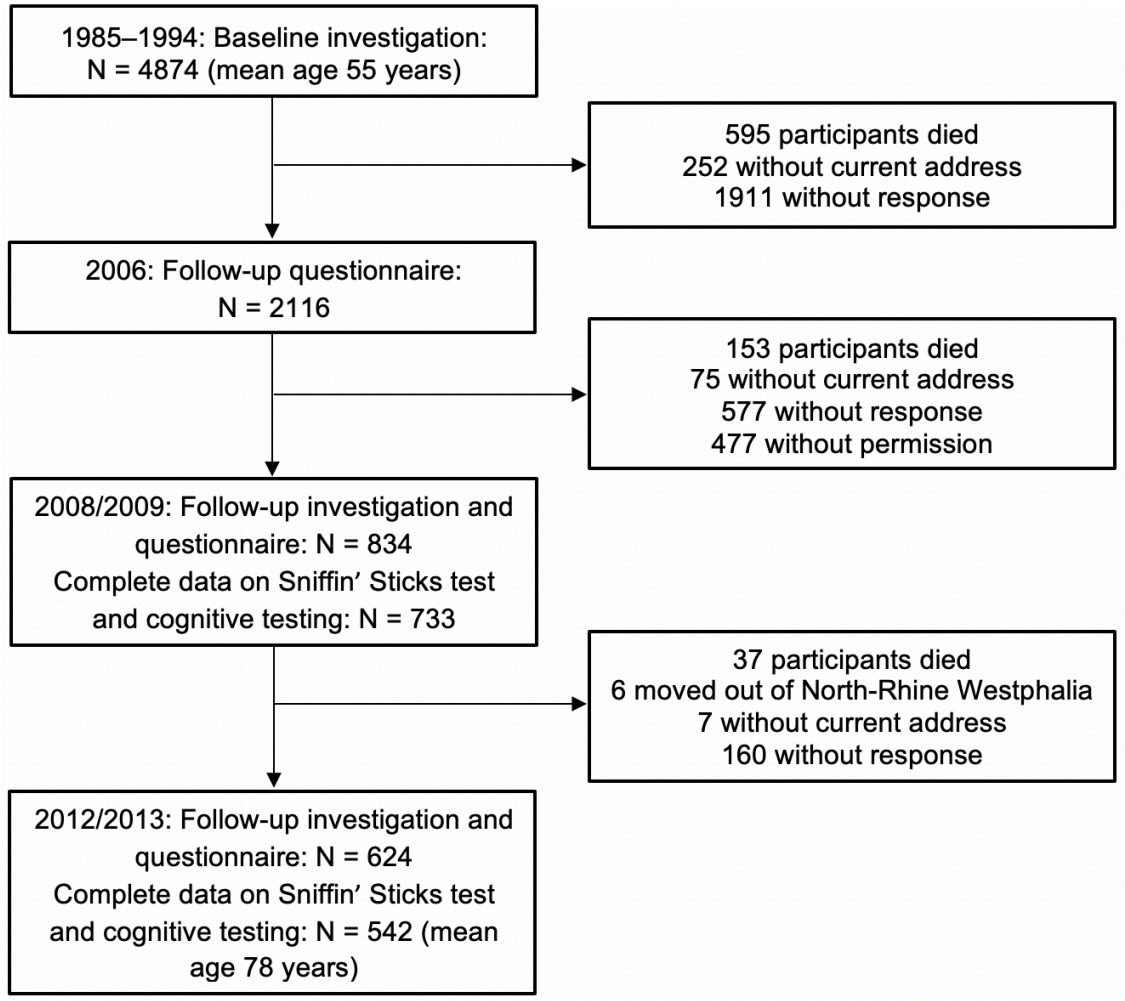


Figure 3: Chronological composition of study population
from 1985 (N = 4874) to 2013 (N = 624).

For the present study, the inclusion criteria were defined as complete data of the Sniffin' Sticks test and the assessment of cognition for the follow-up investigations in 2008/2009 and 2012/2013. These were met by 733 women in 2008/2009 and 542 women in 2012/2013.

All study participants provided written, informed consent. Ethical approval was obtained from the Ethical Committee of the Heinrich-Heine-Universität Düsseldorf (study number: 3507).

2.2 Assessment of air pollution exposure

Air pollution exposure was defined as the women's exposure to NO_2, PM_{10}, $PM_{2.5}$, and filter absorbance of PM with an aerodynamic diameter of 2.5 µm or less ($PM_{2.5}$abs) in our study. Air pollution exposure was assessed in detail and assigned to the address coordinates of study participants. The concentrations of air pollutants were estimated using the Geographic Information System (GIS) technique.

Initially, concentrations of ambient air pollutants were measured at stationary monitoring stations throughout the Ruhr area. These monitoring stations were maintained by the NRW State Environment Agency and covered the Ruhr area in an 8 km grid. Thus, all monitoring stations were located within a small distance of the participant's home address. They were mainly designed to reflect urban to regional scale spatial variations in air quality. TSP were gathered at the monitoring stations with a low volume sampler (air flow: $1 m^3/h$) and then measured using β-ray absorption. At this time, PM_{10} was not routinely measured at all monitoring stations. Therefore, the exposure of PM_{10} was calculated by multiplying TSP measurements with a conversion factor which was estimated at other monitoring stations in the Ruhr area where parallel measurements of TSP and PM_{10} were carried out. The concentrations of NO_2 were measured half-hourly by means of chemiluminescence. The long-term exposure to ambient air pollution was defined as 5-year means of PM_{10} and NO_2 of the years 2003–2007. For this purpose, the individual exposure to ambient air pollution was estimated using the monitoring station closest to the participant's home address (Schikowski *et al.*, 2005, 2008; Gehring *et al.*, 2006).

For a more accurate assessment of the individual long-term exposure, the air pollution concentrations at the participant's home address were estimated with the help of the ESCAPE study (European Study of Cohorts for Air Pollution Effects). Land use regression (LUR) models and data of a measurement campaign (2008–2011) gained in the framework of the ESCAPE study were used. More details of standardized ESCAPE protocols and methods for the development of exposure models are given elsewhere (Eeftens *et al.*, 2012; Beelen *et al.*, 2013).

LUR models provide information about the spatial distributions of the annual mean concentrations which are used as an approximation for the long-term averages of all exposure markers. These validated models were used to assign exposure estimates to the study participants' home address. From 2008 to 2011, campaigns to monitor air pollution were carried out in the study area. Changes in air pollutant levels during the seasons were assessed by 14-day measurements during the cold, warm, and intermediate season and adjusted with data of a reference station that measured over the entire period.

The air pollutant concentrations of PM_{10}, $PM_{2.5}$, and $PM_{2.5}$abs were measured at 20 stations in the Ruhr area. PM_{10} and $PM_{2.5}$ were collected on pre-weighed Teflon filters. $PM_{2.5}$abs was measured as the blackness of $PM_{2.5}$ filters and transformed into absorbance. This variable is often used as a proxy for elemental carbon which is the dominant light-absorbing substance when it comes to air pollution. Simultaneous measurements of NO_2 were performed using passive sampling devices at 40 representative distributed monitoring stations.

In order to better depict the spatial variation of the annual mean concentrations and to obtain a regression equation, measured concentrations were associated with the characteristics of land use. This included data on local transport, road networks, land use, ports, industry, as well as population and household density which were edited and evaluated by the GIS technique. The potential geographic predictors were identified and assigned to each monitoring station. In order to include these spatial differences, LUR models were developed for each area. The regression models obtained were then used to estimate annual pollutant concentrations at the home address of each study participant.

In this way, individual estimates of air pollutant concentrations in the residential ambient air at time of the baseline and the follow-up investigations could be derived. These estimated concentrations of PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2 were assigned to each study participant's home address to receive individual exposure values (Teichert *et al.*, 2013; Vossoughi *et al.*, 2014; Perez *et al.*, 2015; Schikowski *et al.*, 2015).

2.3 Assessment of odor identification

The odor identification was tested with Sniffin' Sticks (Burghart Messtechnik GmbH, 2006) for the follow-up investigation in 2008/2009. Initially, participants answered a questionnaire including the following questions:

> "1. How good do you think you can smell? 1.1 very bad, 1.2 rather bad, 1.3 normal, 1.4 good, 1.5 very good."
>
> "2. How good do you breathe through your nose? 2.1 very bad, 2.2 rather bad, 2.3 normal, 2.4 good, 2.5 very good."
>
> "3. Is smell important to you? 3.1 very important, 3.2 important, 3.3 normal, 3.4 rather not important, 3.5 not important."
>
> "4. Do you feel that you can: 4.1 smell very well, 4.2 cannot smell everything, 4.3 sometimes cannot smell something that other people can smell, 4.4 smell very bad?"
>
> "5. Do you currently have a cold? 5.1 yes, 5.2 no."
>
> "6. Do you currently have hay fever? 6.1 yes, 6.2 no."
>
> "7. Which of the following products did you use today? 7.1 perfume, 7.2 deodorant, 7.3 hairspray."

It was also ensured that the participants had not drunk anything other than water and that they had neither eaten anything, nor chewed gum, nor smoked 15 minutes before the test. In addition, the measurements were carried out in a quiet, well-ventilated room.

The test itself consisted of 16 felt-tipped pens which were presented to the participants one at a time in intervals of 20 to 30 seconds. The Sniffin' Sticks looked like commercially available pens with a length of 14 cm and an inner diameter of 1.3 cm. The pens were filled with different liquid odors dissolved in propylene glycol and stored and used according to the manufacturer's instructions. The test leaders followed the study protocol described below.

For odor presentation, the cap was removed by the test leader, being careful not to touch tip of the Sniffin' Stick. It was placed at a distance of approximately 2 cm

in the middle under both nostrils. The Sniffin' Stick was not allowed to touch the participants' skin and neither was to move it around. The participants were then asked to smell or inhale. After 2–3 seconds, the cap was closed and the participants were given lists of 4 descriptors and asked to identify the odor using a forced-choice protocol. The lists of 4 descriptors were as follows (*correct odor in italics*): (1) *orange*, strawberry, blackberry, pineapple; (2) smoke, *leather*, adhesive, grass; (3) honey, chocolate, vanilla, *cinnamon*; (4) chives, spruce, *peppermint*, onion; (5) coconut, walnut, *banana*, cherry; (6) peach, apple, *lemon*, grapefruit; (7) *licorice*, gummy bear, chewing gum, cookie; (8) cigarette, *coffee*, wine, candle smoke; (9) *clove*, pepper, cinnamon, mustard; (10) pear, plum, peach, *pineapple*; (11) chamomile, raspberry, *rose*, cherry; (12) bread, *fish*, cheese, ham; (13) mustard, rubber, menthol, *turpentine*; (14) onion, sauerkraut, *garlic*, carrot; (15) *anise*, rum, honey, spruce; (16) melon, peach, orange, *apple*. The result was noted without indication of the correctness of the answer. Refusals to provide an answer were treated as incorrect. The participants' scores ranged from 0 to 16 correctly identified odors (Hummel *et al.*, 1997; Kobal *et al.*, 2000).

Due to the complexity of odor identification in which different – including neurological – systems are involved, the Sniffin' Sticks test was only carried out after the assessment of cognition.

2.4 Assessment of cognition

Impairment in cognition was examined using the validated neuropsychological CERAD-Plus (Consortium to Establish a Registry for Alzheimer's Disease) test battery (Memory Clinic University Hospital Basel, 2005).

Cognitive impairment develops very slowly which makes the diagnosis of neurodegenerative diseases considerably more difficult. Those who are affected by memory loss, attention and concentration difficulties often do not notice these deficits themselves. By interviewing relatives of participants, their everyday activities were assessed using the B-ADL (Bayer Activities of Daily Living Scale) (Erzigkeit *et al.*, 2001). Initially, a brief eye test was used to determine whether participants were able to perform the tasks assigned to them by the neuropsychological tests. The execution of neuropsychological tests might also be influenced by the current emotional state. Therefore, each participant was tested for depressive symptoms by the questionnaire ADS (General depression scale, German: Allgemeine Depressionsskala) before examining cognition. ADS was designed as a self-assessment and screening tool to evaluate the severity of depressive symptoms and is also used to differentiate depression from MCI. The frequency of typical depression symptoms (affective, cognitive, somatic, and social) in the past week was assessed on the basis of 20 statements. 4 statements were inverse and served as a criterion for the validity of the answers. They were designed to control correct understanding and to reveal false information through response patterns, distortions, and inattentive answers. The evaluation was easily done with paper and pencil in less than 1 minute and the total score was rated on the basis of validated cut-offs (Hautzinger *et al.*, 2012).

The CERAD-Plus test battery only became established in German-speaking countries at the end of the 20th century, but was already developed according to existing methods by the National Institute on Aging in the U.S.A. in 1986. The test battery was originally designed to generate standardized, validated measures for assessing AD, but has since developed into a widely used, age-differentiated screening tool for other types of neurodegenerative diseases as well. It is still primarily used in dementia diagnostics, e.g. for early detection and identification of treatment options, but can also differentiate between normal cognitive abilities and MCI, dementia and neurodegenerative diseases.

The CERAD-Plus test battery consists of 10 different subtests carried out directly by the participants. In order to obtain a complete picture of one's individual cognition, each subtest examines different cognitive abilities. The sum of all CERAD subtest scores was used to examine general cognition. The integrated standardization program of the German version of the CERAD-Plus test battery transforms all subtest scores into normalized z-scores, standardized according to age, level of education (in years), and gender. Z-scores are individually calculated according to the following formula:

$$z = \frac{(transformed)\ raw\ score - prognosis}{SD\ of\ RR}.$$

The prognosis corrects the influence of demographic variables using a reference population, which is described in detail elsewhere (Aebi, 2003; Luck *et al.*, 2009). "SD of RR" represents the standard deviation (SD) of the residuals of the regression model that best reflect the influences of the demographic variables (RR). Z-scores close to 0 indicate performance within the normal range of the CERAD-Plus test battery based on age, level of education, and gender. The mean of these z-scores was used to form a CERAD total score (Berres *et al.*, 2000). General cognition was also assessed by the Mini Mental State Examination (MMSE). The CERAD subtests are briefly specified below, while a more detailed description has been published elsewhere (Morris *et al.*, 1989; Mirra *et al.*, 1991; Welsh *et al.*, 1994; Aebi, 2003; Chandler *et al.*, 2005; Fillenbaum *et al.*, 2008; Karrasch *et al.*, 2013). The neuropsychological tests were carried out by trained interviewers and overviewed by a psychologist. The participants with previously diagnosed neurodegenerative diseases were not included in the analysis.

In the subtest "Semantic fluency", the participants listed as many animals as possible in 1 minute. The number of different animals added up to a total score. The skills in language production, especially speed and lightness, semantic memory, language, executive function, and cognitive flexibility were tested.

In the short version of the "Boston Naming Test" (BNT), the participants were asked to name 15 items which were individually and successively presented over a period of 2–10 seconds as line drawings on DIN-A5 paper. The participants received 1 point for every correct answer which were added up to a total score of

15 points. The original BNT version consists of 60 drawings, whereas for the CERAD-Plus test battery, a short version of 15 drawings is used. The subtest examined visual perception, object recognition, in particular the function of temporal lobes, object localization, in particular the functioning of parietal lobes, word finding and naming, in particular the Wernicke's area.

The participants had a time limit of 1 minute to enumerate words starting with the letter "s" in the subtest "Phonetic fluency". The number of correctly named "s"-words was added to calculate a total score. It examined language production regarding speed and lightness, phonetic memory, lexical memory, general memory, cognitive flexibility, and decision-making.

Study participants were asked to read aloud 10 words from the test booklet in a total of 3 rounds in the subtest "Word list learning". The words were presented one after the other at intervals of 2 seconds and in different order in each round. The participants had a maximum of 90 seconds per round to memorize the words. Subsequently, the participants were asked to express what they memorized. Correctly remembered words were marked in the questionnaire and added up to a total score for all 3 rounds. The number of intrusions, namely words that did not appear in the word list, but were remembered on a false basis, was also noted and differentiated from the words that were guessed. The maximum number of correct answers in the 3 rounds was 30. This subtest acquired the ability to learn unrelated verbal information in a short time.

In the subtest "Word list recall", the participants were asked to freely recall the 10 words learned in the subtest "Word list learning" described above. The words were retrieved a few minutes after being learned. Similar to what is described above, the number of intrusions was noted and additionally differentiated from guessed words. There was also a time limit of 90 seconds for recalling the words and the number of correct answers was noted and calculated for a total score of up to 10 points. It was checked whether the test participants could remember newly learned verbal information over a period of a few minutes. Thus, the delayed verbal episodic memory was tested.

The subtest "Word list recognition discriminability" was also based on the subtest "Word list learning". The previously learned words should be recognized from a list along with 10 distractors. The 20 words were presented to the participants

one by one and in succession. The rating took into account the number of correctly recognized words that were previously presented in the subtest “Word list learning” (correct “yes” responses = hits) and the number of correctly recognized distractors (correct “no” responses = correct rejections). Only “yes” or “no” responses were allowed. The sum of correct responses was then calculated (in %) based on the following formula and with 100% being the highest score:

$$discriminability = \left(1 - \frac{(10 - hits) + (10 - correct\ rejections)}{20}\right) \times 100.$$

This subtest helped to check whether the test participants benefited from easier access conditions and allowed a distinction between a memory disorder with deficits in storing or accessing contents.

In the subtest “Figure copying”, the participants were asked to copy 4 figures of increasing complexity as precisely as possible: 1 circle, 1 rhombus, 2 intersecting rectangles, and 1 cube. 1 point was given for an approximately circular figure, if the circle was closed, and if a gap existed the distance between lines had to be less than 3 mm. The rhombus had to have all 4 sides (1 point). Lines within the drawing had to be about the same length (1 point) and closed, meaning that if a gap existed, the distance between lines had to be less than 3 mm (1 point). For the drawing of the 2 intersecting rectangles, points were assigned if both figures had 4 sides (1 point) and if the drawing looked similar to the original (1 point). A maximum of 4 points were given for the drawing of the cube: 1 point if the figure was 3-dimensional, 1 point if the front was correctly oriented left or right, 1 point if the inner lines were drawn correctly, and 1 point if the opposite sides were parallel (within 10 degrees). The maximum total score was 11 points. VCP, visual-spatial thinking, and instant memory skills were examined in this subtest.

The figures that were drawn in the subtest of VCP (“Figure copying”) had to be recalled in the subtest “Figure recall”. The scores of the redrawn figures were based on the same system explained above for the subtest of VCP and the maximum total score was also 11 points. This subtest examined nonverbal modality and delayed figural memory.

In the “Trail Making Test A”, the participants had to connect numbers in ascending order. Before starting, an exercise was carried out as an example to become familiar with the type of the task. The following instructions were given: “Please

start at 1 and draw a line from 1 to 2, from 2 to 3, from 3 to 4 etc., until you are finished.” Errors were corrected immediately by the interviewer and the test was stopped after 5 minutes if the task was not completed. The time required and the number of errors were noted. In the “Trail Making Test B”, the level of difficulty increased. The participants had to alternate first a number and then a letter with each other in ascending order. Another exercise was performed as an example before the actual task. The participants received the following instructions: “Please draw a line from 1 to A, from A to 2, from 2 to B, from B to 3, from 3 to C etc., until you are finished.” The required time and the number of errors were also noted. For the variable “Trail Making Test B/A”, the z-score of Trail B was divided by the z-score of Trail A. Trail A focused on psychomotor speed, while Trail B reflected the performance of the executive function. Together the subtests examined attention, visuo-construction performance, discrimination, concentration, problem-solving, and decision-making.

The MMSE is a validated screening method for the assessment of the general cognition. In particular, it is designed to examine orientation, concentration, memory, language, and constructive practice. The MMSE consists of 20 questions, such as “Which year is it?” or “Please write any complete sentence on this paper.” A score of 1 was noted for each correct answer and a score of 0 for each incorrect answer. The maximum score was 30 points. The observed skills with regard to orientation are particularly significant, since disorders in this area are often the first symptoms of AD onset. The MMSE is also used to assess the severity of dementia. The sensitivity is satisfactory in the middle range of scores. Moderately severe dementia symptoms can often already be present at scores below 20 points, given that language disorders, lack of language skills, acute stress events, drug side effects, or a low level of education have been ruled out beforehand. In contrast, the procedure is insufficiently sensitive to mild symptoms. Hence, we did not concentrate exclusively on the MMSE, but assumed its equivalence. We applied the same assumptions on the individual CERAD subtests and the CERAD-Plus total score.

2.5 Assessment of covariates

All participants provided information on potential covariates based on a self-administered standardized questionnaire at the time of the baseline (1985–1994) and follow-up investigations (2008/2009 and 2012/2013). Detailed information on known risk factors for reduced odor identification and MCI was collected (Ahlström *et al.*, 1987; Katzman and Kawas, 1994; Gehring *et al.*, 2006; Doty, 2009; Künzli, Perez and Rapp, 2010; Schikowski *et al.*, 2015).

First, basic questions regarding age and education were asked. Educational level was defined as the maximum period of education achieved either by the participating women or their husbands. Although z-scores in the CERAD-Plus test battery are already standardized for educational level, this variable was particularly relevant for evaluating cognition. Questions were asked about the number of school years, and the number of years for vocational training or for achieving a university degree. Standards were defined according to the school attended: 8 years of education were noted for attending elementary school, 10 years for middle school and 13 years for high school. It was irrelevant whether someone had repeated or skipped a school year. Similarly, standards were defined for the number of years for vocational training or for achieving a university degree: 2–3 years of education were noted for vocational training, 9 semesters for a university degree and 7 semesters for a degree at a university of applied sciences. Furthermore, it was noted which professions the participants had learned. If participants had completed 2 or more vocational trainings, only the highest degree was relevant. The years of education were subsequently calculated. Accordingly, participants were put into 3 levels of education: low (<10 years), medium (=10 years), and high (>10 years).

Body-Mass-Index (BMI), as a measure for assessing the weight of a person in relation to body size, was calculated.

Potential risk factors for respiratory diseases were assessed, such as smoking status (current, past, and never smoker), the number of pack-years for ever smokers, and environmental tobacco exposure at home as definition for passive smoking. In addition, the questionnaire included questions about current symptoms or diseases.

We also gathered information regarding chronic diseases, such as diabetes, bronchial asthma, chronic bronchitis, hay fever, COPD, hypertension, and medication, including possibly sedative drugs. Hay fever and cold were also inquired as possible covariates. All returned questionnaires were checked by the examining doctor.

Finally, the housing situation of the women was assessed and assigned to the categories urban and rural living.

2.6 Statistical analysis

For this statistical analysis, the study population included only women who participated in the Sniffin' Sticks test and the CERAD-Plus test battery for the follow-up investigation in 2008/2009 (N = 733) and with complete information for the assessment of odor identification and cognition in 2012/2013 (N = 542) (Figure 3).

The study population was described separately according to the time of the follow-up investigations in 2008/2009 and 2012/2013. In the description, the mean and the associated standard deviation (SD) for continuous variables (e.g. "age") and the number in percent for dichotomous "yes" or "no" variables (e.g. "cold") are presented. The description of air pollution exposure, odor identification, and cognition were also described. Air pollution exposure was analyzed using the median and associated interquartile ranges (IQR) with unit steps for each pollutant. The Sniffin' Sticks total number was calculated from the number of correctly identified odors from a total of 16 Sniffin' Sticks presented (in %) which was also used as a variable for further analyses. Z-scores were used as outcomes for the assessment of cognition.

With the help of linear regression models, we investigated whether there was a difference in the distribution of odor identification associated with air pollution exposure in a cross-sectional analysis for the follow-up investigation in 2008/2009. Multiple linear regression models were estimated for the association between reduced odor identification and cognitive impairment at 2-time points (2008/2009 and 2012/2013). We conducted a cross-sectional analysis for the association between long-term air pollution exposure and cognitive impairment for the follow-up investigations in 2008/2009 and 2012/2013.

On a hypothesized pathway from air pollution exposure to cognitive impairment, the olfactory system might be of importance. We performed a mediation analysis on the association between air pollution and cognition using additional adjustment for odor identification (Figure 4). We analyzed whether odor identification mediated the effect of air pollution exposure on cognitive impairment in a cross-sectional mediation analysis in 2008/2009 and 2012/2013.

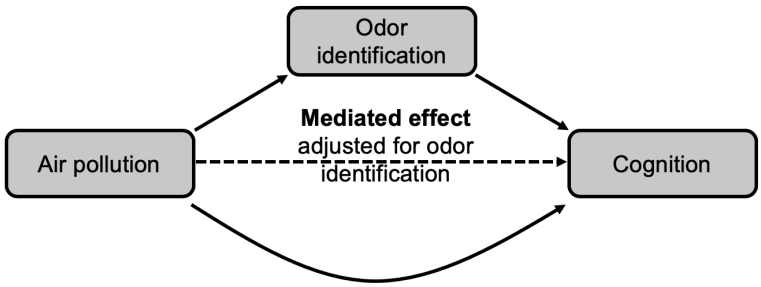


Figure 4: Mediation analysis: associations between air pollution, odor identification, and cognition.

The analyses were performed by using the R (R Development Core Team, 2013) package mediation (Tingley *et al.*, 2014) on the basis of a normal approximation. The most commonly used method for testing mediation investigates the association between the exposure (air pollution) and the mediator variable (odor identification), the association between the mediator and the outcome variable (cognition), and examines whether the direct effect of exposure on outcome was changed through odor identification as a mediator variable. Justification for performing the mediation analysis was given by Judd and Kenny (1981). In general, a variable functions as a mediator to the extent that it accounts for the association between exposure and outcome. We carried out the analysis of causal mediation from a counterfactual perspective with possible interactions between exposure and mediator. An approach that relied on the quasi-Bayesian Monte Carlo method also based on normal approximation (Imai, Keele and Tingley, 2010). Direct and indirect effects were defined, as well as a total effect as the sum of direct and indirect effects. The counterfactual framework allowed for these definitions to work even in models with interactions. In this case, the indirect effect referred to the effect caused by the mediator examined. Meanwhile, the direct effect related to the remaining effect that was not mediated by the mediator (Ikram and VanderWeele, 2015). The proportion of the indirect effect in the total effect was used to assess the extent to which odor identification mediated the association between air pollution exposure and cognitive impairment for the follow-up investigation in 2008/2009 (Baron and Kenny, 1986; MacKinnon, Krull and Lockwood, 2000; Liu, Jones and Glymour, 2010; McFall *et al.*, 2010; Valeri and VanderWeele, 2013; VanderWeele, 2013).

Additionally, we carried out an interaction analysis with odor identification as a moderator variable (Figure 5). We investigated whether the effect of air pollution exposure on cognitive impairment (2008/2009 and 2012/2013) changed depending on how well the test participants performed in the odor identification test.

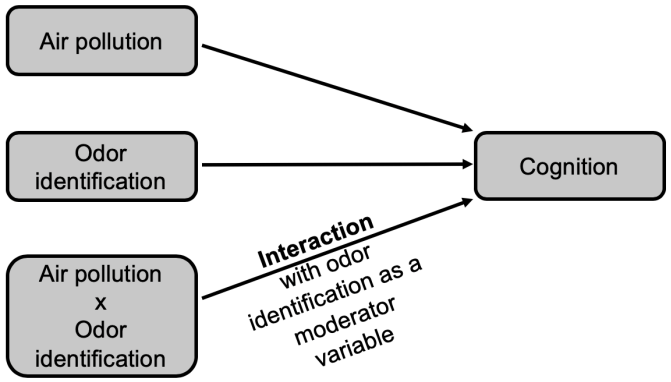


Figure 5: Interaction analysis: associations between air pollution, odor identification, and cognition.

For the interaction analysis, we first evaluated the associations between the variables: exposure (air pollution), moderator (odor identification), and outcome (cognition). The outcome variable of cognition was associated with 3 causal pathways: the impact of air pollution as exposure, the impact of odor identification as moderator, and the interaction between exposure and moderator. According to Baron and Kenny (1986), the moderator hypothesis should be supported if the interaction was significant. Testing a moderator variable would be irrespective of whether significant main effects also occurred for exposure and moderator, since these were not directly relevant conceptually. Thus, the interaction analysis tested whether the causal relationship between 2 variables changed depending on a moderator variable. In contrast to the association between exposure and mediator in the mediation analysis (exposure occurs causally before the mediator), moderator and exposure are at the same level in terms of their role as antecedent or exogenous causal variables for certain criteria effects in the interaction analysis. As a result, the moderator variable acts as an independent variable, while mediated associations change the roles from effects to causes depending on the focus of the mediation analysis (Aiken and West, 1986;

MacKinnon, Krull and Lockwood, 2000; Kraemer *et al.*, 2001, 2002, 2008; McFall *et al.*, 2010; Valeri and VanderWeele, 2013).

Previous studies of the SALIA cohort investigated whether cognitive impairment was associated with air pollution exposure (Schikowski *et al.*, 2015). Results indicated an assiociation between long-term air pollution exposure and the cognitive performance of some CERAD subtests. In particular, there was an association between the air pollutants PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2 and the CERAD subtests of semantic memory (“Boston Naming Test”) and visual construction (“Figure copying”). Therefore, our analysis of the association between air pollution and cognitive impairment looked at the general performance of the CERAD-Plus test battery, with a special focus on the results of the subtests mentioned above.

Both, crude associations (only adjusted for age) and associations additionally adjusted for other potential covariates, were estimated. Our analysis controlled for a large set of covariates. The main model was adjusted for age, level of education, BMI, smoking status, environmental tobacco exposure at home, cold, hay fever, as well as urban and rural living. In addition to the main model, an extended model was also estimated in a sensitivity analysis. The extended model was additionally adjusted for diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension.

Presented are the β-estimate, associated 95%-confidence intervals (95%-CI), and p-values. A 95%-CI indicates the range in which the true mean lies with a 95% probability. The statistical analysis was carried out using the statistical software R version 3.1.3 for Windows.

3 Results

3.1 Characteristics of study population, air pollution, and outcome

3.1.1 Description of the study population

The study population consisted of 733 participants with complete information on the Sniffin' Sticks test and the assessment of cognition for the follow-up investigation in 2008/2009 and 542 participants with a second cognitive assessment in 2012/2013. Table 1 presents the population characteristics of this part of the SALIA cohort for the follow-up investigations in A) 2008/2009 and B) 2012/2013.

For the follow-up investigation in 2008/2009, the mean age of participating women was 73.6 years. According to our definition, 81.4% of the women or their partners had an educational level of at least 10 years of schooling. The majority (N = 596) of the group reported to be never smokers, but 60% had been exposed to environmental tobacco smoke at home. At that time, about 10% of the women reported having a cold or suffering from hay fever. The sample size of the groups representing non-industrialized and industrialized regions were comparable. In order to illustrate how the descriptive data of the study population changed over time, the values for the follow-up investigation in 2012/2013 are also presented in Table 1. For the follow-up investigation in 2012/2013, the mean age was 77.5 years. The groups with different levels of education and the smoking status of the participating women remained stable. In 2012/2013, 13% of the women reported having a cold and 10% suffered from hay fever. Finally, the sample size of the groups representing non-industrialized and industrialized regions also remained comparable in each group at around 50%.

Table 1: Characteristics of the study population (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).

	Mean (± SD[a]) or number (%)	
	A) Follow-up investigation in 2008/2009.	**B) Follow-up investigation in 2012/2013.**
Sample size	733	542
Age [years]	73.6 (3.0)	77.5 (3.2)
Educational level:[b]		
Low [<10 years]	133 (18.1%)	102 (18.8%)
Medium [=10 years]	352 (48.0%)	251 (46.3%)
High [>10 years][c]	245 (33.4%)	189 (34.9%)
BMI [kg/m^2][d]	27.32 (4.5)	28.45 (4.5)
Smoking status:		
Current smoker	19 (2.6%)	17 (3.1%)
Past smoker	118 (16.1%)	85 (15.7%)
Never smoker[c]	596 (81.3%)	437 (80.6%)
Number of pack years for ever smokers[e]	3.4 (10.9)	18.7 (20.3)
Environmental tobacco exposure at home	441 (60.2%)	335 (61.8%)
Diabetes[f]	88 (12.0%)	73 (13.5%)
Bronchial asthma[f]	61 (8.3%)	39 (7.2%)
Chronic bronchitis[f]	90 (12.3%)	51 (9.4%)
COPD[g]	0.8 (0.1)	0.7 (0.1)
Hypertension[h]	482 (65.8%)	362 (66.8%)
Hay fever[f]	78 (10.6%)	50 (9.2%)
Cold[f]	60 (8.2%)	71 (13.1%)
Urban living	392 (53.5%)	270 (49.8%)
Rural living	341 (46.5%)	272 (50.2%)

[a]: SD = standard deviation. [b]: Educational level = highest number of school years attended by either the women or her husband. [c]: information missing on a limited number of subjects. [d]: BMI = body mass index = body weight (kg) / body height^2 (m^2). [e]: Number of pack years for ever-smokers = number of cigarettes smoked per day x number of years the person has smoked / 20 (the size of 1 pack is 20 cigarettes). [f]: Diagnosed by a physician as asked by standardized interview. [g]: COPD = chronic obstructive pulmonary disease: Tiffeneau-Index = forced expiratory volume in 1 second (FEV_1) / forced vital capacity (FVC). [h]: Hypertension = blood pressure >140/90 mmHg. Diagnosed by a physician as asked by standardized interview.

3.1.2 Description of air pollution exposure

The characteristics of air pollution variables at the time of the follow-up investigation in 2008/2009 are presented in Table 2. Exposure to air pollution was assessed using the median and associated interquartile ranges (IQR) with unit steps for each pollutant.

For the follow-up investigation in 2008/2009, the participating women showed the highest exposure to air pollution with the median level of 26.4 µg/m^3 for PM_{10} (IQR = 2.2) with a minimum level of 23.9 µg/m^3 and a maximum level of 33.9 µg/m^3. The median level was 17.4 µg/m^3 for $PM_{2.5}$, 1.4 x 10^{-5}/m for $PM_{2.5}$abs, and 26.0 µg/m^3 for NO_2.

Table 2: Characteristics of air pollution exposure (follow-up investigation in 2008/2009).

Exposure	**Sample size**	**Median (IQR[a])**	**Minimum–Maximum**
PM_{10} [µg/m^3]	733	26.4 (2.2)	23.9–33.8
$PM_{2.5}$ [µg/m^3]	733	17.4 (1.9)	15.6–21.9
$PM_{2.5}$abs [10^{-5}/m]	733	1.4 (0.4)	1.0–4.00
NO_2 [µg/m^3]	733	26.0 (9.7)	19.7–70.3

PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide. [a]: IQR = interquartile range.

3.1.3 Description of odor identification

The results of the performance of the Sniffin' Sticks test for the follow-up investigation in 2008/2009 are presented in Table 3. The odor identification was assessed on the basis of the number of correctly identified Sniffin' Sticks and in percent. A total number was calculated from the number of correctly identified odors from a total of 16 Sniffin' Sticks presented. The Sniffin' Sticks total number is shown as the mean with the corresponding SD.

The mean of the total number of correctly identified Sniffin' Sticks out of 16 was 12.2 with a SD of 2.4.

Table 3: Performance of the Sniffin' Sticks test (follow-up investigation in 2008/2009 (N = 733)).

Sniffin' Stick	**Correctly identified number (%)**
Stick 1 – orange	619 (84.5%)
Stick 2 – leather	571 (77.9%)
Stick 3 – cinnamon	454 (61.9%)
Stick 4 – peppermint	676 (92.2%)
Stick 5 – banana	630 (86.0%)
Stick 6 – lemon	387 (52.8%)
Stick 7 – licorice	639 (87.2%)
Stick 8 – coffee	524 (71.5%)
Stick 9 – cloves	644 (87.9%)
Stick 10 – pineapple	442 (60.3%)
Stick 11 – rose	660 (90.0%)
Stick 12 – fish	684 (93.3%)
Stick 13 – turpentine	458 (62.5%)
Stick 14 – garlic	584 (79.7%)
Stick 15 – anise	602 (82.1%)
Stick 16 – apple	338 (46.1%)
Total number, mean (± SD[a])[b]	12.2 (2.4)

[a]: SD = standard deviation. [b]: Sniffin' Sticks Total number = total number of correctly identified Sniffin' Sticks is also the variable used for further analyses.

3.1.4 Description of cognition

The description of the performance of the CERAD-Plus test battery for the follow-up investigations in A) 2008/2009 and B) 2012/2013 is shown in Table 4. The assessment of cognition is presented as the mean of normalized z-scores for each subtest with corresponding SD.

In 2008/2009, the arithmetic mean of the CERAD total score (sum of z-scores of the CERAD subtests) valued slightly below 0 (mean (± SD) = −0.2 (0.5)). The arithmetic mean of the CERAD total score was <−0.1 with a SD of 0.6 for the follow-up investigation 2012/2013.

Table 4: Characteristics of z-scores of the CERAD-Plus test battery (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).

CERAD-Plus test battery	**Mean (± SD[a])**	
	A) Follow-up investigation in 2008/2009.	**B) Follow-up investigation in 2012/2013.**
Sample size	733	542
Semantic fluency (SeFl)	−0.1 (0.9)	0.1 (1.0)
Boston Naming Test (BNT)	0.6 (1.1)	0.4 (1.2)
Phonetic fluency (PhFl)	0.5 (1.1)	0.3 (1.0)
Word list learning (WL-L)	−0.8 (1.1)	<−0.1 (1.2)
Word list recall (WL-R)	−0.2 (1.1)	−0.1 (1.1)
Word list recognition discriminability (WL-D)	<0.1 (1.1)	−0.1 (1.1)
Figure copying (Fig-C)	−0.9 (1.3)	−0.3 (1.3)
Figure recall (Fig-R)	−0.6 (1.1)	−0.3 (1.1)
Trail Making Test A (TMT-A)	−0.2 (1.0)	0.1 (1.1)
Trail Making Test B (TMT-B)	0.3 (1.0)	−0.1 (1.1)
Trail Making Test B/A (TMT-B/A)	0.6 (1.0)	−0.1 (1.1)
Mini Mental State Examination (MMSE)	−0.7 (1.2)	−0.5 (1.2)
CERAD total score[b]	−0.2 (0.5)	<−0.1 (0.6)

[a]: SD = standard deviation. [b]: CERAD total score = the mean of the sum of z-scores of all CERAD subtests.

It is very unusual for cognitive function to improve over time. A possible explanation for the improvement of the performance of the CERAD-Plus test might be a learning effect together with a healthy survivor bias between the 2-time points in 2008/2009 and 2012/2013. Therefore, we additionally checked the results of the 542 participants who participated in both follow-up investigations (Table 5).

These 542 participants scored −0.1 in the CERAD total score for the first follow-up investigation (2008/2009). Thus, this subgroup had already performed better compared to the mean CERAD total score of −0.2 of the whole study population (N = 733) in 2008/2009. The t-test is presented to assess whether the mean of the performance of the CERAD-Plus test at the first investigation differs from the performance of the CERAD-Plus test for the second follow-up investigation.

Table 5: Distribution of z-scores of the CERAD-Plus test battery of **participants of both follow-up investigations** (N = 542).

CERAD-Plus test battery	**Mean (± SD[a])**		**t-test[c]**
	First follow-up investigation.	**Second follow-up investigation.**	
Semantic fluency (SeFl)	<0.1 (0.9)	0.1 (1.0)	0.357
Boston Naming Test (BNT)	0.7 (1.1)	0.4 (1.2)	<0.001
Phonetic fluency (PhFl)	0.6 (1.1)	0.3 (1.0)	<0.001
Word list learning (WL-L)	−0.7 (1.2)	<−0.1 (1.2)	<0.001
Word list recall (WL-R)	−0.1 (1.1)	−0.1 (1.1)	0.298
Word list recognition discriminability (WL-D)	<0.1 (1.0)	−0.1 (1.1)	0.014
Figure copying (Fig-C)	−0.8 (1.3)	−0.3 (1.3)	<0.001
Figure recall (Fig-R)	−0.5 (1.1)	−0.3 (1.1)	0.005
Trail Making Test A (TMT-A)	−0.1 (1.0)	0.1 (1.1)	0.003
Trail Making Test B (TMT-B)	0.4 (1.0)	−0.1 (1.1)	<0.001
Trail Making Test B/A (TMT-B/A)	0.6 (1.0)	−0.1 (1.1)	<0.001
Mini Mental State Examination (MMSE)	−0.6 (1.1)	−0.5 (1.2)	0.045
CERAD total score[b]	−0.10 (0.5)	<−0.1 (0.57)	0.025

[a]: SD = standard deviation. [b]: CERAD total score = the mean of the sum of z-scores of all CERAD subtests. [c]: T-test = parametric test to determine the difference of means between the performance of the CERAD-Plus test for the first and second follow-up investigation.

3.2 Association between air pollution and odor identification

Table 6 presents the association between air pollution and odor identification for the follow-up investigation in 2008/2009. The β-estimate, associated 95%-CI, and the p-value are presented for PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2. All associations were adjusted for potential covariates.

The analysis revealed no association between long-term air pollution exposure and odor identification. An increase of 1 IQR of PM was not associated with a poorer performance of the Sniffin' Sticks test in either the crude model adjusted for age (e.g. PM_{10}: p-value = 0.515, β = 0.06 (95%-CI −0.12; 0.25)) or the main model adjusted for age, educational level, BMI, smoking status, living in an urban vs. rural area, cold, and hay fever (e.g. PM_{10}: p-value = 0.731, β = 0.04 (95%-CI −0.19; 0.28)). The associations between $PM_{2.5}$, $PM_{2.5}$abs, and NO_2 and the total number of correctly identified Sniffin' Sticks were neither significant.

Table 6: Association between air pollution and odor identification (follow-up investigation in 2008/2009 (N = 733)).

	Crude			Adjusted		
Exposure	**β-estimate**	**95%-CI**	**p-value**	**β-estimate**	**95%-CI**	**p-value**
PM_{10} [µg/m^3]	0.06	−0.12; 0.25	0.515	0.04	−0.19; 0.28	0.731
$PM_{2.5}$ [µg/m^3]	<0.01	−0.24; 0.24	0.995	−0.06	−0.39; 0.26	0.707
$PM_{2.5}$abs [10^{-5}/m]	<0.01	−0.16; 0.16	0.972	0.01	−0.19; 0.22	0.914
NO_2 [µg/m^3]	−0.02	−0.24; 0.20	0.849	−0.06	−0.36; 0.25	0.706

Presented are β-estimates, 95%-confidence intervals (95%-CI), and p-values for an increase by 1 interquartile range (IQR) in air pollution. The crude associations were adjusted for age only. Additional adjustments were made for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide.

We also conducted a linear regression analysis for the association between air pollution and the total number of correctly identified Sniffin' Sticks additionally adjusted for more covariates (namely diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension) for the follow-up investigation in 2008/2009 (Table 7).

In the extended model, an increase of 1 IQR of PM_{10} was neither associated with a poorer performance of the Sniffin' Sticks test (e.g. PM_{10}: p-value = 0.979, β <0.01 (95%-CI −0.25; 0.26)), nor were the associations significant between $PM_{2.5}$, $PM_{2.5}$abs, and NO_2, and the total number of correctly identified Sniffin' Sticks.

Table 7: Association between air pollution and odor identification **additionally adjusted for diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension** (follow-up investigation in 2008/2009 (N = 733)).

Exposure	**β-estimate**	**95%-CI**	**p-value**
PM_{10} [µg/m^3]	<0.01	−0.25; 0.26	0.979
$PM_{2.5}$ [µg/m^3]	−0.10	−0.45; 0.25	0.587
$PM_{2.5}$abs [10^{-5}/m]	0.02	−0.21; 0.24	0.886
NO_2 [µg/m^3]	−0.06	−0.4; 0.28	0.719

Presented are β-estimates, 95%-confidence intervals (95%-CI), and p-values for an increase by 1 interquartile range (IQR) in air pollution. All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, hay fever and **additionally for diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension**. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide.

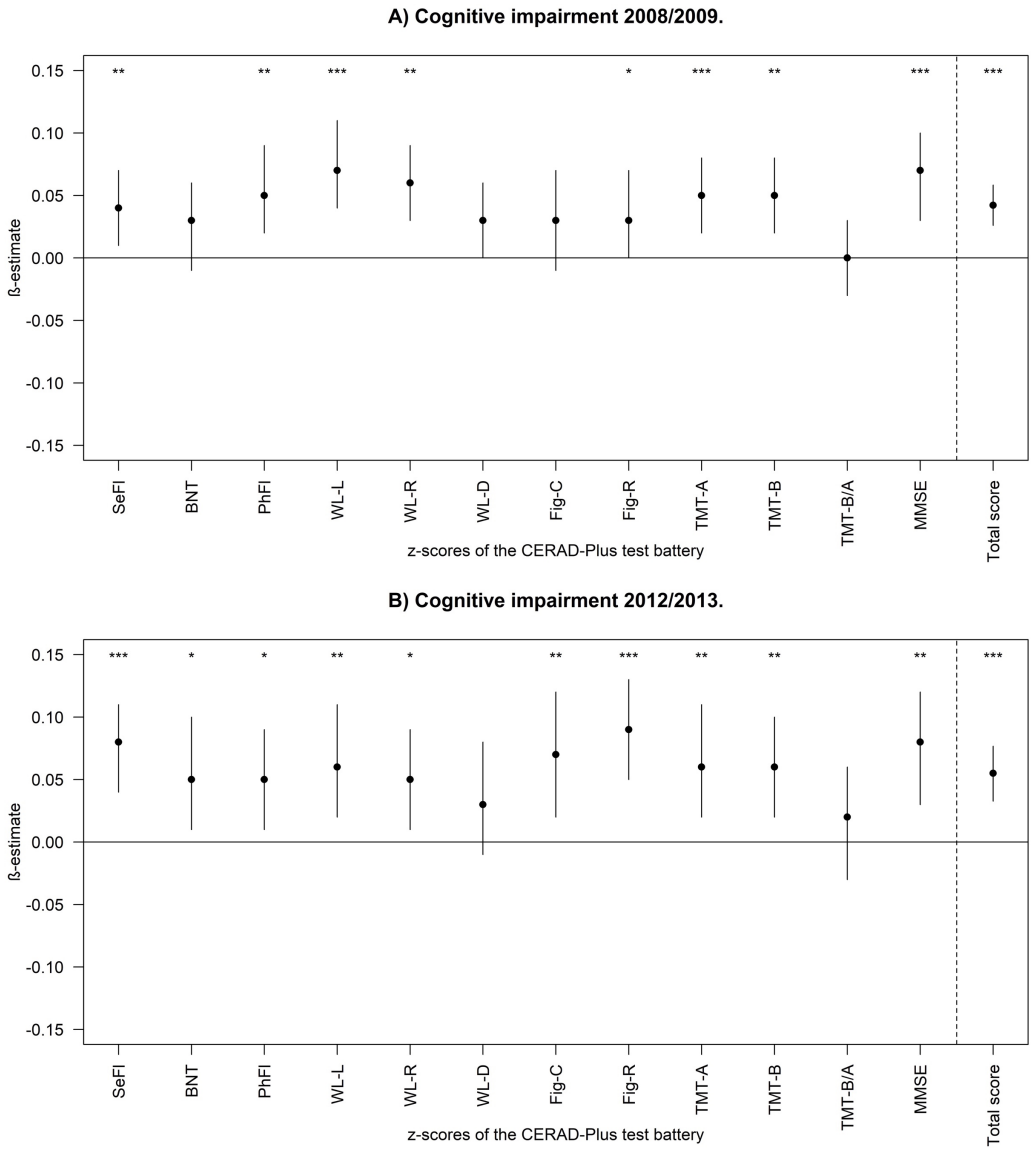
3.3 Association between odor identification and cognition

The association between odor identification and cognition at 2-time points in A) 2008/2009 and B) 2012/2013 are presented in Figure 4. Here again, adjustments were made for a large set of covariates.

For the follow-up investigation in 2008/2009, an increase of 1 point in the total number of correctly identified Sniffin' Sticks was significantly associated with an increase in most of CERAD subtests and the CERAD total score (p-value <0.001, β = 0.76 (95%-CI 0.47; 1.05)) in the main model. This association was also shown for the performance of CERAD-Plus test battery for the follow-up investigation in 2012/2013 (e.g. CERAD total score: p-value <0.001, β = 0.99 (95%-CI 0.59; 1.38)).

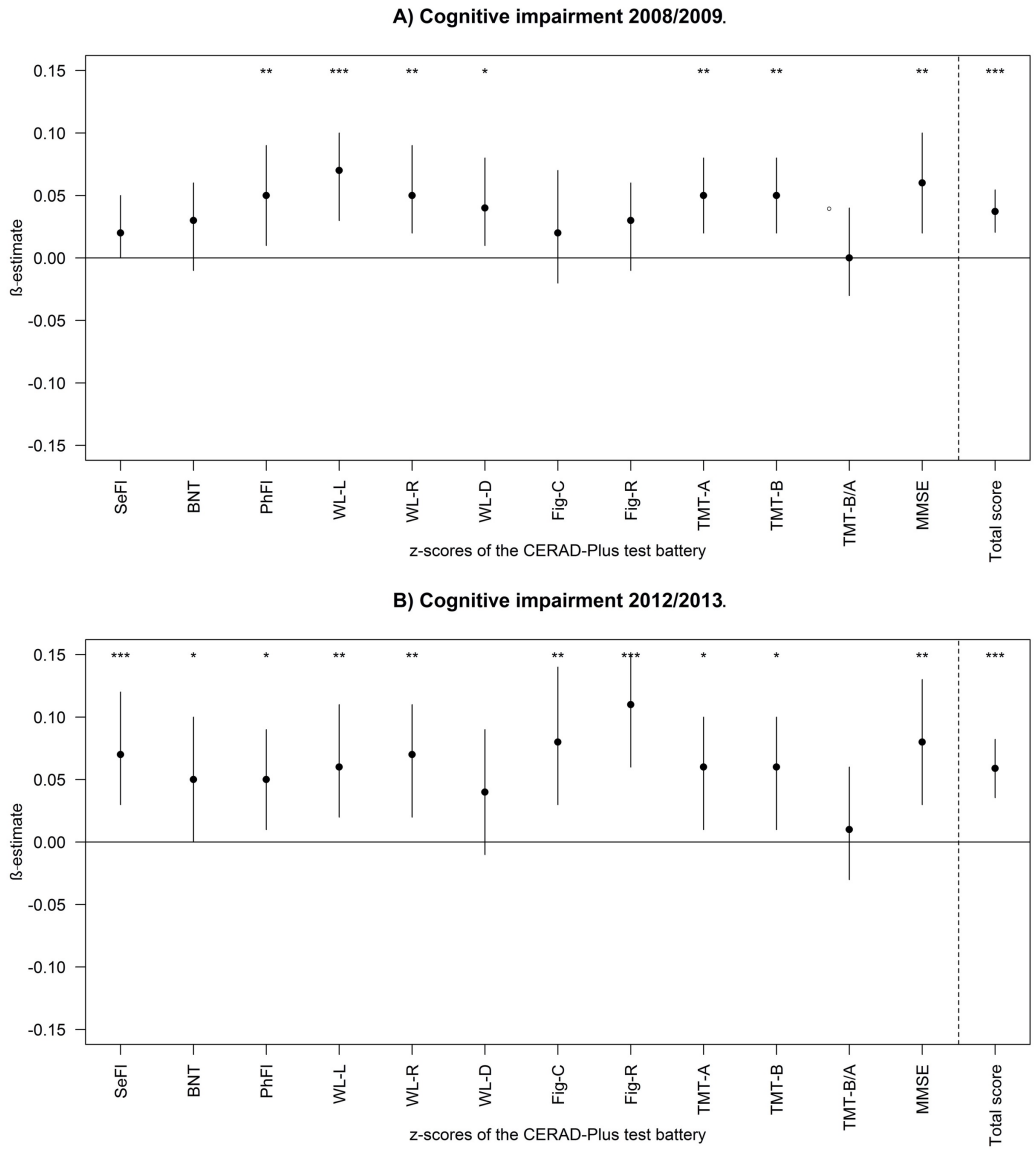
The extended model was additionally adjusted for diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension, and showed similar results for the association between odor identification and cognition for the follow-up in A) 2008/2009 and B) 2012/2013 (Figure 5).

Here, the difference in the distribution of the performance of the CERAD-Plus test was significantly associated with the total number of correctly identified Sniffin' Sticks for the follow-up investigations in 2008/2009 and 2012/2013 (e.g. CERAD total score: p-value <0.001, β = 0.67 (95%-CI 0.37; 0.98), 2008/2009; p-value <0.001, β = 1.06 (95%-CI 0.64; 1.48), 2012/2013).



Presented are β-estimates with corresponding 95%-confidence intervals (95%-CI) and p-values (p-value <0.05: *, p-value <0.01: **; p-value <0.001: ***). All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. SeFI = Semantic fluency, BNT = Boston Naming Test; PhFI = Phonetic fluency; WL-L = Word list learning; WL-R = Word list recall; WL-D = Word list recognition discriminability; Fig-C = Figure copying; Fig-R = Figure recall; TMT-A = Trail Making Test A; TMT-B = Trail Making Test B; TMT-B/A = Trail Making Test B / Trail Making Test A; MMSE = Mini Mental State Examination; Total score = the mean of the sum of z-scores of all CERAD subtests.

Figure 6: Associations between odor identification and z-scores of the CERAD-Plus test battery (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).



Presented are β-estimates with corresponding 95%-confidence intervals (95%-CI) and p-values (p-value <0.05: *, p-value <0.01: **; p-value <0.001: ***). All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, hay fever and **additionally for diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension**. SeFl = Semantic fluency, BNT = Boston Naming Test; PhFl = Phonetic fluency; WL-L = Word list learning; WL-R = Word list recall; WL-D = Word list recognition discriminability; Fig-C = Figure copying; Fig-R = Figure recall; TMT-A = Trail Making Test A; TMT-B = Trail Making Test B; TMT-B/A = Trail Making Test B / Trail Making Test A; MMSE = Mini Mental State Examination; Total score = the mean of the sum of z-scores of all CERAD subtests.

Figure 7: Associations between odor identification and z-scores of the CERAD-Plus test battery **additionally adjusted for diabetes, bronchial asthma, chronic bronchitis, COPD, and hypertension** (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).

3.4 Association between air pollution and cognition

The effects of long-term air pollution exposure on the performance of the CERAD-Plus test battery for the follow-up investigation in 2008/2009 (Table 8) and 2012/2013 (Table 9) are presented with β-estimates, corresponding 95%-CI, and p-values. Again, all associations were adjusted for important risk factors.

The exposure to air pollution was associated with lower performance of the CERAD-Plus test battery for the follow-up investigations in 2008/2009. Congruent with previous studies of the SALIA cohort (Schikowski *et al.*, 2015), the subtest of VCP ("Figure copying") was negatively associated with all air pollution values, with the strongest association for NO_2 (p-value <0.001, β = −0.28 (95%-CI −0.44; −0.13)). An increase of 1 IQR of PM_{10}, $PM_{2.5}$, and $PM_{2.5}$abs was also significantly associated with lower performance of the subtest of VCP (PM_{10}: p-value = 0.022, β = −0.14 (95%-CI −0.27; −0.02); $PM_{2.5}$: p-value = 0.049, β = −0.17 (95%-CI −0.34; <0.01); $PM_{2.5}$abs: p-value = 0.028, β = −0.12 (95%-CI −0.22; −0.01)). The subtest "Word list learning" and PM_{10} were also significantly associated for the follow-up investigation in 2008/2009 (p-value = 0.019, β = 0.13 (95%-CI 0.02; 0.24)).

For the follow-up investigation in 2012/2013, a strong association was observed between PM_{10} and cognitive impairment (CERAD total score: p-value = 0.036, β = 1.28 (95%-CI 0.09; 2.48). An increase of 1 IQR of PM_{10} was significantly associated with lower performance in the subtest "Word list learning" (p-value = 0.035, β = 0.14 (95%-CI 0.01; 0.28)). The association between NO_2 and the subtest of VCP was also significant (p-value = 0.027, β = −0.24 (95%-CI −0.45; −0.03)).

Table 8: Association between air pollution and cognition (follow-up investigation in **2008/2009** (N = 733)).

		SeFl	BNT	PhFl	WL-L	WL-R	WL-D	Fig-C	Fig-R	TMT-A	TMT-B	TMT-B/A	MMSE	Total score
PM_{10} [µg/m³]	**β-estimate**	0.02	−0.02	0.06	0.13	0.09	−0.02	−0.14	0.05	0.03	0.04	0.01	0.10	0.71
	95%-CI	−0.07; 0.10	−0.13; 0.08	−0.04; 0.16	0.02; 0.24	−0.01; 0.20	−0.12; 0.08	−0.27; −0.02	−0.05; 0.15	−0.06; 0.12	−0.06; 0.13	−0.08; 0.10	−0.01; 0.21	−0.21; 1.62
	p-value	0.708	0.658	0.277	0.019*	0.091	0.699	0.022*	0.376	0.533	0.476	0.917	0.081	0.130
$PM_{2.5}$ [µmg/m³]	**β-estimate**	0.04	−0.06	0.10	0.10	0.08	−0.07	−0.17	0.07	−0.01	0.04	0.05	0.09	0.56
	95%-CI	−0.08; 0.15	−0.20; −0.04	−0.04; 0.24	−0.05; 0.25	−0.06, 0.23	−0.21; 0.07	−0.34; <0.01	−0.07; 0.20	−0.14; 0.11	−0.09; 0.18	−0.08; 0.18	−0.07; 0.24	−0.69; 1.82
	p-value	0.555	0.456	0.151	0.185	0.254	0.320	0.049*	0.350	0.841	0.535	0.454	0.283	0.380
$PM_{2.5}$abs [10^{-5}/m]	**β-estimate**	0.01	0.01	0.05	0.08	0.08	−0.02	−0.12	0.01	0.01	0.07	0.05	0.05	0.55
	95%-CI	−0.07; 0.08	−0.08; 0.10	−0.03; 0.14	−0.01; 0.18	−0.01; 0.17	−0.11; −0.07	−0.22; −0.01	−0.07; 0.10	−0.07; 0.09	−0.02; 0.15	−0.03; 0.14	−0.05; 0.15	−0.23; 1.32
	p-value	0.884	0.845	0.226	0.072	0.090	0.609	0.028*	0.775	0.791	0.124	0.195	0.298	0.167
NO_2 [µg/m³]	**β-estimate**	0.01	−0.10	−0.04	0.03	0.05	−0.11	−0.28	−0.01	−0.03	<0.01	0.02	0.04	−0.44
	95%-CI	−0.01; 0.12	−0.23; 0.04	−0.17; 0.09	−0.11; 0.17	−0.09; 0.18	−0.24; 0.02	−0.44; −0.13	−0.14; 0.12	−0.14; 0.09	−0.12; 0.12	−0.11; 0.14	−0.11; 0.18	−1.62; 0.74
	p-value	0.865	0.163	0.542	0.715	0.501	0.108	<0.001*	0.888	0.670	0.984	0.795	0.630	0.470

Presented are β-estimates, 95%-confidence intervals (95%-CI), and p-values for an increase by 1 interquartile range (IQR) in air pollution (p-value <0.05: *). All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide; SeFl = Semantic fluency, BNT = Boston Naming Test; PhFl = Phonetic fluency; WL-L = Word list learning; WL-R = Word list recall; WL-D = Word list recognition discriminability; Fig-C = Figure copying; Fig-R = Figure recall; TMT-A = Trail Making Test A; TMT-B = Trail Making Test B; TMT-B/A = Trail Making Test B / Trail Making Test A; MMSE = Mini Mental State Examination; Total score = the mean of the sum of z-scores of all CERAD subtests.

Table 9: Association between air pollution and cognition (follow-up investigation in **2012/2013** (N = 542)).

		SeFl	BNT	PhFl	WL-L	WL-R	WL-D	Fig-C	Fig-R	TMT-A	TMT-B	TMT-B/A	MMSE	Total score
PM_{10} [µg/m³]	**β-estimate**	−0.05	0.04	0.01	0.14	0.11	0.08	0.04	0.09	0.08	0.08	<0.01	0.12	1.28
	95%-CI	−0.16; 0.07	−0.10; 0.17	−0.11; 0.12	0.01; 0.28	−0.02; 0.23	−0.05; 0.21	−0.11; 0.19	−0.04; 0.21	−0.04; 0.21	−0.05; 0.20	−0.12; 0.13	−0.02; 0.21	0.09; 2.48
	p-value	0.430	0.630	0.909	0.035*	0.103	0.236	0.640	0.165	0.208	0.215	0.955	0.090	0.036*
$PM_{2.5}$ [µg/m³]	**β-estimate**	−0.08	<0.01	0.01	0.17	0.07	0.06	−0.05	0.08	0.04	0.01	−0.04	0.08	0.62
	95%-CI	−0.25; 0.08	−0.20; 0.19	−0.16; 0.18	−0.02; 0.37	−0.11; 0.25	−0.12; 0.24	−0.27; 0.16	−0.10; 0.26	−0.14; 0.22	−0.16; 0.19	−0.22; 0.14	−0.12; 0.27	−1.09; 2.33
	p-value	0.314	0.973	0.895	0.074	0.466	0.524	0.637	0.374	0.696	0.870	0.676	0.431	0.479
$PM_{2.5}$abs [10^{-5}/m]	**β-estimate**	−0.02	0.02	0.01	0.05	0.03	0.04	0.01	0.04	0.08	0.04	−0.05	0.04	−0.50
	95%-CI	−0.12; 0.09	−0.10; 0.15	−0.10; 0.12	−0.07; 0.18	−0.09; 0.15	−0.07; 0.16	−0.13; 0.15	−0.07; 0.16	−0.03; 0.20	−0.08; 0.15	−0.17; 0.07	−0.08; 0.17	−0.61; 1.61
	p-value	0.725	0.745	0.892	0.411	0.591	0.465	0.853	0.474	0.161	0.537	0.410	0.508	0.376
NO_2 [µg/m³]	**β-estimate**	0.03	−0.03	0.11	0.14	0.01	<0.01	−0.24	0.06	<0.01	−0.05	−0.09	0.04	0.29
	95%-CI	−0.13; 0.19	−0.22; 0.16	−0.05; 0.28	−0.05; 0.33	−0.17; 0.19	−0.17; 0.18	−0.45; −0.03	−0.11; 0.18	−0.17; 0.18	−0.23; 0.12	−0.27; 0.08	−0.15; 0.24	−1.38; 1.96
	p-value	0.680	0.756	0.180	0.144	0.931	0.960	0.027*	0.477	0.957	0.539	0.297	0.653	0.735

Presented are β-estimates, 95%-confidence intervals (95%-CI), and p-values for an increase by 1 interquartile range (IQR) in air pollution (p-value <0.05: *). All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide; SeFl = Semantic fluency, BNT = Boston Naming Test; PhFl = Phonetic fluency; WL-L = Word list learning; WL-R = Word list recall; WL-D = Word list recognition discriminability; Fig-C = Figure copying; Fig-R = Figure recall; TMT-A = Trail Making Test A; TMT-B = Trail Making Test B; TMT-B/A = Trail Making Test B / Trail Making Test A; MMSE = Mini Mental State Examination; Total score = the mean of the sum of z-scores of all CERAD subtests.

3.5 Mediation analysis between air pollution and cognitive impairment mediated by odor identification

We performed a mediation analysis on the pathway from air pollution to cognitive impairment using additional adjustment for odor identification to answer the a priori hypothesis (Table 10). The estimated percent of the association between long-term air pollution exposure and cognitive impairment that was mediated by a decrease in odor identification are given. Presented are also indirect, direct, and total effects with β-estimates, corresponding 95%-CI, and p-values. We adjusted all associations for age, educational level, BMI, smoking status, living in an urban vs. rural area, cold, and hay fever.

Odor identification did not mediate the association between air pollution and the performance of the CERAD-Plus test battery. For the follow-up investigation in 2008/2009, the estimated proportion mediated by odor identification for the association between long-term exposure to ambient air pollution and cognitive impairment was less than 3% for PM_{10} (p-value = 0.812). The association between air pollution – PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2 for the follow-up investigation in 2008/2009 – and a decrease in cognitive impairment in 2012/2013 were not significantly mediated by a reduction in odor identification (estimated proportion mediated = −1.3%, p-value = 0.868 for PM_{10}).

Odor identification did not mediate the association between air pollution and further CERAD subtests (e.g. VCP: estimated proportion mediated was less than 1% for PM_{10} in 2012/2013). A selection of mediation analyses for further CERAD subtests is presented in Appendix A.

Table 10: Mediation analysis between air pollution and **cognitive impairment** mediated by odor identification (follow-up investigations in A) 2008/2009 (N=733) and B) 2012/2013 (N=542)).

	Average indirect effect			Average direct effect			Total effect			Estimated proportion mediated	
	β-estimate	95%-CI	p-value	β-estimate	95%-CI	p-value	β-estimate	95%-CI	p-value	Proportion mediated %[a]	p-value
A) Cognitive impairment 2008/2009.											
PM_{10} [µg/m^3]	0.02	−0.15; 0.21	0.826	0.87	<0.01; 1.70	0.048	0.89	0.02; 1.72	0.042	2.7	0.812
$PM_{2.5}$ [µg/m^3]	−0.04	−0.31; 0.22	0.746	0.91	−0.31; 2.09	0.176	0.87	−0.34; 2.07	0.198	−3.3	0.844
$PM_{2.5}$ abs [10^{-5}/m]	0.02	−0.14; 0.18	0.830	0.69	−0.09; 1.44	0.082	0.71	−0.07; 1.47	0.078	2.6	0.816
NO_2 [µg/m^3]	−0.03	−0.26; 0.21	0.832	−0.30	−1.45; 0.84	0.616	−0.33	−1.48; 0.83	0.618	5.2	0.802
B) Cognitive impairment 2012/2013.											
PM_{10} [µg/m^3]	−0.02	−0.29; 0.25	0.862	1.52	0.42; 2.59	0.006	1.49	0.41; 2.55	0.006	−1.3	0.868
$PM_{2.5}$ [µg/m^3]	−0.15	−0.57; 0.22	0.412	1.08	−0.50; 2.59	0.224	0.93	−0.69; 2.51	0.314	−9.2	0.634
$PM_{2.5}$ abs [10^{-5}/m]	−0.01	−0.27; 0.25	0.930	0.91	−0.18; 1.99	0.106	0.90	−0.21; 2.01	0.120	−0.6	0.978
NO_2 [µg/m^3]	−0.01	−0.47; 0.28	0.666	−0.22	−1.04; 2.07	0.522	0.44	−1.13; 2.06	0.600	−1.7	0.950

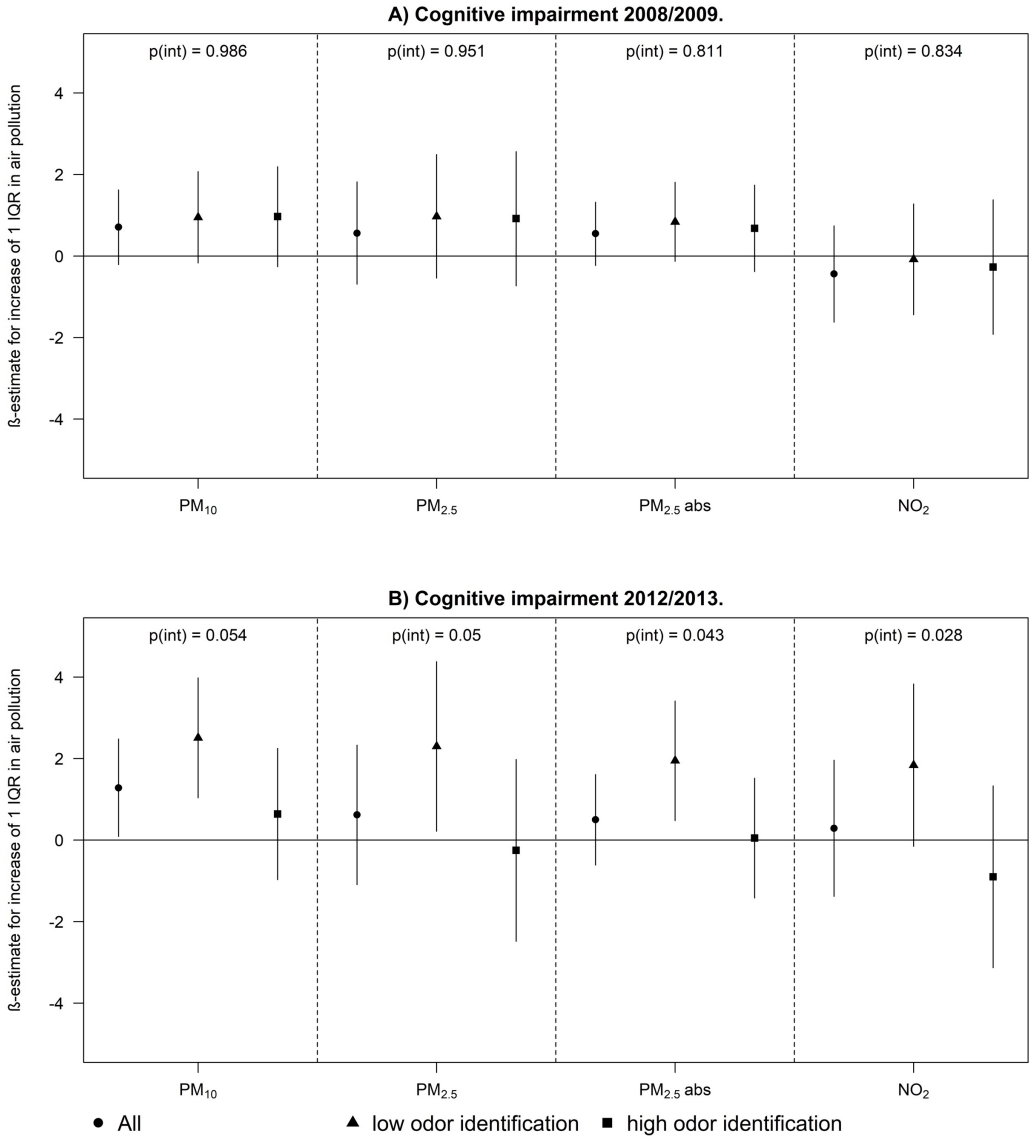
Estimated percent of the association between long-term air pollution exposure and cognitive impairment that was mediated by a decrease in odor identification. Approach utilizing exposure and mediator averages with interaction. Exposure: air pollution; mediator: odor identification; outcome: **cognitive impairment** (CERAD total score). Presented are indirect, direct, and total effects (β-estimates, 95%-confidence intervals (95%-CI), and p-values). All models were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide. [a]: Proportion mediated = mediated effect / (direct effect + mediated effect) x 100.

3.6 Interaction analysis between air pollution and cognitive impairment with reduced odor identification

This analysis was conducted to investigate whether the association between air pollution exposure and cognitive impairment changes with odor identification as a moderator variable for the follow-up investigations in A) 2008/2009 and B) 2012/2013 (Figure 6). The association between air pollution and cognition was categorized in subgroups defined by low (<median of total number of correctly identified Sniffin' Sticks) vs. high odor identification (≥median of total number of correctly identified Sniffin' Sticks). Presented are β-estimates with corresponding 95%-CI for an increase of 1 IQR in air pollution and p-values for the interaction terms. All associations were again adjusted for a large set of covariates.

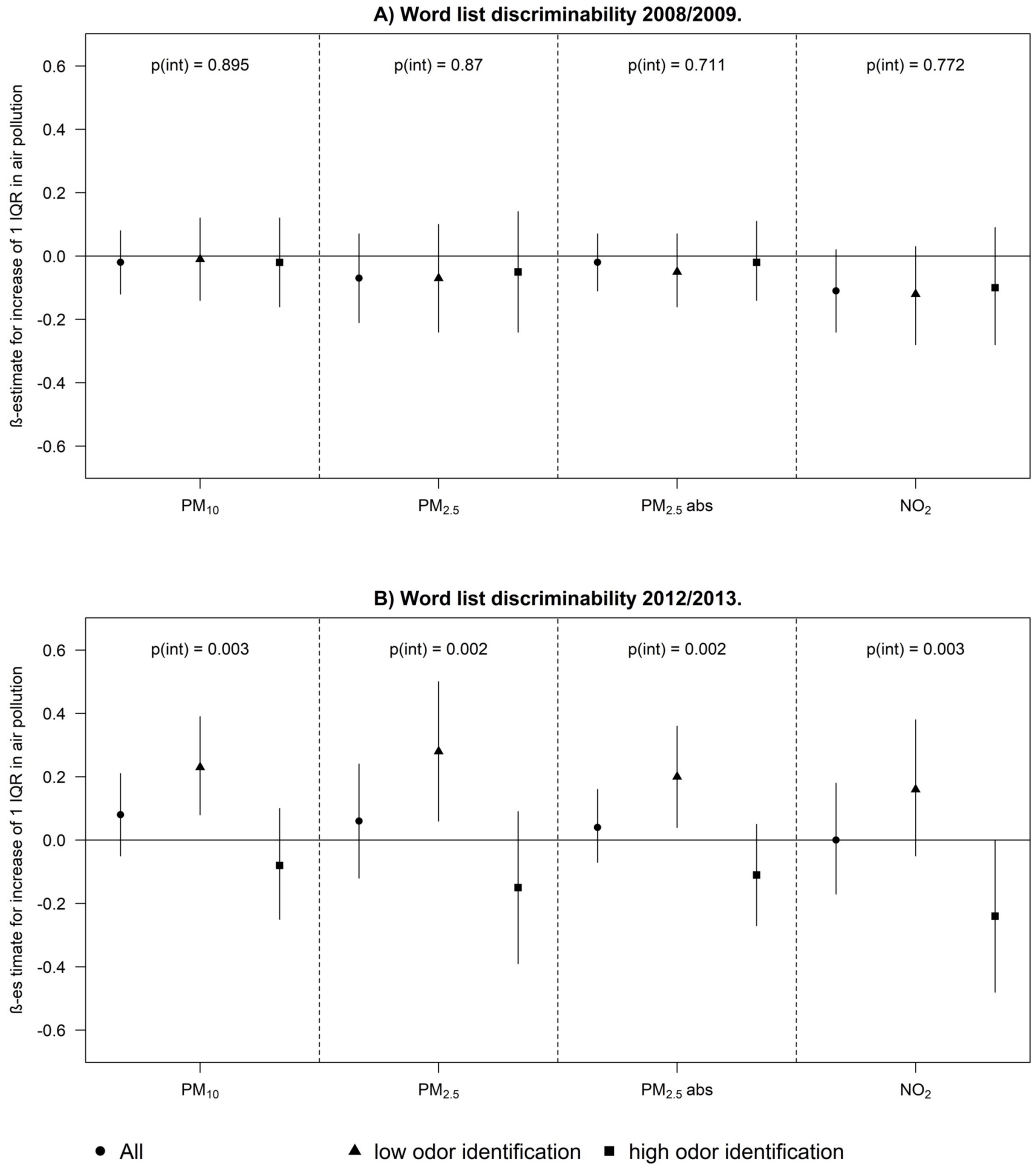
The association between air pollution exposure and cognitive impairment changed depending on odor identification as a moderator variable. Results for the follow-up investigation in 2008/2009 varied: interaction was significant for $PM_{2.5}$abs and the CERAD subtests "Word list learning" (p-value = 0.016), while there was no significant interaction between air pollution exposure, odor identification, and the CERAD total score (e.g. p-value = 0.986 for PM_{10}). The effect of air pollution exposure on cognitive impairment for the follow-up investigation in 2012/2013 interacted significantly with the performance of the odor identification test. The interaction analysis showed significance for all air pollution values and the CERAD total score, with the strongest interaction for NO_2 (p-value = 0.028). The CERAD subtest "Word list discrimination" also interacted significantly with all air pollution values and the odor identification test (e.g. p-value = 0.003 for PM_{10}) (Figure 7).

The interaction analysis for the CERAD subtests "Semantic fluency" (p-value = 0.030 for $PM_{2.5}$abs) and "Word list learning" (p-value = 0.039 for $PM_{2.5}$) were significant as well. The CERAD subtest of VCP, however, did not show significance in the interaction analysis (Appendix B).



Presented are β-estimates with corresponding 95%-confidence intervals (95%-CI) for the association between air pollution and **cognitive impairment** (CERAD total score) in subgroups defined by low (<median of total number of correctly identified Sniffin' Sticks) vs. high odor identification (≥median of total number of correctly identified Sniffin' Sticks) for an increase of 1 interquartile range (IQR) in air pollution. All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide.

Figure 8: Interaction analysis between air pollution and **cognitive impairment** with odor identification as a moderator variable (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).



Presented are β-estimates with corresponding 95%-confidence intervals (95%-CI) for the association between air pollution and impairment in the CERAD subtest **"Word list discriminability"** in subgroups defined by low (<median of total number of correctly identified Sniffin' Sticks) vs. high odor identification (≥median of total number of correctly identified Sniffin' Sticks) for an increase of 1 interquartile range (IQR) in air pollution. All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide.

Figure 9: Interaction analysis between air pollution and the CERAD subtest **"Word list discriminability"** with odor identification as a moderator variable (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).

4 Discussion

4.1 Summary of key results

Our study investigated chronic health effects of long-term ambient air pollution exposure in study participants of a population-based cohort of elderly women. Data was first collected in the early 90s and follow-up investigations were carried out until 2012/2013. We were unable to demonstrate an association between long-term air pollution and reduced odor identification. Odor identification and cognition, on the other hand, were associated cross-sectionally. In our linear regression analysis, long-term exposure to air pollution was associated with cognitive impairment in 2008/2009 and 2012/2013. We conducted further analyses to better understand the role of the olfactory system on the pathway between air pollution exposure and cognitive impairment. Although no association between air pollution and the mediator (odor identification) was found, the mediation analysis was nevertheless carried out to answer the a priori hypothesis. The interaction analysis indicates that the association between air pollution exposure and area-specific cognitive abilities changes depending on reduced odor identification.

4.2 Study design and methods

We recognize various methodological strengths, but also limitations of our study which should be taken into account when interpreting the results. Direct comparison with previous studies is generally limited due to differences in study design and population, assessment of exposure, odor identification, and cognition, adjustment for covariates, and statistical methods.

Our study was carried out using data of the population-based SALIA cohort. The study population consisted of older women of the same age, who have lived at a single address in the Ruhr area since the beginning of the SALIA investigations in the early 1980s and who have been participating in the follow-up investigations since then.

A key strength of the SALIA cohort is the over 20-year follow-up period which allows us to investigate the chronic health effects of exposure to air pollution and to provide valid results. The epidemiological study design is generally well suited for the study of chronic health effects of long-term air pollution. We were able to investigate a randomly selected group of older women under real conditions and under the adjustment of important risk factors for a possible association between air pollution exposure, reduced odor identification, and cognitive impairment. Another strength is the composition of the study population. The study participants mainly consisted of housewives who were neither highly exposed to air pollution, nor particularly ill or susceptible to the health effects of air pollution exposure. Therefore, they can be representative of a general population sample of elderly women. As housewives, the study participants have probably spent a lot of time at home, which is why this cohort was particularly well suited for estimating air pollution in the immediate vicinity of the participant's home address.

On the other hand, it must be recognized that our study area, the Ruhr area, is historically known for high levels of air pollution due to its coal and mining history. Air quality has changed dramatically due to preventive interventions in the 20 years between the baseline and the most recent follow-up investigations of the SALIA cohort (baseline data not shown). It is plausible that health effects were mainly caused by the extreme air pollution in the past decades and only became visible after a longer period of time. Exposure levels to air pollution used for this

study were much lower compared to the baseline investigation and may no longer be associated with the performance of the Sniffin' Sticks and the neuropsychological tests. Under real conditions, it is difficult to uncover the differences between the lowest and highest levels of air pollution because the differences between the selected groups who were exposed to the different levels of air pollution are often small. Therefore, it is necessary to study larger study populations. We may have missed significant associations because the moderate sample size of our study population limits the ability to detect true effects. All participants provided detailed information on potential and known covariates using a self-administered standardized questionnaire. Although we have adjusted for a large set of covariates, it is possible that other factors induce a loss of odor identification and a predisposition to cognitive impairment. Our results may also have been affected by loss of follow-up. During the more than 20 years of the study period, only the most resistant participants may have survived. Thus, one limitation of our study may be a selection towards healthy and surviving participants. Additionally, it should be noted that there may have been a non-responder bias in the baseline investigation. In contrast to the general population, our study participants mainly had a higher level of education. This would probably have led to an underestimation of the real impact of air pollution exposure on the performance of the Sniffin' Sticks and the neuropsychological tests. However, we do not assume that this could bias the results since participation was not related to air pollution exposure and study participants with higher level of education were randomly distributed throughout the study area (Gehring *et al.*, 2006; Ranft *et al.*, 2009; Schikowski *et al.*, 2015).

Research in the format of epidemiological studies is particularly useful for studying the effects of air pollution on public and individual health over a long period of time. Conclusions apply especially when the study period is long, when a large sample of randomly selected participants is observed, and when as many different risk factors as possible are assessed. In this context, it is also important to identify particularly vulnerable times in life and to examine them extra carefully. The analyses carried out in our study meet all of these requirements: our study period was more than 20 years, our study participants were older women from the Ruhr area who were selected at random, extensive investigations were performed and multiple risk factors were identified and taken into account. Such

study designs promise to continue to uncover previously unknown associations between air pollution and health effects.

One of the major strengths of our study is that air pollution was detailly assessed in the immediate vicinity of the participants' home address. The exact geographic coordinates of the address of each study participant were determined and corresponding air pollution levels were assigned based on measurements and LUR models from the ESCAPE campaign 2008–2011 (Eeftens *et al.*, 2012; Beelen *et al.*, 2013). LUR models estimated long-term average concentrations of exposure markers and provided information about the spatial distribution of the annual mean. GIS data on the land use characteristics near monitoring stations were also taken into account. As a result, the exposure assessment was less susceptible to random variation in one area and one year. Another strength is the 20-year follow-up period of the SALIA cohort. The pattern of air pollution of the investigated study area has remained the same over the past few decades. Therefore, the exposure assessment of each participants' home address reflected long-term exposure (Ranft *et al.*, 2009; Vierkötter *et al.*, 2010).

Exposure to air pollution and its chronic effects on human health are difficult to assess. Although we are always exposed to all components at the same time, for research purposes, a few air pollutants must be sufficient as a marker for the entire pollutant mixture. In our study, PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2 have been studied separately. Since these air pollutants are known to highly correlate with the mixture of air pollutants in an anthropological environment, such as the Ruhr area, they were used as a proxy for the complex interactions of this air pollutant mixture. Ultimately, we only provide indirect evidence for the effects of air pollution. Associations between certain pollutant markers and health effects are not necessarily the only causal consequence of a single air pollutant. The observed effects could rather be attributed to the complex interactions of the mixture. It is conceivable that only the mixture of all components together has harmful effects on health. The use of more complex regression models is crucial to investigate the additive or synergistic effects of various air pollutants.

The differences between the effects of the air pollutants we used may partly be due to the fact that twice as many calibration points were used for NO_2 as for PM. However, according to previous studies, a differential measurement error from

this source is not necessarily an explanation for the difference in effects (Vierkötter *et al.*, 2010). It is known that UFP from combustion processes in motor vehicles vary on a small-scale spatial environment around busy roads whereas the mass distribution of particles on a large-scale is dominated by larger particles (Zhu *et al.*, 2002; Ranft *et al.*, 2009). LUR models can estimate traffic-related air pollution with sufficient spatial resolution (Teichert *et al.*, 2013). They were validated to ensure that the predictability of these models was not overestimated and to explain the substantial fraction of the variations in air pollution concentrations in most areas (Wang *et al.*, 2013). Thus, the assessment of air pollution in our study is particularly reliable. However, it should be noted, that different markers of air pollution may have similar spatial distributions over time, but the composition of the air pollutant mixture itself may change significantly due to new fuels and implemented engine technologies over time (Schikowski *et al.*, 2015). In general, it is difficult to fully assess the mixture of air pollutants in studies. Experimental studies mainly deal with the investigation of the toxicological properties of individual pollutants, since the mixture of pollutants in the ambient air is difficult to reproduce in laboratory experiments on animals or volunteers. Research in the format of epidemiological studies use 1 or more pollutant as markers for a pollutant mixture and can at least estimate approximations for the complex interactions of this mixture (Künzli, Perez and Rapp, 2010). Therefore, our exposure assessment is suitable for researching the association between ambient air pollution and chronic health effects.

Air pollution has been assessed in different studies using different measurement methods. In the future, it will be important to agree on uniform markers for the air pollutant mixture. Given the complexity of air pollution, it is not surprising that no single method or marker has emerged that can be used universally. In order to be able to assess the effects of air pollution on health in studies, not only further research approaches will be required, but openness and willingness to discuss.

The Sniffin' Sticks test is a validated screening tool for the assessment of olfactory function (Hummel *et al.*, 1997, 2001; Kobal *et al.*, 2000; Doty, 2006). It should be noted that the Sniffin' Sticks test used in this study is a simplified version addressing only odor identification, but neither the odor thresholds, nor the odor discrimination. The elaborate version of the test provides detailed

information about the olfactory system. First, the assessment of odor identification in our study was chosen in order to satisfy the time restrictions when testing study participants in the SALIA investigations. Second, the highest test-retest reliability was continuously found for odor identification. Third, compared to other tests, the Sniffin' Sticks were generally better suited to determine a reduced olfactory function over a lifetime. In particular, the test participants' ability to identify odors decreased over the age of 65 years. Fourth, the test was repeatedly validated to screen for olfactory dysfunction (Doty *et al.*, 1995; Hummel *et al.*, 1997, 2001). Different definitions were used for hyposmia. Kobal *et al.* (2000) defined hyposmia as the 10th percentile score of 16- to 35-year-old test subjects. Hummel *et al.* (2001) decided to exclude 4 odors from the screening test on the basis of identification rates below 55% for garlic, turpentine, and apple and because anise and licorice were regarded as too similar. Their 12-item odor identification test used the 90th percentile with 10 points as the cut-off value for hyposmia. Due to the inconsistencies, and because definitions were made based on age groups that did not correlate with the mean age of our study cohort, we decided to use the original 16-item Sniffin' Sticks test and preferred the total number of correctly identified Sniffin' Sticks out of 16 for our analysis instead. According to normative data for Sniffin' Sticks (Hummel *et al.*, 2007), the participating women in our study performed within the expected range of their age group.

Nonetheless, the measurement of the olfactory system using Sniffin' Sticks remains only a proxy to assess odor identification. Odor identification is complex and influenced by both, olfactory and neurological components. Even if the neurological component is controlled by the CERAD-Plus test, this might affect the validity of our results. Therefore, our results can only ever be seen as indicators for further investigations in this field of research (Ranft *et al.*, 2009).

In addition, it is very difficult to assess transport routes using epidemiological studies. Our study was based on the hypothesis of a transport route from nose to brain along the olfactory nerve which is generally difficult to prove right or wrong. In this context, it should also be acknowledged that epidemiological studies by nature cannot prove hypothesized transport routes (Rückerl *et al.*, 2011).

The olfactory system as a possible transport route related to air pollutants and their health effects is promising, but further research is needed. Future approaches should pay particular attention to the use of uniform definitions for a reduced olfactory function and establish instruments that examine in detail the complex processing of olfactory information in connection with cognitive abilities.

The CERAD-Plus test battery has long been a validated, normed tool and is known to be very sensitive for the early detection of cognitive impairment. The CERAD z-scores are already individually adjusted for age, level of education, and gender. Z-scores close to 0 indicate that participants performed on average within the normal range of the CERAD-Plus test battery. The test allows the comprehensive and individual assessment of area-specific cognitive abilities. In this way, we were able to assess the chronic effects of air pollution on different areas of the brain (Aebi, 2003; Fillenbaum *et al.*, 2008; Karrasch *et al.*, 2013).

The assessment of cognition was analyzed at 2-time points. Chronic health effects develop over a long period of time and on the basis of individual genes, habits, living and working situations which is why cognitive impairment is very difficult to test, even with the help of neuropsychological tests. Daily changes are also typical for such neuropsychological disorders. Therefore, poor performance of cognitive tests is only an indicator of cognitive impairment. The performance of our study participants improved in most subtests over time which is highly unlikely. We were able to determine that the women who had taken part in both follow-up investigations had already performed better compared to the whole study population at the first follow-up investigation. However, we suspect the influence of a learning effect and healthy survival bias, which is why the analysis with data of the second assessment of cognition must be interpreted taking this assumption into account. Furthermore, we cannot rule out selection bias, because the risk of cognitive impairment increases sharply due to age-related factors. Many different causes may be important in developing cognitive impairment in the elderly. Even after the adjustment for important risk factors, these other causes could in turn have changed the effect of air pollution exposure on cognition. Failure to address the cause of death may have led to an underestimation of the association between air pollution and MCI, because some participants may have died from neurological disorders. In addition, women with

neurological disorders may no longer have participated in the study which could have masked an association between exposure to air pollution and cognitive impairment in this age group (Ranft *et al.*, 2009; Schikowski *et al.*, 2015).

Complex neurological tests are used to assess cognition in studies. Cognitive abilities are summarized under various general terms. The term cognition is also not used uniformly. However, comparisons between different studies are more difficult if terms are not used consistently. This should be kept in mind for future research.

For odor identification to act as a mediator for the effect of air pollution on the outcome of cognitive impairment, air pollution exposure should precede the mediator variable of odor identification and the mediator should in turn precede the outcome variable of cognitive impairment. Air pollution exposure and odor identification should also be correlated, and either the mediator's main effect on the outcome, or the interaction between exposure and mediator should be significant. On the other hand, odor identification should be treated as a moderator rather than a mediator variable, if there was no association between air pollution exposure and the moderator variable of odor identification, if the moderator variable preceded air pollution exposure, and if the interaction between exposure and moderator was significant (Baron and Kenny, 1986; Kraemer *et al.*, 2001; Valeri and VanderWeele, 2013). In this context, our study indicates that the association between air pollution and cognitive impairment changes depending on odor identification as a moderator variable. Additional covariates may also be important for the association between exposure, mediator, moderator, and outcome variable. To rule out bias, it must be ensured that the estimates of the analysis are adjusted for all relevant covariates. Relevant covariates should be identified beforehand and integrated into correctly specified models (MacKinnon, Krull and Lockwood, 2000; Valeri and VanderWeele, 2013). Even if we adjusted for multiple covariates, further covariates may be relevant for the associations we examined.

Complex associations can only be discovered with the help of complex analyses. When it comes to air pollution-induced health effects, various hypotheses have to be carefully investigated with adjustment for all important risk factors. However, it must also be accepted if there are other correlations than originally expected.

4.3 Air pollution and odor identification

Our findings of the SALIA cohort showed no association between long-term air pollution exposure and reduced odor identification even after adjusting for additional covariates.

We investigated the association between air pollution and olfactory function in linear regression models adjusted for multiple covariates. However, the validity of our results may be limited, because we only performed a cross-sectional analysis. Air pollution exposure and odor identification were assessed at a single point in time. Therefore, we were unable to estimate the association between air pollution and changes in odor identification of individual participants over time.

When first studies on air pollution-induced health effects were carried out, they mainly focused on the respiratory tract. In the mid 1990s, it was recognized that chemosensory senses, such as tasting or smelling, could also be affected by many potential environmental hazards (Halpern, 1982). It was recognized that different anthropogenic and naturally occurring chemicals and substances also had an impact on the olfactory system (Schiffman and Nagle, 1992). Cumulative exposure to industrial vapors and the association with odor loss decreased with increasing duration since the last exposure (Schwartz *et al.*, 1989). Further studies confirmed the deleterious effects of airborne chemical components on the olfactory system (Norès, Biacabe and Bonfils, 2000; Gobba, 2006). However, the effects of highly concentrated chemical components are difficult to compare with the effects of relatively low concentrations of air pollutants in the environment that were investigated in our study. These studies already indicated that the olfactory mucosa is one of the few places in the nervous system where new neurons constantly emerge – even in adulthood. Our study area was long known for high concentrations of air pollutants. It could be possible that air pollutants previously damaged the olfactory system of our study participants, but this may not have been detectable at the time of our follow-up investigations, since new olfactory neurons had emerged in between.

Few studies investigated the mechanism of uptake and transport of particles from the air into the brain along the olfactory nerve. Initially, the main focus was on easily traceable metals or radioactive labeled isotopes. In comparison to other

metals, the ionic Mn seemed to be taken up particularly well via the olfactory tract and to reach other areas of the brain transneuronally. After intranasal instillation, Mn was detected in the olfactory bulb of different animals. It was assumed that the occupational neurotoxicity of inhaled Mn may be related to an uptake of the metal into the brain via the olfactory tract (Tjälve and Henriksson, 1999). Later, it was confirmed that the olfactory transport route for inhaled Mn oxide particles, referred to as solid UFP, may efficiently reach the CNS and cause inflammatory changes (Elder *et al.*, 2006; Lucchini, Dorman, *et al.*, 2012). Still, it is difficult to provide direct evidence of olfactory transport of Mn in humans. The transport of nasally administered thallium in the human olfactory bulb along the olfactory nerve was investigated using a combination of imaging techniques. Healthy participants were nasally administered thallium into the olfactory cleft. 24 hours later, different imaging techniques were used to visualize the thallium distribution. Thallium was found in the olfactory bulb in the anterior cranial fossa which supported the hypothesis of a functional transport route from nose to brain in humans (Shiga *et al.*, 2011). Later, they suggested that with reduced olfactory function, thallium-based imaging in the olfactory bulb decreased compared to healthy participants and that patients with intact olfactory nerve fibers could be selected using thallium-based imaging to track olfactory dysfunction over a longer period of time (Shiga *et al.*, 2013, 2017). The results of this working group are difficult to compare with ours. On the one hand, a significantly smaller number of participants was examined, on the other hand, the participants were predominantly male and also much younger than our study participants. In addition, thallium was not inhaled in this study, but rather instilled into the olfactory cleft with a syringe. In this context, pathways related to breathing may be very different. It must also be noted that thallium, but also Mn, are generally difficult to compare with the air pollutants we investigated.

One of the first approaches to translocation and accumulation of inhaled particles along the olfactory nerve and in areas of the brain was by Oberdörster *et al.* (2004). A study in rats showed a significant and persistent increase in the olfactory bulb on the days after the inhalation of UFP. Approximately 20% of the inhaled UFP deposited on the rat olfactory mucosa migrated to the olfactory bulb. However, it should be noted that the animals' breathing occurs more frequently through the nose and that results from animal experiments are difficult to compare

with those in cohort studies. Inhalation of certain metal dusts or aerosols at work was associated with different effects on the olfactory system (Sunderman, 2001; Doty, 2006; Gobba, 2006). However, this type of high-level exposure at the workplace is also difficult to compare with our assessment of the ambient air at the home address of our study participants. In general, the deposition of particles in the nasopharyngeal area also depends strongly on the particle size.

One of the first human studies was conducted in Mexico City and examined the effects of ambient air pollutants on the olfactory epithelium. A group of urban dwellers was compared to residents of a relatively clean port city on the Mexican Pacific coast. A group of young men, who had only recently lived in Mexico City, was also examined and observed over a 3-month period. There was an increased number of single-stranded DNA breaks in the olfactory epithelium of city dwellers compared to the controls. The number of single-stranded DNA breaks in young men, who had just moved into the city, also increased during the observation period (Calderón-Garcidueñas *et al.*, 1996). Compared to our study, the conclusions of this first research approach are limited due to the small sample size. Another limitation is that possible damage to the olfactory function was not measured. However, the effect of ambient air pollution on the olfactory function of Mexico City residents was later investigated. Participants aged 20–63 years were compared to controls from a rural region for perception and threshold of odors of everyday drinks. Here, too, there was an association between air pollution and the olfactory function. However, this association was only significant in the younger age groups and not in the 50–63 age group (Hudson *et al.*, 2006). Later, Mexican researchers showed that urban air pollution impaired intranasal sensitivity to trigeminal nerves and olfactory function, the latter was measured using the Sniffin 'Sticks test (Guarneros *et al.*, 2009). They also found indications for an association between air pollutants and olfactory bulb inflammation and reduced odor identification (Calderón-Garcidueñas *et al.*, 2010). In both studies, the associations were shown in otherwise healthy young men and women, while our study population consisted only of older women. After all, the urban air in Mexico City is very different from to the urban air that our study participants were exposed to in Germany. Air pollution was not assessed, but rather 2 different study areas were selected to represent different levels of air pollution. Even if associations have been found, it cannot be guaranteed whether these are

actually solely due to air pollution or whether other factors have contributed to the associations. It should be noted that inconsistencies may also be due to that fact.

This approach was recently taken up and continued. In the U.S.A., a study population consisting of urban adults aged 57–85 years showed an association between $PM_{2.5}$ and NO_2 exposure and reduced odor identification (Adams *et al.*, 2016; Ajmani *et al.*, 2016). Interestingly, the most deleterious effects were observed in 57- to 64-year-olds, an age group comparable to the baseline age of our study participants. Air pollution was measured at monitoring sites linked to each participant's home address within a 60 km grid and assessed as the moving average of 1 year prior to the health assessment as the main exposure window of interest. The study population included men and women who may not have spent a lot of time at home in the year prior to the health assessment, but rather spent more time in other heavily polluted locations. In contrast, our participants lived at a single address for a long time and were actually exposed to the air pollution that was present in this environment at different times of our exposure assessment. Since most of them were housewives, it can be assumed that they usually also spent more time at home. Additionally, the olfactory function was only measured with 5 Sniffin' Sticks, and categorized based on a cut-off value that was not adapted to different age groups. In contrast, we used 16 Sniffin' Sticks and the number of correctly identified Sniffin' Sticks of our study participants as a variable for the olfactory function.

Although the importance of human olfactory function and the many difficulties that arise from olfactory dysfunction were recognized early on, the corresponding data are still lacking to make the conclusive case between air pollution and sense of smell as it has with heart and lung health (Halpern, 1982; Upadhyay and Holbrook, 2004; Ajmani, Suh and Pinto, 2016; Genter and Doty, 2019).

4.4 Odor identification and cognition

In our study, the performance of the odor identification test was significantly associated with cognition assessed with the neuropsychological CERAD-Plus test battery. The number of correctly identified Sniffin' Sticks was strongly associated with the CERAD total score and most CERAD subtests.

We estimated multiple regression models for the association between reduced odor identification and cognitive impairment at 2-time points. This contributed to the reliability of our results and enabled us to draw conclusions about the associations and changes for individual declines of individual participants.

Our results are consistent with the evidence provided by others. It was discovered early on that olfactory dysfunction and pathologies in the olfactory system were associated with neurodegenerative diseases, such as AD and PD (Doty, Deems and Stellar, 1988; Hyman, Arriagada and Hoesen, 1991). The predictive benefits of odor identification deficits for patients with MCI, who later developed Alzheimer's, were also recognized at an early stage (Devanand *et al.*, 2000). Since then, olfactory dysfunction has been repeatedly demonstrated as an early sign of neurodegenerative diseases (Doty, 2009, 2012; Hüttenbrink *et al.*, 2013; Zou *et al.*, 2016; Marin *et al.*, 2018). AD and PD were also associated with visual-spatial deficits (Crucian and Okun, 2003) which is one of the cognitive abilities in our study that was associated with reduced odor identification. However, our study aimed to investigate the associations between odor identification and cognition in participants, who were not diagnosed with neurodegenerative diseases.

In a large-scale study, the olfactory system of almost 2000 people aged 5–99 years was investigated with an odor identification test. There was a general decrease in the ability to identify odors with aging which accelerated significantly in those over 70 years of age (Doty, Shaman, Applebaum, *et al.*, 1984). Increasing age, even without obvious medical problems, has since been associated with reduced odor identification in several studies (Ship *et al.*, 1996; Murphy *et al.*, 2002; Doty and Kamath, 2014). We have therefore adjusted the association between reduced odor identification and cognitive impairment for the influence of age.

In a study with older participants from the Rush Memory and Aging Project in the U.S.A., the association between olfactory function and decline in different brain areas was investigated. The olfactory identification of all participants was assessed at the baseline investigation and cognitive tests were carried out for up to 3 years. It was concluded that reduced odor identification was associated with impaired global perception and a faster decline in perceptual processing speed and episodic memory in the elderly (Wilson *et al.*, 2006). Despite similar results, it should be pointed out that their study period was not only half as long as ours, but that only 45 study participants were actually followed over the entire study period. In addition, 72.6% of the participants were women, while our study population consisted exclusively of women. A follow-up of this study showed that difficulties in identifying odors predicted subsequent development of MCI in the elderly without manifest cognitive impairment (Wilson, Schneider, *et al.*, 2007) which is also in line with our results. They even performed brain autopsies to investigate whether reduced odor identification in old age was related to the accumulation of AD pathologies. For this purpose, several brain areas were examined postmortem for β-amyloid plaques, density of neurofibrillary tangles and Lewy bodies. It was concluded that the difficulty in identifying common odors in old age was partly due to the accumulation of neurofibrillary pathologies in central odor regions (Wilson, Arnold, *et al.*, 2007). Subsequent research demonstrated that reduced odor identification was useful in predicting the transition from MCI to a clinical diagnosis of AD and its association with cognitive decline in the elderly without impaired cognitive abilities (Devanand, 2016). A meta-analysis aimed to evaluate the differences in olfactory function between patients with MCI and AD. The results indicated that olfactory function was susceptible to pathological changes in patients with MCI and AD and that reduced olfactory function was generally more severe in patients with AD than in patients with MCI. It was concluded that measurements of the olfactory function may be useful for the early detection of AD (Jung, Shin and Lee, 2019). Although we cannot provide any further information on this particular topic, these studies provide an outlook on the importance of the association between reduced odor identification and cognitive impairment which we have confirmed.

In a cohort study with non-demented participants, reduced odor identification and cognitive impairment, namely MCI and AD, were associated (Devanand *et al.*,

2010). Odor identification was particularly associated with cognitive subtests similar to our CERAD subtests, namely immediate and delayed recall, category fluency, and BNT. The association between reduced olfactory function and performance in the CERAD subtest BNT was previously shown in a comparative study between participants with MCI and those without cognitive impairment. Participants with MCI performed worse in all tests compared to the controls, but the performance in odor identification tests was less impaired than the performance in the CERAD subtest BNT (Laakso *et al.*, 2009). A longitudinal study carried out on healthy aging individuals came to similar conclusions (Wehling *et al.*, 2016). In addition, performance in the odor identification test proved to be a significant predictor for verbal episodic memory which we also tested with the CERAD subtest “Word list recall” and obtained similar results. In a large study population of elderly participants with reduced olfactory function but normal cognition at baseline, there was a strong association between the reduced ability to identify odors and the 5-year incidence of cognitive impairment measured using the MMSE (Schubert *et al.*, 2008). In our analysis, the performance in the MMSE test was also associated with reduced odor identification. A study comparing objective olfactory tests and self-reported olfactory dysfunctions showed a significant association between objectively measured olfactory dysfunction and cognitive impairment, assessed, among others, by the neuropsychological CERAD test. The self-reported olfactory dysfunction was also found to be an uncertain indicator (Choi *et al.*, 2018).

It should be pointed out that various tests were used to examine odor identification, such as the 40-item University of Pennsylvania Smell Identification Test (Doty, Shaman, Kimmelman, *et al.*, 1984), the Brief Smell Identification Test derived from it (Doty, Marcus and William Lee, 1996), or the San Diego Odor Identification Test (Murphy, Anderson and Markison, 1994; Murphy *et al.*, 2002). One of the first studies in this context to be carried out with Sniffin’ Sticks examined the usefulness of odor identification as an additional screening test in incipient AD (Quarmley *et al.*, 2017). The elaborate version of the Sniffin’ Sticks test was later used to validate olfactory dysfunction in AD patients (Yu *et al.*, 2018). In a recently published study among adults (18–79 years) in Germany, the cross-sectional association between olfactory and cognitive performance was analyzed using data of the population-based LIFE-Adult-Study (Yahiaoui-Doktor

et al., 2019). Similar to our study, Sniffin' Sticks were used to assess the olfactory function and the neuropsychological CERAD-Plus test to assess cognition, but in contrast to our study methods, only 12 Sniffin' Sticks and 5 CERAD subtests were used. Here, too, there was an association between olfactory and cognitive performance after adjustment for age, gender, level of education, and depressive symptoms. Participants who performed better in the Sniffin' Sticks test also performed better in all CERAD subtests. We were able to replicate and even extend these results. We presented the associations mentioned cross-sectionally. Furthermore, we not only collected more data for the assessment of the olfactory function and cognitive abilities, but also for possible covariates. Finally, we adjusted for multiple other important risk factors.

It should be emphasized that the studies discussed above examined both genders, while our study population was restricted to women. In general, women outperformed men in all of these studies. The underlying background for this is unknown, but the gender-specific differences appear to be undetectable in severe pathologies associated with AD (Devanand, 2016; Jung, Shin and Lee, 2019). More research is needed to shed light on this topic. Additionally, the growing field of gender-specific medicine could also contribute to a new understanding.

Odor identification tests are widely available, easy to use, and cost-effective, yet primarily used as research tools. Although reduced olfactory function has been shown repeatedly to predict cognitive decline in people with previously intact cognitive abilities and to be useful as an early biomarker for AD pathologies. In daily clinical practice, however, odor identification tests are usually only carried out taking into account the specific exclusion criteria and in combination with a thorough clinical, neuropsychological, and neuroimaging assessment. With further insight into the association between olfactory and cognitive performance, the clinical recommendations may also change (Devanand, 2016).

4.5 Air pollution and cognition

In our study, long-term air pollution exposure was associated with lower performance of multiple subtests of the CERAD-Plus test battery.

The association between air pollution and MCI was previously investigated in the SALIA cohort (Ranft *et al.*, 2009; Schikowski *et al.*, 2015). Results indicated that air pollution was associated with general cognition assessed with the CERAD total score, but also with effects on certain brain areas, namely those related to the construction performance tests which are indicators of visuo-spatial deficits.

Air pollution comprises numerous environmental toxins that can affect the brain through different pathways. A growing body of evidence suggests possible effects of ambient air pollution on brain health and CNS diseases, such as chronic neuroinflammation, oxidative stress, cerebrovascular damage, and neurodegenerative diseases (Peters *et al.*, 2006; Block *et al.*, 2012). In vitro studies were the first to point out these neurotoxic effects of urban air pollution (Block *et al.*, 2004; Hartz *et al.*, 2008; Davis *et al.*, 2013; Fagundes *et al.*, 2015). The conclusions have been supported by a large number of animal studies which also examined the effects of air pollution on the CNS (Guxens and Sunyer, 2012; Costa *et al.*, 2014). One of the first evidence of particles in the brain was detected in hamsters after intratracheal instillation of UFP labeled with radioactivity (Nemmar *et al.*, 2001). Mice exposed to concentrated airborne PM showed increased inflammation and neurodegeneration in the brain tissue compared to control animals (Campbell *et al.*, 2005; Veronesi *et al.*, 2005). After 4 weeks of inhalation of diesel exhaust gases, elevated pro-inflammatory markers were also detected in the brain of rats which indicated generalized neuroinflammation (Levesque *et al.*, 2011). In vitro and animal studies are crucial as basic research and contribute to a better understanding of the underlying mechanisms of the development of neurodegenerative diseases in humans. In contrast, they are very limited when it comes to the association between air pollution and complex cognitive processes in the human brain that play a role in cognitive tasks and are often used to screen for neurodegenerative diseases.

The role of environmental factors in the development of neurodegenerative diseases was also investigated in humans. One of the first studies in this context

examined the effects of exposure to diesel exhaust gases in the human brain using electroencephalographic signals. Increasing functional effects of brain activity have been shown in human volunteers exposed to dilute diesel exhaust for 1 hour, indicating a general cortical stress response (Crüts *et al.*, 2008). However, the power of this study was very limited due to its small sample size.

In a population-based cohort study, the incidence of dementia was associated with living near heavy traffic (Chen *et al.*, 2017). Further studies have contributed to this association between air pollution exposure and neurodegenerative diseases (Sagai and Tin Win-Shwe, 2015; Heusinkveld *et al.*, 2016; Shou *et al.*, 2019). In fact, postmortem sampling has identified PM in the human brain, albeit of unknown composition and origin. Living in Mexico City with exposure to high levels of air pollution was associated with a significant upregulation of pro-inflammatory markers in the olfactory bulb and frontal cortex, disorders of the BBB, and oxidative stress in children and young adults who died suddenly (Calderón-Garcidueñas *et al.*, 2008). In another study, brain autopsies from children and young adult residents in Mexico showed changes of gene expression associated with oxidative stress and other AD-associated pathology. It was concluded that air pollution may play a role in CNS damage, its effects on the developing brain, and the potential etiology of AD and mood disorders (Calderón-Garcidueñas *et al.*, 2012). It should be kept in mind that concentrations of air pollutants in Mexico City are extreme and it cannot be said with certainty whether such serious pathologies may ever occur in people who are only exposed to moderate levels of air pollution.

They also investigated whether air pollution exposure was associated with children's brain growth and cognition. Children exposed to high levels of air pollution showed differences in the supratentorial volumes of white matter compared to controls. The differences in brain volume were also consistent with cognitive deficits in highly exposed children, suggesting that exposure to air pollution interfered with children's brain development and cognition (Calderón-Garcidueñas *et al.*, 2011). This association between air pollution and cognition in children also confirmed the conclusions of an earlier cohort study. Higher BC levels have been shown to be associated with decreased cognitive function in verbal and nonverbal intelligence, and memory construction tests (Suglia *et al.*,

2007). Another study carried out in southern Spain investigated the effects of NO_2 and cognitive development in children. Although their results were not statistically significant, it was also suspected that traffic-related air pollution negatively affected children's cognitive abilities (Freire *et al.*, 2010). These results provided initial evidence of the association between air pollution and cognition. However, they are not transferable one-to-one to the effects of long-term air pollution exposure and cognitive changes in the elderly.

One of the first studies to investigate air pollution-induced cognitive changes in adults was carried out by Chen and Schwartz (2009). Exposure to annual ozone was associated with an age-related decline in cognitive function of adults. Another study with BC as a marker for traffic-related air pollution investigated its effect on the performance of cognitive tests over a 10-year study period. BC has been found to be associated with the MMSE score in older men (Power *et al.*, 2011). Further studies have contributed to this growing body of research by demonstrating the importance of air pollution for adult cognitive function (Tonne *et al.*, 2014; Ailshire and Clarke, 2015). Because of the complexity of cognition, the cognitive tests used in these studies vary widely, and air pollution markers were associated with different cognitive abilities as a result. In our study, PM_{10} was associated with the CERAD total score and the CERAD subtest "Word list learning". All air pollutant markers were also associated with the CERAD subtest of VCP. These findings are consistent with identified associations between exposure to air pollution and domain-specific cognitive abilities (Gatto *et al.*, 2014). A cohort study, very similar to the one we researched, also consisted only of older women and examined the association between PM and performance in cognitive tests. Long-term exposure to higher PM_{10} and $PM_{2.5}$ levels were significantly associated with poor performance in cognitive tests (Weuve, 2012). However, cognitive tests were partly administered using validated telephone interviews and cognitive function was assessed with a limited number of subtests, whereas assessment of cognition was much more detailed in our study.

The need for more research is obvious. People are getting older and the proportion of the elderly in our society is increasing. Therefore, it is important to understand the effects of toxic exposure over the lifespan and to provide evidence of the particular vulnerability of the elderly.

4.6 Mediation and interaction analysis

To our knowledge, this is the first study to provide epidemiological evidence that long-term air pollution exposure and reduced odor identification interact when it comes to influencing cognition in a cohort study of elderly women with a follow-up period of more than 20 years. The association between exposure to long-term air pollution and cognitive impairment changed depending on odor identification as a moderator variable. We carried out the mediation analysis although there was no association between the exposure (air pollution) and the mediator variable (odor identification) to answer the a priori hypothesis. Consequently, the mediation analysis indicated that this association between air pollution exposure and cognition was not mediated by a reduction in odor identification.

We analyzed the association between air pollution and odor identification at a single point in time and its influence on the performance of cognitive tests at 2-time points. Therefore, although we cannot account for decline in odor identification over time, we can provide information on how cognition changed over time under this influence and after adjustment for age, educational level, BMI, smoking status, living in an urban vs. rural area, cold, and hay fever. We hypothesized that transport of air pollutants into the brain along the olfactory nerve at time A is associated with reduced odor identification, while cognitive impairment is only expected at a later time B. Justification for performing the mediation analysis was missing because air pollution and odor identification were not associated. Nevertheless, the mediation analysis was carried out to answer the a priori hypothesis. In our interaction analysis, the results for an interaction between air pollution exposure and cognitive impairment varied depending on reduced odor identification at time A (2008/2009). However, the effect of air pollution exposure on cognitive impairment at time B (2012/2013) interacted significantly with the performance in the odor identification test. Our results suggest that a functional transport route from nose to brain appears to be important when it comes to MCI after exposure to air pollution.

Several biological pathways have been discussed of how air pollution exposure may be associated with cognition and further result in neurodegenerative diseases. Olfactory neurons are in direct contact with the external environment because they must be easily accessible if they are to sense odor molecules. A

hypothetical transport route for air pollutants to the brain tissue along the olfactory nerve appears to be possible from an anatomical point of view. However, it is believed that if this transport route was open to airborne pollutants, the sense of smell may be impaired before particles reach the brain (Arnold, 2019).

One of the first research teams to investigate air pollution, its effects on the brain, and a transport route associated with the olfactory system carried out studies in Mexico. They found various neuropathologies, such as alterations of the BBB, or degenerating cortical neurons, but also a deteriorated olfactory barrier in dogs from Mexico City that were exposed to high levels of air pollution (Calderón-Garcidueñas *et al.*, 2002). In a follow-up study in Mexico City dogs and less exposed controls, the genomic DNA from nose and brain was assessed, and the presence of metals in the olfactory mucosa, olfactory bulb, and frontal cortex was measured. The aim of this study was to investigate the association between urban air pollution and changes in the nose and brain tissue. In dogs from Mexico City, DNA damage was premature and the inflammatory and stress protein brain responses were early and significant. In addition, the investigated metals were present in a gradient from olfactory mucosa > olfactory bulb > frontal cortex (Calderón-Garcidueñas *et al.*, 2003). The association between air pollution and the acceleration of Alzheimer's-like neuropathologies in dogs may also be essential for a better understanding of neurodegenerative diseases in humans. Additionally, a transport route from nose to brain seemed promising. The research team also investigated whether living in a city with high levels of air pollution was associated with inflammation of the human brain. Severe air pollution was associated with significantly higher pro-inflammatory markers in different brain areas and in the olfactory bulb (Calderón-Garcidueñas *et al.*, 2004). However, it has to be kept in mind that results from animal studies are generally not transferable to humans one-to-one. They also focused on children and young adults from Mexico City and 2 less polluted cities. The brain-region-specific effects of metals, which originate from toxic combustion and industrial plants and are measured with $PM_{2.5}$, were examined in connection with neuropathological changes. The concentrations of metals associated with PM were higher in the frontal lobe and olfactory bulb of Mexico City residents. These results suggested that neurotoxicity was associated with PM metals in young city dwellers. In addition, the olfactory bulb was a target of air pollution and involved

in the neuroinflammatory response (Calderón-Garcidueñas *et al.*, 2013). Although conclusive, their results are difficult to apply to our study. Air pollution in Mexico City exceeds the levels of air pollution our study participants were exposed to in urban areas, even after a long period of time. Neuropathological and inflammatory processes in the brain are also hardly comparable to the performance of cognitive tests. Additionally, the number of humans investigated in their studies was rather small which limited the power of their results. While children and young adults are among the most vulnerable members of society, the older members of society examined in our study are also particularly at risk. Results from these different population groups are not interchangeable.

Other studies also indicated an association between air pollution, the olfactory system, and cognition. A study in animals investigated whether exposure to UFP was associated with deposition on the olfactory mucosa of the nasal region and transport along the olfactory nerve into the olfactory bulb. After different exposure times, the animals' lungs, cerebrum, cerebellum, and olfactory bulb were removed and examined. In the olfactory bulb, there was a significant and persistent increase in the concentrations of the UFP marker with increased exposure times (Oberdörster *et al.*, 2004). The effect of solid UFP after intranasal instillation on the olfactory bulb and hippocampus of mice was also examined. It was shown that pro-inflammatory markers were only induced in the olfactory bulb (Tin-Tin-Win-Shwe *et al.*, 2006). Other animal studies also contributed to correlate solid UFP inhalation and neurodegenerative pathologies with changes of the olfactory system (Colin-Barenque, Souza-Gallardo and Fortoul, 2011; Heusinkveld *et al.*, 2016). Although these animal studies are essential and support our hypothesis of direct nose-to-brain transport, it must be pointed out that findings taken from animal studies are not indicative of human pathologies.

Similar to the 2004 Mexican study, brain tissue from people exposed to different levels of air pollution was magnetically examined for biologically formed nanoparticles of the strongly magnetic mineral magnetite. These nanoparticles arise as combustion-derived, iron-rich particles which are often associated with other transition metals. The presence of magnetite nanoparticles was associated with age and level of air pollution, with the highest magnetite content found in the brain of a 32-year-old Mexico City resident. It was shown that smaller magnetite

pollutant particles may reach the brain directly via the olfactory bulb. Their presence indicated a direct route of transportation involving the olfactory system (Maher *et al.*, 2016). Although this study suggested a functional transport route from nose to brain, many other transport routes may also have been responsible for the presence of these particles in the brain tissue.

Exposure to different metals and their effects on neurobehavioral tests were investigated in a study with healthy children in Italy. They reported an association between Mn concentrations in the soil and anomalies of motor coordination and odor identification of study participants (Lucchini, Guazzetti, *et al.*, 2012). Their study sample was large and the exposure assessment was carried out very thoroughly. In addition, exposure to Mn was previously shown to be associated with an increased prevalence of PD in this study area (Lucchini *et al.*, 2007). Altogether, the results indicated the dangerous health effects of long-term increased Mn concentrations in the environment, supporting our hypothesis of an important role of odor identification for the association between environmental pollutants and cognition. However, no statements could be made regarding the chronological sequence. A study with teenagers who had spent their entire lives in areas contaminated with Mn showed that long-term exposure to Mn affected attention, neuromotor and olfactory functions. The olfactory function was assessed using the Sniffin' Sticks test and brain imaging was performed while sniffing to show the olfactory psychophysical response in the olfactory regions of the brain (Iannilli *et al.*, 2016). However, it is important to keep in mind that this was a pilot study with very few study participants. The results are generally consistent with our hypothesis, but the large differences between Mn and the air pollutants we examined should be emphasized. Additionally, study results with children are not applicable to our study participants of elderly women.

Previous studies of the SALIA cohort were among the first to link traffic-related air pollution exposure with reduced odor identification and impaired cognitive function in the elderly (Ranft *et al.*, 2009). 399 participants of the SALIA cohort met the inclusion criteria. At this point in time, exposure to background pollution in the ambient air was estimated by the pollutant concentrations, averaged over the 5 years prior to the baseline study and over the 5 years from 2002–2006, and calculated using GIS. Distance of the home address to the next busy street with

a traffic density of more than 10000 cars was also used as a marker for air pollution. The results indicated that distance to traffic was a significant and consistent risk factor for odor identification and MCI for participants under 74 years of age. However, there were no adverse effects of traffic exposure on cognition in women over 74 years of age. The effect of living near a busy street was equally pronounced in urban and rural areas. We were able to confirm these results and add further insights into the association between air pollution exposure, odor identification, and cognition even though we did not use “distance of residential address to the next busy street” as a marker for air pollution in our study, but PM_{10}, $PM_{2.5}$, $PM_{2.5}$abs, and NO_2. For the assessment of air pollution exposure, we carried out LUR models with data of a measurement campaign in 2008/2009. Despite the differences, our results may contribute to ideas regarding the chronological sequence. The association between exposure to long-term air pollution and cognitive impairment changed depending on odor identification and this effect was only significant in 2012/2013. In addition, cognitive impairment was also associated with air pollution in participants over 74 years of age.

In summary, the transport of ambient air pollutants along the olfactory nerve has been demonstrated in several studies and the association between air pollution exposure, odor identification, and neuropathology is supported by various findings. As far as we know, our results are the first to indicate that exposure to air pollution and odor identification interact when it comes to influencing cognitive abilities. However, further research is needed to better understand the underlying mechanisms and pathological pathways of these associations. Long-term studies in larger population samples are particularly needed to corroborate our results.

4.7 Conclusion

The goal of this study was to investigate the associations between long-term air pollution, odor identification, and cognition. Our epidemiological study is among the first to highlight the fact that the association between long-term air pollution exposure and cognitive impairment changed depending on odor identification as a moderator variable in a population-based cohort of elderly women who were followed for more than 20 years.

The exact way in which air pollutants damage the organism remains an important topic for further research. Clean air is a fundamental right of everyone. Mindless, short-lived, or small-scale measures will not help improve the quality of the air we breathe today. Our heads of states and government must acknowledge air pollution as one of the most dangerous threats to public health and agree on common measures to protect people. There should be a focus on how air pollution is measured to ensure that study results are more comparable. It is crucial to further investigate possible transport routes in order to better understand how and why air pollution is associated with various health effects. More research is also needed to investigate the possible association between air pollution and the olfactory system, and its role as a possible transport route for air pollutants deserve further attention. Research should particularly focus on air pollution-induced cognitive impairment. Uniform definitions and appropriate measurement tools are necessary to assess normal vs. accelerated brain aging. The incidence of neurological diseases is increasing in aging societies, which is why progress in diagnostic and therapeutic options, early detection and prevention are of great importance. Olfactory loss as an early symptom of AD needs further research, but may contribute to the early diagnosis of the disease in the future. Symptoms and diseases of patients need to be considered diligently and critically in the context of their environment. Therefore, doctors are not only required to expand their scientific knowledge in a rapidly developing field. They should also take a leadership role in this research field, educate patients, promote and advocate actions, and call for wide-ranging strategies that need to be taken to reduce ambient air pollution and its consequences.

5 References

Ackermann-Liebrich, U. *et al.* (1997) “Lung function and long term exposure to air pollutants in Switzerland,” *American Journal of Respiratory and Critical Care Medicine*. American Thoracic Society, 155(1), pp. 122–129. doi: 10.1164/ajrccm.155.1.9001300.

Adam, M. *et al.* (2015) “Adult lung function and long-term air pollution exposure. ESCAPE: a multicentre cohort study and meta-analysis,” *European Respiratory Journal*, 45(1), pp. 38–50. doi: 10.1183/09031936.00130014.

Adams, D. R. *et al.* (2016) “Nitrogen dioxide pollution exposure is associated with olfactory dysfunction in older U.S. adults,” *International Forum of Allergy and Rhinology*. John Wiley and Sons Inc., 6(12), pp. 1245–1252. doi: 10.1002/alr.21829.

Aebi, C. (2003) “Validierung der neuropsychologischen Testbatterie CERAD-NP : Eine Multi-Center Studie,” *Dissertation*. doi: 10.5451/unibas-002728525.

Ahlström, R. *et al.* (1987) “A comparison of odor perception in smokers, nonsmokers, and passive smokers,” *American Journal of Otolaryngology*, 8(1), pp. 1–6. doi: 10.1016/S0196-0709(87)80011-X.

Aiken, L. S. and West, S. G. (1986) “Use and interpretation of regression analysis models containing interactions and power polynomials,” *Unpublished manuscript, Arizona State University, Tempe*.

Ailshire, J. A. and Clarke, P. (2015) “Fine Particulate Matter Air Pollution and Cognitive Function Among U.S. Older Adults,” *The Journals of Gerontology Series B: Psychological Sciences and Social Sciences*, 70(2), pp. 322–328. doi: 10.1093/geronb/gbu064.

Ailshire, J. A. and Crimmins, E. M. (2014) “Fine Particulate Matter Air Pollution and Cognitive Function Among Older US Adults,” *American Journal of Epidemiology*, 180(4), pp. 359–366. doi: 10.1093/aje/kwu155.

Ajmani, G. S. *et al.* (2016) “Fine particulate matter exposure and olfactory dysfunction among urban-dwelling older US adults,” *Environmental Research*, 151, pp. 797–803. doi: 10.1016/j.envres.2016.09.012.

Ajmani, G. S. *et al.* (2017) “Smoking and olfactory dysfunction: A systematic literature review and meta-analysis,” *Laryngoscope*. John Wiley and Sons Inc., 127(8), pp. 1753–1761. doi: 10.1002/lary.26558.

Ajmani, G. S., Suh, H. H. and Pinto, J. M. (2016) “Effects of ambient air pollution exposure on olfaction: A review,” *Environmental Health Perspectives*. Public Health Services, US Dept of Health and Human Services, pp. 1683–1693. doi: 10.1289/EHP136.

Allred, E. N. *et al.* (1989) “Short-Term Effects of Carbon Monoxide Exposure on the Exercise Performance of Subjects with Coronary Artery Disease,” *New England Journal of Medicine*, 321(21), pp. 1426–1432. doi: 10.1056/NEJM198911233212102.

Alzheimer, A. (1906) “Über den eigenartigen schweren Krankheitsprozess der Hirnrinde,” *Neurologisches Centralblatt*, 25, p. 1134.

Analitis, A. *et al.* (2006) “Short-term effects of ambient particles on cardiovascular and respiratory mortality.,” *Epidemiology (Cambridge, Mass.)*, 17(2), pp. 230–233. doi: 10.1097/01.ede.0000199439.57655.6b.

Arnold, C. (2019) “Sensory overload? Air pollution and impaired olfaction,” *Environmental Health Perspectives*. Public Health Services, US Dept of Health and Human Services. doi: 10.1289/EHP3621.

Atkinson, R. W. *et al.* (1999) “Short-term associations between outdoor air pollution and visits to accident and emergency departments in London for respiratory complaints,” *European Respiratory Journal*, 13(2), pp. 257–265. doi: 10.1183/09031936.99.13225799.

Aumüller, G. *et al.* (2014) “Duale Reihe Anatomie, Thieme,” *Edited by G. Aumüller et al. Stuttgart: Georg Thieme Verlag.* doi: 10.1055/b-002-99154.

Baron, R. M. and Kenny, D. A. (1986) “The Moderator-Mediator Variable Distinction in Social The Moderator-Mediator Variable Distinction in Social Psychological Research: Conceptual, Strategic, and Statistical Considerations,” *Journal of Personality and Social Psychology*, 51(6), pp. 1173–1182. doi: 10.1037/0022-3514.51.6.1173.

Beelen, R. *et al.* (2008) “Long-term effects of traffic-related air pollution on mortality in a Dutch cohort (NLCS-AIR study),” *Environmental Health Perspectives*, 116(2), pp. 196–202. doi: 10.1289/ehp.10767.

Beelen, R. *et al.* (2013) “Development of NO2 and NOx land use regression models for estimating air pollution exposure in 36 study areas in Europe – The ESCAPE project,” *Atmospheric Environment*, 72(September 2016), pp. 10–23. doi: 10.1016/j.atmosenv.2013.02.037.

Berres, M. *et al.* (2000) “Normal ranges of neuropsychological tests for the diagnosis of Alzheimer’s disease,” *Studies in health technology and informatics*. IOS Press, 77, pp. 195–199. doi: 10.3233/978-1-60750-921-9-195.

Block, M. L. *et al.* (2004) “Nanometer size diesel exhaust particles are selectively toxic to dopaminergic neurons: the role of microglia, phagocytosis, and NADPH oxidase,” *FASEB Journal : official publication of the Federation of American Societies for Experimental Biology*, 18(13), pp. 1618–1620. doi: 10.1096/fj.04-1945fje.

Block, M. L. *et al.* (2012) “The outdoor air pollution and brain health workshop,” *NeuroToxicology*, pp. 972–984. doi: 10.1016/j.neuro.2012.08.014.

Block, M. L. and Calderón-Garcidueñas, L. (2009) “Air pollution: mechanisms of neuroinflammation and CNS disease,” *Trends in Neurosciences*, 32(9), pp. 506–516. doi: 10.1016/j.tins.2009.05.009.

Bock, O., Haeger, M. and Voelcker-Rehage, C. (2019) “Structure of executive functions in young and in older persons,” *PLoS ONE*. Public Library of Science, 14(5). doi: 10.1371/journal.pone.0216149.

Bokina, A. I. *et al.* (1976) “Investigation of the mechanism of action of atmospheric pollutants on the central nervous system and comparative evaluation of methods of study,” *Environmental Health Perspectives*, Vol. 13, pp. 37–42. doi: 10.2307/3428235.

Brämerson, A. *et al.* (2004) “Prevalence of Olfactory Dysfunction: The Skövde Population-Based Study,” *Laryngoscope*, 114(4), pp. 733–737. doi: 10.1097/00005537-200404000-00026.

Braun-Fahrländer, C. *et al.* (1997) “Respiratory health and long-term exposure to air pollutants in Swiss schoolchildren,” *American Journal of Respiratory and Critical Care Medicine*. American Thoracic Society, 155(3), pp. 1042–1049. doi: 10.1164/ajrccm.155.3.9116984.

Briet, M. *et al.* (2007) “Endothelial function and chronic exposure to air pollution in normal male subjects,” *Hypertension*, 50(5), pp. 970–976. doi: 10.1161/HYPERTENSIONAHA.107.095844.

Brook, R. D. *et al.* (2002) “Inhalation of Fine Particulate Air Pollution and Ozone Causes Acute Arterial Vasoconstriction in Healthy Adults,” *Circulation*, 105(13), pp. 1534–1536. doi: 10.1161/01.CIR.0000013838.94747.64.

Brook, R. D. *et al.* (2004) “Air pollution and cardiovascular disease: A statement for

healthcare professionals from the expert panel on population and prevention science of the American Heart Association," *Circulation*, pp. 2655–2671. doi: 10.1161/01.CIR.0000128587.30041.C8.

Brook, R. D. (2008) "Cardiovascular effects of air pollution," *Clinical Science*, 115(6), pp. 175–187. doi: 10.1042/CS20070444.

Brook, R. D. *et al.* (2010) "Particulate Matter Air Pollution and Cardiovascular Disease: An Update to the Scientific Statement From the American Heart Association," *Circulation*, 121(21), pp. 2331–2378. doi: 10.1161/CIR.0b013e3181dbece1.

Brook, R. D., Brook, J. R. and Rajagopalan, S. (2003) "Air pollution: The 'heart' of the problem," *Current Hypertension Reports*, 5(1), pp. 32–39. doi: 10.1007/s11906-003-0008-y.

Brunekreef, B. (2007) "Health effects of air pollution observed in cohort studies in Europe," *Journal of Exposure Science & Environmental Epidemiology*, 17(SUPPL. 2), pp. 61–65. doi: 10.1038/sj.jes.7500628.

BPB (Bundeszentrale für politische Bildung) (2019) "Demografischer Wandel," Available at: https://www.bpb.de/politik/innenpolitik/demografischer-wandel/ (Accessed: February 4, 2020).

Burghart Messtechnik GmbH (2006) "Burghart Instruments (®)," *Wedel, Germany*. Available at: http://www.burghart-mt.de/de/medizintechnik/sniffin-sticks-taste-strips.html (Accessed: February 23, 2020).

Burns, A. and Zaudig, M. (2002) "Mild cognitive impairment in older people," *Lancet*. Elsevier Limited, pp. 1963–1965. doi: 10.1016/S0140-6736(02)11920-9.

Calderón-Garcidueñas, L. *et al.* (1996) "DNA strand breaks in human nasal respiratory epithelium are induced upon exposure to urban pollution," *Environmental Health Perspectives*. Public Health Services, US Dept of Health and Human Services, 104(2), pp. 160–168. doi: 10.1289/ehp.96104160.

Calderón-Garcidueñas, L. *et al.* (2002) "Air pollution and brain damage," *Toxicologic Pathology*, 30(3), pp. 373–389. Available at: http://www.ncbi.nlm.nih.gov/pubmed/12051555.

Calderón-Garcidueñas, L. *et al.* (2003) "DNA Damage in Nasal and Brain Tissues of Canines Exposed to Air Pollutants Is Associated with Evidence of Chronic Brain Inflammation and Neurodegeneration," *Toxicologic Pathology*, 31(5), pp. 524–538. doi: 10.1080/01926230390226645.

Calderón-Garcidueñas, L. *et al.* (2004) "Brain Inflammation and Alzheimer's-Like Pathology in Individuals Exposed to Severe Air Pollution," *Toxicologic Pathology*, 32(6), pp. 650–658. doi: 10.1080/01926230490520232.

Calderón-Garcidueñas, L. *et al.* (2008) "Long-term Air Pollution Exposure Is Associated with Neuroinflammation, an Altered Innate Immune Response, Disruption of the Blood-Brain Barrier, Ultrafine Particulate Deposition, and Accumulation of Amyloid β-42 and α-Synuclein in Children and Young Adult," *Toxicologic Pathology*, 36(2), pp. 289–310. doi: 10.1177/0192623307313011.

Calderón-Garcidueñas, L. *et al.* (2010) "Urban air pollution: influences on olfactory function and pathology in exposed children and young adults," *Experimental and toxicologic pathology*, 62(1), pp. 91–102. doi: 10.1016/j.etp.2009.02.117.

Calderón-Garcidueñas, L. *et al.* (2011) "Exposure to severe urban air pollution influences cognitive outcomes, brain volume and systemic inflammation in clinically healthy children," *Brain and Cognition*. Academic Press Inc., 77(3), pp. 345–355. doi: 10.1016/j.bandc.2011.09.006.

Calderón-Garcidueñas, L. *et al.* (2012) "Neuroinflammation, Hyperphosphorylated Tau,

Diffuse Amyloid Plaques, and Down-Regulation of the Cellular Prion Protein in Air Pollution Exposed Children and Young Adults," *Journal of Alzheimer's Disease*, 28(1), pp. 93–107. doi: 10.3233/JAD-2011-110722.

Calderón-Garcidueñas, L. *et al.* (2013) "The impact of environmental metals in young urbanites' brains," *Experimental and Toxicologic Pathology*, 65(5), pp. 503–511. doi: 10.1016/j.etp.2012.02.006.

Campbell, A. *et al.* (2005) "Particulate Matter in Polluted Air May Increase Biomarkers of Inflammation in Mouse Brain," *NeuroToxicology*, 26(1), pp. 133–140. doi: 10.1016/j.neuro.2004.08.003.

Campbell, A. *et al.* (2009) "Particulate Matter Induced Enhancement of Inflammatory Markers in the Brains of Apolipoprotein E Knockout Mice," *Journal of Nanoscience and Nanotechnology*, 9(8), pp. 5099–5104. doi: 10.1166/jnn.2009.GR07.

Capitani, E. *et al.* (1988) "Perceptual attention in aging and dementia measured by Gottschaldt's Hidden Figure Test," *Journals of Gerontology*, 43(6). doi: 10.1093/geronj/43.6.P157.

Chandler, M. J. *et al.* (2005) "A total score for the CERAD neuropsychological battery," *Neurology*, 65(1), pp. 102–106. doi: 10.1212/01.wnl.0000167607.63000.38.

Chen, G. *et al.* (2015) "Traffic-related air pollution and lung cancer: A meta-analysis," *Thoracic Cancer*. John Wiley and Sons Inc., 6(3), pp. 307–318. doi: 10.1111/1759-7714.12185.

Chen, H. *et al.* (2017) "Living near major roads and the incidence of dementia, Parkinson's disease, and multiple sclerosis: a population-based cohort study," *The Lancet*. Elsevier, 389(10070), pp. 718–726. doi: 10.1016/S0140-6736(16)32399-6.

Chen, J.-C. and Schwartz, J. (2009) "Neurobehavioral effects of ambient air pollution on cognitive performance in US adults," *NeuroToxicology*, 30(2), pp. 231–239. doi: 10.1016/j.neuro.2008.12.011.

Chen, J. C. (2010) "Editorial: Geographic determinants of stroke mortality: Role of ambient air pollution," *Stroke*, pp. 839–841. doi: 10.1161/STROKEAHA.110.578476.

Choi, J. S. *et al.* (2018) "Olfactory dysfunction and cognition among older adults in the United States," *International Forum of Allergy and Rhinology*. John Wiley and Sons Inc., 8(5), pp. 648–654. doi: 10.1002/alr.22078.

Cohen, A. J. *et al.* (2004) "Urban air pollution," *Edited by Ezzati, M. et al. Comparative Quantification of Health Risks: Global and regional burden of disease attributable to selected risk factors*, pp. 1377–1458. ISBN: 9241580313.

Colin-Barenque, L., Souza-Gallardo, L. M. and Fortoul, T. I. (2011) "Toxic effects of inhaled manganese on the olfactory bulb: An ultrastructural approach in mice," *Journal of Electron Microscopy*, 60(1), pp. 73–78. doi: 10.1093/jmicro/dfq073.

Costa, L. G. *et al.* (2014) "Neurotoxicants Are in the Air: Convergence of Human, Animal, and In Vitro Studies on the Effects of Air Pollution on the Brain," *BioMed Research International*, 2014, pp. 1–8. doi: 10.1155/2014/736385.

Craik, F. I. M., Luo, L. and Sakuta, Y. (2010) "Effects of Aging and Divided Attention on Memory for Items and Their Contexts," *Psychology and Aging*. American Psychological Association Inc., 25(4), pp. 968–979. doi: 10.1037/a0020276.

Craik, F. I. M. and Rose, N. S. (2012) "Memory encoding and aging: A neurocognitive perspective," *Neuroscience and Biobehavioral Reviews*, pp. 1729–1739. doi: 10.1016/j.neubiorev.2011.11.007.

Creasey, H. and Rapoport, S. I. (1985) "The aging human brain," *Annals of Neurology*, 17(1), pp. 2–10. doi: 10.1002/ana.410170103.

Crucian, G. P. and Okun, M. S. (2003) “Visual spatial ability in Parkinson’s disease,” *Frontiers in Bioscience*, 8(6), p. 1171. doi: 10.2741/1171.

Crüts, B. *et al.* (2008) “Exposure to diesel exhaust induces changes in EEG in human volunteers,” *Particle and Fibre Toxicology*, 5(1), pp. 1–4. doi: 10.1186/1743-8977-5-4.

Cullen, M. M. and Leopold, D. A. (1999) “Disorders of smell and taste,” *Medical Clinics of North America*, 83(1), pp. 57–74. doi: 10.1016/S0025-7125(05)70087-0.

D’Mello, C., Le, T. and Swain, M. G. (2009) “Cerebral microglia recruit monocytes into the brain in response to tumor necrosis factora signaling during peripheral organ inflammation,” *Journal of Neuroscience*, 29(7), pp. 2089–2102. doi: 10.1523/JNEUROSCI.3567-08.2009.

Davis, D. a. *et al.* (2013) “Urban air pollutants reduce synaptic function of CA1 neurons via an NMDA/NȮ pathway in vitro,” *Journal of Neurochemistry*, 127(4), pp. 509–519. doi: 10.1111/jnc.12395.

Delacourte, A. *et al.* (1999) “The biochemical pathway of neurofibrillary degeneration in aging and Alzheimer’s disease,” *American Academy of Neurology*, pp. 1158–1165. doi: 10.1212/wnl.52.6.1158

Delfino, R. J. *et al.* (2009) “Air pollution exposures and circulating biomarkers of effect in a susceptible population: Clues to potential causal component mixtures and mechanisms,” *Environmental Health Perspectives*, 117(8), pp. 1232–1238. doi: 10.1289/journla.ehp.0800194.

Devanand, D. P. *et al.* (2000) “Olfactory deficits in patients with mild cognitive impairment predict Alzheimer’s disease at follow-up,” *American Journal of Psychiatry*, 157(9), pp. 1399–1405. doi: 10.1176/appi.ajp.157.9.1399.

Devanand, D. P. *et al.* (2010) “Olfactory identification deficits and MCI in a multi-ethnic elderly community sample,” *Neurobiology of Aging*, 31(9), pp. 1593–1600. doi: 10.1016/j.neurobiolaging.2008.09.008.

Devanand, D. P. *et al.* (2015) “Olfactory identification deficits and increased mortality in the community,” *Annals of Neurology*, 78(3), pp. 401–411. doi: 10.1002/ana.24447.

Devanand, D. P. (2016) “Olfactory Identification Deficits, Cognitive Decline, and Dementia in Older Adults,” *American Journal of Geriatric Psychiatry*. Elsevier B.V., 24(12), pp. 1151–1157. doi: 10.1016/j.jagp.2016.08.010.

DIMDI (Deutsches Institut für Medizinische Dokumentation und Information) (2020) “ICD-10-GM Version 2020,” Available at: https://www.dimdi.de/static/de/klassifikationen/icd/icd-10-gm/kode-suche/htmlgm2020/block-f00-f09.htm (Accessed: January 13, 2020).

Dockery, D. W. *et al.* (1993) “An Association between Air Pollution and Mortality in Six U.S. Cities,” *New England Journal of Medicine*, 329(24), pp. 1753–1759. doi: 10.1056/NEJM199312093292401.

Dominici, F. *et al.* (2003) “National maps of the effects of particulate matter on mortality: exploring geographical variation,” *Environmental health perspectives*, 111(1), pp. 39–44. doi: 10.1289/ehp.5181.

Donaldson, K. *et al.* (2003) “Oxidative stress and calcium signaling in the adverse effects of environmental particles (PM10),” *Free Radical Biology and Medicine*, 34(11), pp. 1369–1382. doi: 10.1016/S0891-5849(03)00150-3.

Doty, R. (2009) “The Olfactory System and Its Disorders,” *Seminars in Neurology*, 29(01), pp. 74–81. doi: 10.1055/s-0028-1124025.

Doty, R. L., Shaman, P., Applebaum, S. L., *et al.* (1984) “Smell identification ability: Changes with age,” *Science*, 226(4681), pp. 1441–1443. doi: 10.1126/science.6505700.

Doty, R. L., Shaman, P., Kimmelman, C. P., *et al.* (1984) “University of pennsylvania smell identification test: A rapid quantitative olfactory function test for the clinic,” *Laryngoscope*, 94(2), pp. 176–178. doi: 10.1288/00005537-198402000-00004.

Doty, R. L. *et al.* (1991) “Olfactory dysfunction in three neurodegenerative diseases,” *Geriatrics*, 46(SUPPL. 1), pp. 47–51. doi: 10.29333/ejgm/82202.

Doty, R. L. *et al.* (1995) “A Study of the Test-retest Reliability of Ten Olfactory Tests,” *Chemical Senses*, 20(6), pp. 645–656. doi: 10.1093/chemse/20.6.645.

Doty, R. L. (2006) “Olfactory dysfunction and its measurement in the clinic and workplace,” *International Archives of Occupational and Environmental Health*, 79(4), pp. 268–282. doi: 10.1007/s00420-005-0055-6.

Doty, R. L. (2012) “Olfactory dysfunction in Parkinson disease,” *Nature Reviews Neurology*. Nature Publishing Group, 8(6), pp. 329–339. doi: 10.1038/nrneurol.2012.80.

Doty, R. L., Deems, D. A. and Stellar, S. (1988) “Olfactory dysfunction in parkinsonism: A general deficit unrelated to neurologic signs, disease stage, or disease duration,” *Neurology*, 38(8), pp. 1237–1237. doi: 10.1212/WNL.38.8.1237.

Doty, R. L. and Kamath, V. (2014) “The influences of age on olfaction: A review,” *Frontiers in Psychology*, p. 20. doi: 10.3389/fpsyg.2014.00020.

Doty, R. L., Marcus, A. and William Lee, W. (1996) “Development of the 12-item cross-cultural smell identification test(cc-sit),” *Laryngoscope*, 106(3), pp. 353–356. doi: 10.1097/00005537-199603000-00021.

Dubois, B. and Albert, M. L. (2004) “Amnestic MCI or prodromal Alzheimer’s disease?,” *Lancet Neurology*, pp. 246–248. doi: 10.1016/S1474-4422(04)00710-0.

Edenhofer, O. *et al.* (2014) “Climate Change 2014: Mitigation of Climate Change. Contribution of Working Group III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change,” *Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.* Available at: https://www.ipcc.ch/site/assets/uploads/2018/02/ipcc_wg3_ar5_full.pdf#page=1&zoom=auto,-127,792 (Accessed: February 4, 2020).

Edwards, R. *et al.* (2006) “Does living near heavy industry cause lung cancer in women? A case-control study using life grid interviews,” *Thorax*, 61(12), pp. 1076–1082. doi: 10.1136/thx.2005.057620.

Eeftens, M. *et al.* (2012) “Development of Land Use Regression Models for PM 2.5, PM 2.5 Absorbance, PM 10 and PM coarse in 20 European Study Areas; Results of the ESCAPE Project,” *Environmental Science & Technology*, 46(20), pp. 11195–11205. doi: 10.1021/es301948k.

Elder, A. *et al.* (2006) “Translocation of Inhaled Ultrafine Manganese Oxide Particles to the CentralNervous System,” *Environmental Health Perspectives*, 114(8), pp. 1172–1178. doi: 10.1289/ehp.9030.

Erzigkeit, H. *et al.* (2001) “The Bayer-Activities of Daily Living Scale (B-ADL): Results from a Validation Study in Three European Countries,” *Dementia and Geriatric Cognitive Disorders*, 12(5), pp. 348–358. doi: 10.1159/000051280.

European Environment Agency (2019) “Air quality in Europe 2019,” *Copenhagen.* Available at: https://www.eea.europa.eu//publications/air-quality-in-europe-2019 (Accessed: July 1, 2020).

European Environment Agency (2020) “Sources of air pollution in Europe,” Available at: https://www.eea.europa.eu/signals/signals-2013/infographics/sources-of-air-pollution-in-europe/view (Accessed: June 14, 2020).

Fagundes, L. S. *et al.* (2015) “Direct contact with particulate matter increases oxidative

stress in different brain structures," *Inhalation Toxicology*, 27(10), pp. 462–467. doi: 10.3109/08958378.2015.1060278.

Feron, V. J. *et al.* (2001) "Health Risks Associated with Inhaled Nasal Toxicants," *Critical Reviews in Toxicology*, 31(3), pp. 313–347. doi: 10.1080/20014091111712.

Fillenbaum, G. G. *et al.* (2008) "Consortium to Establish a Registry for Alzheimer's Disease (CERAD): The first twenty years," *Alzheimer's & Dementia*, 4(2), p. ALZJJALZ200708005. doi: 10.1016/j.jalz.2007.08.005.

Freire, C. *et al.* (2010) "Association of traffic-related air pollution with cognitive development in children," *Journal of Epidemiology & Community Health*, 64(3), pp. 223–228. doi: 10.1136/jech.2008.084574.

Frye, R. E., Schwartz, B. S. and Doty, R. L. (1990) "Dose-Related Effects of Cigarette Smoking on Olfactory Function," *JAMA: The Journal of the American Medical Association*. American Medical Association, 263(9), pp. 1233–1236. doi: 10.1001/jama.1990.03440090067028.

Gakidou, E. *et al.* (2017) "Global, regional, and national comparative risk assessment of 84 behavioural, environmental and occupational, and metabolic risks or clusters of risks, 1990-2016: A systematic analysis for the Global Burden of Disease Study 2016," *The Lancet*. Lancet Publishing Group, 390(10100), pp. 1345–1422. doi: 10.1016/S0140-6736(17)32366-8.

Gatto, N. M. *et al.* (2014) "Components of air pollution and cognitive function in middle-aged and older adults in Los Angeles," *NeuroToxicology*, 40, pp. 1–7. doi: 10.1016/j.neuro.2013.09.004.

Gauderman, W. J. *et al.* (2005) "Childhood asthma and exposure to traffic and nitrogen dioxide," *Epidemiology*, 16(6), pp. 737–743. doi: 10.1097/01.ede.0000181308.51440.75.

Gehring, U. *et al.* (2006) "Long-term exposure to ambient air pollution and cardiopulmonary mortality in women," *Epidemiology*, 17(5), pp. 545–551. doi: 10.1097/01.ede.0000224541.38258.87.

Genter, M. B. and Doty, R. L. (2019) "Toxic exposures and the senses of taste and smell," *Handbook of Clinical Neurology*. Elsevier B.V., pp. 389–408. doi: 10.1016/B978-0-444-63855-7.00022-8.

Gobba, F. (2006) "Olfactory toxicity: long-term effects of occupational exposures," *International Archives of Occupational and Environmental Health*, 79(4), pp. 322–331. doi: 10.1007/s00420-005-0043-x.

Götschi, T. *et al.* (2008) "Long-Term Effects of Ambient Air Pollution on Lung Function," *Epidemiology*, 19(5), pp. 690–701. doi: 10.1097/EDE.0b013e318181650f.

Guarneros, M. *et al.* (2009) "Mexico city air pollution adversely affects olfactory function and intranasal trigeminal sensitivity," *Chemical Senses*. Oxford Academic, 34(9), pp. 819–826. doi: 10.1093/chemse/bjp071.

Guxens, M. and Sunyer, J. (2012) "A review of epidemiological studies on neuropsychological effects of air pollution," *Swiss medical weekly*, 141(January), p. w13322. doi: 10.4414/smw.2011.13322.

Halperin, W. E. *et al.* (1983) "Nasal cancer in a worker exposed to formaldehyde," *JAMA*, 249(4), pp. 510–602. Available at: http://www.ncbi.nlm.nih.gov/pubmed/6848853 (Accessed: February 14, 2020).

Halpern, B. P. (1982) "Environmental factors affecting chemoreceptors: an overview," *Environmental Health Perspectives*, 44, pp. 101–105. doi: 10.1289/ehp.8244101.

Harada, C. N., Natelson Love, M. C. and Triebel, K. L. (2013) "Normal cognitive aging," *Clinics in Geriatric Medicine*, pp. 737–752. doi: 10.1016/j.cger.2013.07.002.

Hartz, A. M. S. *et al.* (2008) “Diesel exhaust particles induce oxidative stress, proinflammatory signaling, and P-glycoprotein up-regulation at the blood-brain barrier,” *The FASEB Journal*, 22(8), pp. 2723–2733. doi: 10.1096/fj.08-106997.

Hautzinger, M. *et al.* (2012) “Allgemeine Depressionsskala (ADS),” *Hogrefe Verlag*. Available at: https://www.testzentrale.de/shop/allgemeine-depressionsskala.html (Accessed: January 14, 2020).

Hawkes, C. (2006) “Olfaction in Neurodegenerative Disorder,” in *Taste and Smell*. Basel: KARGER, pp. 133–151. doi: 10.1159/000093759.

Hayden, K. M. and Welsh-Bohmer, K. A. (2011) “Epidemiology of cognitive aging and alzheimer’s disease: Contributions of the cache county utah study of memory, health and aging,” *Current Topics in Behavioral Neurosciences*. Springer Verlag, 10, pp. 3–31. doi: 10.1007/7854_2011_152.

Hedley, A. J. *et al.* (2002) “Cardiorespiratory and all-cause mortality after restrictions on sulphur content of fuel in Hong Kong: An intervention study,” *Lancet*. Elsevier Limited, 360(9346), pp. 1646–1652. doi: 10.1016/S0140-6736(02)11612-6.

Herbert, R. A. *et al.* (1996) “Two-Year and Lifetime Toxicity and Carcinogenicity Studies of Ozone in B6C3F1 Mice,” *Toxicologic Pathology*. Sage PublicationsSage CA: Thousand Oaks, CA, 24(5), pp. 539–548. doi: 10.1177/019262339602400502.

Heusinkveld, H. J. *et al.* (2016) “Neurodegenerative and neurological disorders by small inhaled particles,” *NeuroToxicology*. Elsevier, 56, pp. 94–106. doi: 10.1016/j.neuro.2016.07.007.

Hildebrandt, K. *et al.* (2009) “Short-term effects of air pollution: a panel study of blood markers in patients with chronic pulmonary disease,” *Particle and Fibre Toxicology*, 6(1), pp. 1–25. doi: 10.1186/1743-8977-6-25.

Hödl, A. K., Bonelli, R. M. and Kapfhammer, H. P. (2005) “Mild cognitive impairment: A review of the literature,” *MMW Fortschritte der Medizin*, 147(23), pp. 40–43. Available at: http://www.ncbi.nlm.nih.gov/pubmed/15981904 (Accessed: February 6, 2020).

Hoffmann, B. *et al.* (2007) “Residential exposure to traffic is associated with coronary atherosclerosis,” *Circulation*, 116(5), pp. 489–496. doi: 10.1161/CIRCULATIONAHA.107.693622.

Holt, G. R. (1996) “Effects of air pollution on the upper aerodigestive tract,” in *Otolaryngology - Head and Neck Surgery*, pp. 201–204. doi: 10.1016/S0194-5998(96)70165-1.

Horn, J. L. and Cattell, R. B. (1967) “Age differences in fluid and crystallized intelligence,” *Acta Psychologica*, 26(C), pp. 107–129. doi: 10.1016/0001-6918(67)90011-x.

Horn, R. *et al.* (1996) “Atrophy of Hippocampus in patients with alzheimer’s disease and other diseases with memory impairment,” *Dementia and Geriatric Cognitive Disorders*, 7(4), pp. 182–186. doi: 10.1159/000106876.

Howieson, D. B. *et al.* (1993) “Neurologic function in the optimally healthy oldest old: Neuropsychological evaluation,” *Neurology*, 43(10), pp. 1882–1886. doi: 10.1212/wnl.43.10.1882.

Hudson, R. *et al.* (2006) “Effect of air pollution on olfactory function in residents of Mexico City,” *Chemical Senses*. Oxford University Press, 31(1), pp. 79–85. doi: 10.1093/chemse/bjj019.

Hummel, T. *et al.* (1997) “"Sniffin’ sticks", Olfactory performance assessed by the combined testing of odor identification, odor discrimination and olfactory threshold,” *Chemical Senses*, 22(1), pp. 39–52. doi: 10.1093/chemse/22.1.39.

Hummel, T. *et al.* (2001) “Screening of olfactory function with a four-minute odor

identification test: Reliability, normative data, and investigations in patients with olfactory loss," *Annals of Otology, Rhinology and Laryngology*, 110(10), pp. 976–981. doi: 10.1177/000348940111001015.

Hummel, T. *et al.* (2007) "Normative data for the "Sniffin' Sticks" including tests of odor identification, odor discrimination, and olfactory thresholds: An upgrade based on a group of more than 3,000 subjects," *European Archives of Oto-Rhino-Laryngology*, 264(3), pp. 237–243. doi: 10.1007/s00405-006-0173-0.

Hummel, T. and Nordin, S. (2005) "Olfactory disorders and their consequences for quality of life," *Acta Oto-Laryngologica*, 125(2), pp. 116–121. doi: 10.1080/00016480410022787.

Humphrey, D. G. and Kramer, A. F. (1999) "Age-related differences in perceptual organization and selective attention: Implications for display segmentation and recall performance," *Experimental Aging Research*, 25(1), pp. 1–26. doi: 10.1080/036107399244110.

Hüttenbrink, K.-B. *et al.* (2013) "Olfactory Dysfunction," *Deutsches Aerzteblatt Online*, 110(1–2), pp. 1–8. doi: 10.3238/arztebl.2013.0001.

Hyman, B. T., Arriagada, P. V. and Hoesen, G. W. Van (1991) "Pathologic Changes in the Olfactory System in Aging and Alzheimer's Diseasea," *Annals of the New York Academy of Sciences*, 640(1), pp. 14–19. doi: 10.1111/j.1749-6632.1991.tb00184.x.

Iannilli, E. *et al.* (2016) "Effects of manganese exposure on olfactory functions in teenagers: A pilot study," *PLoS ONE*. Public Library of Science, 11(1), p. e0144783. doi: 10.1371/journal.pone.0144783.

Ikram, M. A. and VanderWeele, T. J. (2015) "A proposed clinical and biological interpretation of mediated interaction," *European Journal of Epidemiology*, 30(10), pp. 1115–1118. doi: 10.1007/s10654-015-0087-5.

Imai, K., Keele, L. and Tingley, D. (2010) "A general approach to causal mediation analysis," *Psychological Methods*, 15(4), pp. 309–334. doi: 10.1037/a0020761.

Jacobs, L. *et al.* (2010) "Air Pollution–Related Prothrombotic Changes in Persons with Diabetes," *Environmental Health Perspectives*, 118(2), pp. 191–196. doi: 10.1289/ehp.0900942.

Janssen, N. A. *et al.* (2012) "Health effects of black carbon," *World Health Organization Regional Office for Europe*. World Health Organization, Regional Office for Europe. Available at: http://www.euro.who.int/en/health-topics/environment-and-health/air-quality/publications/2012/health-effects-of-black-carbon (Accessed: January 30, 2020).

Judd, C. M. and Kenny, D. A. (1981) "Process Analysis: Estimating Mediation in Treatment Evaluations," *Evaluation Review*. Sage PublicationsSage CA: Thousand Oaks, CA, 5(5), pp. 602–619. doi: 10.1177/0193841X8100500502.

Jung, H. J., Shin, I. S. and Lee, J. E. (2019) "Olfactory function in mild cognitive impairment and Alzheimer's disease: A meta-analysis," *Laryngoscope*. John Wiley and Sons Inc., pp. 362–369. doi: 10.1002/lary.27399.

Karrasch, M. *et al.* (2013) "CERAD test performance and cognitive impairment in Parkinson's disease," *Acta Neurologica Scandinavica*, 128(6), pp. 409–413. doi: 10.1111/ane.12138.

Katzman, R. and Kawas, C. (1994) "The epidemiology of dementia and Alzheimer disease," R. Gardner III et al. (eds.) *Alzheimer disease*. Thomson Brooks/Cole Publishing Co., pp. 105–122. doi: tbd.

Kemper, S., Herman, R. and Lian, C. (2003) "Age differences in sentence production," *Journals of Gerontology - Series B Psychological Sciences and Social Sciences*, 58(5), pp. P260–268. doi: 10.1093/geronb/58.5.P260.

Kobal, G. *et al.* (2000) “Multicenter investigation of 1,036 subjects using a standardized method for the assessment of olfactory function combining tests of odor identification, odor discrimination, and olfactory thresholds,” *European Archives of Oto-Rhino-Laryngology*, 257(4), pp. 205–211. doi: 10.1007/s004050050223.

Kraemer, H. C. *et al.* (2001) “How do risk factors work together ? Mediators , moderators, and independent, overlapping, and proxy risk factors,” *The American Journal of Psychiatry*, 158(6), pp. 848–856. doi: 10.1176/appi.ajp.158.6.848.

Kraemer, H. C. *et al.* (2002) “Mediators and moderators of treatment effects in randomized clinical trials,” *Archives of General Psychiatry*. American Medical Association, pp. 877–883. doi: 10.1001/archpsyc.59.10.877.

Kraemer, H. C. *et al.* (2008) “How and why criteria defining moderators and mediators differ between the Baron & Kenny and MacArthur approaches,” *Health psychology : official journal of the Division of Health Psychology, American Psychological Association*, 27(2S), pp. 101–108. doi: 10.1037/0278-6133.27.2(Suppl.).S101.

Kretzschmar, H. A. and Neumann, M. (2000) “Die neuropathologische diagnostik neurodegenerativer und demenzieller krankheiten,” *Pathologe*, 21(5), pp. 364–374. doi: 10.1007/s002920000402.

Kuhlmey, A. and Schaeffer, D. (2008) *Alter, Gesundheit und Krankheit: Handbuch Gesundheitswissenschaften*, *Verlag Hans Huber*. Available at: https://slidex.tips/download/adelheid-kuhlmey-doris-schaeffer-hrsg-alter-gesundheit-und-krankheit-handbuch-ge (Accessed: February 4, 2020).

Künzli, N. *et al.* (2000) “Public-health impact of outdoor and traffic-related air pollution: A European assessment,” *Lancet*. Lancet Publishing Group, 356(9232), pp. 795–801. doi: 10.1016/S0140-6736(00)02653-2.

Künzli, N., Perez, L. and Rapp, R. (2010) “Air Quality and Health, European Respiratory Journal,” Available at: https://www.ersnet.org/pdf/publications/air-quality-ENG.pdf (Accessed: March 21, 2018).

Laakso, M. P. *et al.* (2009) “Olfactory identification in non-demented elderly population and in mild cognitive impairment: A comparison of performance in clinical odor identification versus Boston Naming Test,” *Journal of Neural Transmission*, 116(7), pp. 891–895. doi: 10.1007/s00702-009-0235-8.

Lautenbacher, S. and Gauggel, S. (2010) “Neuropsychologie psychischer Störungen, Neuropsychologie psychischer Störungen,” *Springer Berlin Heidelberg.* doi: 10.1007/978-3-540-72340-0.

Levesque, S. *et al.* (2011) “Diesel Exhaust Activates and Primes Microglia: Air Pollution, Neuroinflammation, and Regulation of Dopaminergic Neurotoxicity,” *Environmental Health Perspectives*, 119(8), pp. 1149–1155. doi: 10.1289/ehp.1002986.

Lezak, Muriel D *et al.* (2004) “Neuropsychological Assessment. 4th Edition,” *Edited by M. D. Lezak, D. B. Howieson, and D. W. Loring. New York: Oxford University Press.* ISBN: 0195111214.

Liu, S., Jones, R. N. and Glymour, M. M. (2010) “Implications of Lifecourse Epidemiology for Research on Determinants of Adult Disease,” *Public Health Reviews*, 32(2), pp. 489–511. doi: 10.1007/BF03391613.

Loft, S. (2009) “Air quality and climate change,” *Ugeskrift for laeger*, 171(44), pp. 3168–3171. Available at: http://www.ncbi.nlm.nih.gov/pubmed/19857393 (Accessed: February 4, 2020).

Logan, W. P. D. (1953) “Mortality in the London Fog Incident,” *The Lancet*, 261(6755), pp. 336–338. doi: 10.1016/S0140-6736(53)91012-5.

Lucchini, R. G. *et al.* (2007) “High prevalence of parkinsonian disorders associated to

manganese exposure in the vicinities of ferroalloy industries," *American Journal of Industrial Medicine*, 50(11), pp. 788–800. doi: 10.1002/ajim.20494.

Lucchini, R. G., Dorman, D. C., *et al.* (2012) "Neurological impacts from inhalation of pollutants and the nose–brain connection," *NeuroToxicology*, 33(4), pp. 838–841. doi: 10.1016/j.neuro.2011.12.001.

Lucchini, R. G., Guazzetti, S., *et al.* (2012) "Tremor, olfactory and motor changes in Italian adolescents exposed to historical ferro-manganese emission," *NeuroToxicology*, 33(4), pp. 687–696. doi: 10.1016/j.neuro.2012.01.005.

Luck, T. *et al.* (2009) "CERAD-NP-Testbatterie: Alters-, geschlechts- und bildungsspezifische Normen ausgewählter Subtests. Ergebnisse der German Study on Ageing, Cognition and Dementia in Primary Care Patients (AgeCoDe)," *Zeitschrift für Gerontologie und Geriatrie*, 42(5), pp. 372–384. doi: 10.1007/s00391-009-0031-y.

Luszcz, M. A. and Bryana, J. (1999) "Toward understanding age-related memory loss in late adulthood," *Gerontology*, 45(1), pp. 2–9. doi: 10.1159/000022048.

MacKinnon, D. P., Krull, J. L. and Lockwood, C. M. (2000) "Equivalence of the mediation, confounding and suppression effect," *Prevention science : the official journal of the Society for Prevention Research*, 1(4), pp. 173–181. doi: 10.1023/a:1026595011371.

Maher, B. A. *et al.* (2016) "Magnetite pollution nanoparticles in the human brain," *Proceedings of the National Academy of Sciences of the United States of America*. National Academy of Sciences, 113(39), pp. 10797–10801. doi: 10.1073/pnas.1605941113.

Maisonet, M. *et al.* (2004) "A review of the literature on the effects of ambient air pollution on fetal growth," *Environmental research*, 95(1), pp. 106–15. doi: 10.1016/j.envres.2004.01.001.

Mar, T. F. *et al.* (2005) "Fine particulate air pollution and cardiorespiratory effects in the elderly," *Epidemiology*, 16(5), pp. 681–687. doi: 10.1097/01.ede.0000173037.83211.d6.

Marin, C. *et al.* (2018) "Olfactory Dysfunction in Neurodegenerative Diseases," *Current Allergy and Asthma Reports*. Current Medicine Group LLC 1, p. 42. doi: 10.1007/s11882-018-0796-4.

Matulionis, D. H. (1974) "Ultrastructure of olfactory epithelia in mice after smoke exposure," *Annals of Otology, Rhinology & Laryngology*. SAGE PublicationsSage CA: Los Angeles, CA, 83(2), pp. 192–201. doi: 10.1177/000348947408300207.

McFall, G. P. *et al.* (2010) "Testing covariates of Type 2 diabetes-cognition associations in older adults: Moderating or mediating effects?," *Neuropsychology*, 24(5), pp. 547–562. doi: 10.1037/a0019246.

Medina, S. *et al.* (2009) "The apheis project: Air pollution and health-A European information system," *Air Quality, Atmosphere and Health*, 2(4), pp. 185–198. doi: 10.1007/s11869-009-0050-2.

Memory Clinic University Hospital Basel (2005) "CERAD-Plus test battery, revised edition 2005. German version," Available at: https://www.memoryclinic.ch/de/main-navigation/neuropsychologen/cerad-plus/ (Accessed: February 23, 2020).

Miller, K. A. *et al.* (2007) "Long-Term Exposure to Air Pollution and Incidence of Cardiovascular Events in Women," *New England Journal of Medicine*, 356(5), pp. 447–458. doi: 10.1056/NEJMoa054409.

Mills, N. L. *et al.* (2009) "Adverse cardiovascular effects of air pollution," *Nature Clinical Practice. Cardiovascular medicine*, 6(1), pp. 36–44. doi: 10.1038/ncpcardio1399.

Mirra, S. S. *et al.* (1991) "The Consortium to Establish a Registry for Alzheimer's Disease (CERAD): Part II. Standardization of the neuropathologic assessment of Alzheimer's

disease," *Neurology*, 41(4), pp. 479–479. doi: 10.1212/WNL.41.4.479.

Mitis, F., Iavarone, I. and Martuzzi, M. (2007) "Health impact of ozone in 13 Italian cities," *Epidemiologia e prevenzione*, 31(6), pp. 323–332. Available at: http://www.ncbi.nlm.nih.gov/pubmed/18326424.

Morgan, K. T. and Monticello, T. M. (1990) "Airflow, gas deposition, and lesion distribution in the nasal passages," *Environmental Health Perspectives*. Environmental Health Perspectives, 85, pp. 209–218. doi: 10.1289/ehp.85-1568327.

Morris, J. C. *et al.* (1989) "The consortium to establish a registry for alzheimer's disease (CERAD). Part I. Clinical and neuropsychological assessment of alzheimer's disease," *Neurology*, 39(9), pp. 1159–1165. doi: 10.1212/wnl.39.9.1159.

Müller, A. *et al.* (2002) "Olfactory function in Parkinsonian syndromes," *Journal of Clinical Neuroscience*, 9(5), pp. 521–524. doi: 10.1054/jocn.2001.1071.

Murphy, C. *et al.* (2002) "Prevalence of olfactory impairment in older adults," *Journal of the American Medical Association*, 288(18), pp. 2307–2312. doi: 10.1001/jama.288.18.2307.

Murphy, C., Anderson, J. A. and Markison, S. (1994) "Psychophysical Assessment of Chemosensory Disorders in Clinical Populations," *Olfaction and Taste XI*. Springer Japan, pp. 609–613. doi: 10.1007/978-4-431-68355-1_251.

Naegele, G. *et al.* (2013) "Aus Politik und Zeitgeschichte: Alternde Gesellschaft," Available at: https://www.bpb.de/shop/zeitschriften/apuz/153013/alternde-gesellschaft (Accessed: February 4, 2020).

Nafstad, P. *et al.* (2003) "Lung cancer and air pollution: A 27 year follow up of 16 209 Norwegian men," *Thorax*. BMJ Publishing Group, 58(12), pp. 1071–1076. doi: 10.1136/thorax.58.12.1071.

National Research Council (2008) "Estimating Mortality Risk Reduction and Economic Benefits from Controlling Ozone Air Pollution, Estimating Mortality Risk Reduction and Economic Benefits from Controlling Ozone Air Pollution," *Washington, D.C.: National Academies Press.* doi: 10.17226/12198.

Nemery, B., Hoet, P. H. M. and Nemmar, A. (2001) "The Meuse Valley fog of 1930: An air pollution disaster," *Lancet*. Elsevier Limited, 357(9257), pp. 704–708. doi: 10.1016/S0140-6736(00)04135-0.

Nemmar, A. *et al.* (2001) "Passage of intratracheally instilled ultrafine particles from the lung into the systemic circulation in hamster," *American Journal of Respiratory and Critical Care Medicine*. American Lung Association, 164(9), pp. 1665–1668. doi: 10.1164/ajrccm.164.9.2101036.

Norès, J. M., Biacabe, B. and Bonfils, P. (2000) "Chemicals toxic to the olfactory system. Analysis and description," *Presse medicale (Paris, France : 1983)*, 29(32), pp. 1773–17781. Available at: http://www.ncbi.nlm.nih.gov/pubmed/11098278 (Accessed: February 14, 2020).

Oberdörster, G. *et al.* (2004) "Translocation of Inhaled Ultrafine Particles to the Brain," *Inhalation Toxicology*, 16(6–7), pp. 437–445. doi: 10.1080/08958370490439597.

Oberdörster, G., Elder, A. and Rinderknecht, A. (2009) "Nanoparticles and the Brain: Cause for Concern?," *Journal of Nanoscience and Nanotechnology*, 9(8), pp. 4996–5007. doi: 10.1166/jnn.2009.GR02.

Oosterman, J. M. *et al.* (2010) "Assessing mental flexibility: Neuroanatomical and neuropsychological correlates of the trail making test in elderly people," *Clinical Neuropsychologist*, 24(2), pp. 203–219. doi: 10.1080/13854040903482848.

Perez, L. *et al.* (2015) "Air pollution and atherosclerosis: A cross-sectional analysis of

four European cohort studies in the ESCAPE study," *Environmental Health Perspectives*, 123(6), pp. 597–605. doi: 10.1289/ehp.1307711.

Peters, A. *et al.* (1997) "Increased plasma viscosity during an air pollution episode: A link to mortality?," *Lancet*, 349(9065), pp. 1582–1587. doi: 10.1016/S0140-6736(97)01211-7.

Peters, A. (2001) "Particulate air pollution is associated with an acute phase response in men. Results from the MONICA-Augsburg Study," *European Heart Journal*, 22(14), pp. 1198–1204. doi: 10.1053/euhj.2000.2483.

Peters, A. (2005) "Particulate matter and heart disease: Evidence from epidemiological studies," *Toxicology and Applied Pharmacology*, 207(2), pp. 477–482. doi: 10.1016/j.taap.2005.04.030.

Peters, A. *et al.* (2006) "Translocation and potential neurological effects of fine and ultrafine particles a critical update," *Particle and fibre toxicology*, 3(13), pp. 1–13. doi: 10.1186/1743-8977-3-13.

Petersen, R. C. (2004) "Mild cognitive impairment as a diagnostic entity," in *Journal of Internal Medicine*, pp. 183–194. doi: 10.1111/j.1365-2796.2004.01388.x.

Pope, C. A. (1989) "Respiratory disease associated with community air pollution and a steel mill, Utah Valley," *American Journal of Public Health*, 79(5), pp. 623–628. doi: 10.2105/AJPH.79.5.623.

Pope, C. A. *et al.* (2002) "Lung cancer, cardiopulmonary mortality, and long-term exposure to fine particulate air pollution," *JAMA*, 287(9), pp. 1132–1141. doi: 10.1001/jama.287.9.1132.

Pope, C. A. and Dockery, D. W. (2006) "Health Effects of Fine Particulate Air Pollution: Lines that Connect," *Journal of the Air & Waste Management Association*, 56(6), pp. 709–742. doi: 10.1080/10473289.2006.10464485.

Power, M. C. *et al.* (2011) "Traffic-Related Air Pollution and Cognitive Function in a Cohort of Older Men," *Environmental Health Perspectives*, 119(5), pp. 682–687. doi: 10.1289/ehp.1002767.

Power, M. C. *et al.* (2016) "Exposure to air pollution as a potential contributor to cognitive function, cognitive decline, brain imaging, and dementia: A systematic review of epidemiologic research," *Neurotoxicology*, 56, pp. 235–253. doi: 10.1016/j.neuro.2016.06.004.

Quarmley, M. *et al.* (2017) "Odor Identification Screening Improves Diagnostic Classification in Incipient Alzheimer's Disease," *Journal of Alzheimer's Disease*. IOS Press, 55(4), pp. 1497–1507. doi: 10.3233/JAD-160842.

R Development Core Team (2013) "R: A Language and Environment for Statistical Computing," *Vienna, Austria.* Available at: http://www.r-project.org (Accessed: March 22, 2018).

Rabbitt, P. and Lowe, C. (2000) "Patterns of cognitive ageing," *Psychological Research*. Springer Verlag, 63(3–4), pp. 308–316. doi: 10.1007/s004269900009.

Ranft, U. *et al.* (2009) "Long-term exposure to traffic-related particulate matter impairs cognitive function in the elderly," *Environmental Research*, 109(8), pp. 1004–1011. doi: 10.1016/j.envres.2009.08.003.

Rassow, J. *et al.* (2016) "Duale Reihe Biochemie, Thieme," *Edited by J. Rassow et al. Stuttgart: Georg Thieme Verlag.* doi: 10.1055/b-003-129341.

Rivest, S. *et al.* (2000) "How the Blood Talks to the Brain Parenchyma and the Paraventricular Nucleus of the Hypothalamus During Systemic Inflammatory and Infectious Stimuli," *Proceedings of the Society for Experimental Biology and Medicine*.

Blackwell Publishing Inc., 223(1), pp. 22–38. doi: 10.1046/j.1525-1373.2000.22304.x.

Rückerl, R. *et al.* (2006) “Air Pollution and Markers of Inflammation and Coagulation in Patients with Coronary Heart Disease,” *American Journal of Respiratory and Critical Care Medicine*, 173(4), pp. 432–441. doi: 10.1164/rccm.200507-1123OC.

Rückerl, R. *et al.* (2007) “Air pollution and inflammation (Interleukin-6, C-reactive protein, fibrinogen) in myocardial infarction survivors,” *Environmental Health Perspectives*, 115(7), pp. 1072–1080. doi: 10.1289/ehp.10021.

Rückerl, R. *et al.* (2011) “Health effects of particulate air pollution: A review of epidemiological evidence,” *Inhalation Toxicology*, 23(10), pp. 555–592. doi: 10.3109/08958378.2011.593587.

La Rue, A. *et al.* (1992) “Cognitive performance in relatives of patients with probable Alzheimer disease: An age at onset effect?,” *Journal of Clinical and Experimental Neuropsychology*, 14(4), pp. 533–538. doi: 10.1080/01688639208402842.

Ryan, J. J., Sattler, J. M. and Lopez, S. J. (2000) “Age effects on Wechsler Adult Intelligence Scale-III subtests,” *Archives of clinical neuropsychology : the official journal of the National Academy of Neuropsychologists*, 15(4), pp. 311–407. Available at: http://www.ncbi.nlm.nih.gov/pubmed/14590227 (Accessed: February 5, 2020).

Sagai, M. and Tin Win-Shwe, T. (2015) “Oxidative Stress Derived from Airborne Fine and Ultrafine Particles and the Effects on Brain-Nervous System: Part 2,” *Nihon eiseigaku zasshi. Japanese journal of hygiene*, pp. 220–229. doi: 10.1265/jjh.70.220.

Salthouse, T. A. *et al.* (1995) “Aging of attention: Does the ability to divide decline?,” *Memory & Cognition*, 23(1), pp. 59–71. doi: 10.3758/BF03210557.

Salthouse, T. A., Babcock, R. L. and Shaw, R. J. (1991) “Effects of adult age on structural and operational capacities in working memory,” *Psychology and aging*, 6(1), pp. 118–127. doi: 10.1037/0882-7974.6.1.118.

Santos, D. V *et al.* (2004) “Hazardous Events Associated With Impaired Olfactory Function,” *Archives of Otolaryngology–Head & Neck Surgery*, 130(3), p. 317. doi: 10.1001/archotol.130.3.317.

Sarazin, M. and Dubois, B. (2002) “Mild cognitive impairment or pre-demential Alzheimer’s disease?,” *Revue Neurologique*, 158(10 SUPPL.), pp. 30–34.

Schiffman, S. S. and Nagle, H. T. (1992) “Effect of environmental pollutants on taste and smell,” *Otolaryngology - Head and Neck Surgery*, pp. 693–700. doi: 10.1177/019459989210600613.

Schikowski, T. *et al.* (2005) “Long-term air pollution exposure and living close to busy roads are associated with COPD in women,” *Respiratory Research*, 6(152). doi: 10.1186/1465-9921-6-152.

Schikowski, T. *et al.* (2008) “Contribution of smoking and air pollution exposure in urban areas to social differences in respiratory health,” *BMC Public Health*, 8, pp. 1–10. doi: 10.1186/1471-2458-8-179.

Schikowski, T. *et al.* (2015) “Association of air pollution with cognitive functions and its modification by APOE gene variants in elderly women,” *Environmental Research*. Elsevier, 142, pp. 10–16. doi: 10.1016/j.envres.2015.06.009.

Schubert, C. R. *et al.* (2008) “Olfaction and the 5-Year Incidence of Cognitive Impairment in an Epidemiological Study of Older Adults,” *Journal of the American Geriatrics Society*, 56(8), pp. 1517–1521. doi: 10.1111/j.1532-5415.2008.01826.x.

Schubert, C. R. *et al.* (2009) “Olfactory Impairment in Adults,” *Annals of the New York Academy of Sciences*, 1170(1), pp. 531–536. doi: 10.1111/j.1749-6632.2009.04102.x.

Schulz, H. *et al.* (2019) “Breathing: Ambient Air Pollution and Health - Part II,”

Pneumologie. Georg Thieme Verlag, pp. 347–373. doi: 10.1055/a-0895-6494.

Schünke, M. *et al.* (2018) "PROMETHEUS Kopf, Hals und Neuroanatomie," *Stuttgart: Georg Thieme Verlag.* doi: 10.1055/b-006-149644.

Schwartz, B. S. *et al.* (1989) "Olfactory function in chemical workers exposed to acrylate and methacrylate vapors," *American Journal of Public Health*, 79(5), pp. 613–618. doi: 10.2105/AJPH.79.5.613.

Schwartz, J. (2004) "Air pollution and children's health," *Pediatrics*, 113(SUPPL. 4), pp. 1037–1043. doi: 10.1542/peds.113.4.S1.1037.

Seaton, A. *et al.* (1999) "Particulate air pollution and the blood," *Thorax*, 54(11), pp. 1027–1032. doi: 10.1136/thx.54.11.1027.

Shiga, H. *et al.* (2011) "Evaluation of the Olfactory Nerve Transport Function by SPECT-MRI Fusion Image with Nasal Thallium-201 Administration," *Molecular Imaging and Biology*, 13(6), pp. 1262–1266. doi: 10.1007/s11307-010-0461-3.

Shiga, H. *et al.* (2013) "Assessment of Olfactory Nerve by SPECT-MRI Image with Nasal Thallium-201 Administration in Patients with Olfactory Impairments in Comparison to Healthy Volunteers," *PLoS ONE*, 8(2), p. e57671. doi: 10.1371/journal.pone.0057671.

Shiga, H. *et al.* (2017) "Prognostic value of olfactory nerve damage measured with thallium-based olfactory imaging in patients with idiopathic olfactory dysfunction," *Scientific Reports*. Nature Publishing Group, 7(1), p. 3581. doi: 10.1038/s41598-017-03894-4.

Ship, J. A. *et al.* (1996) "Longitudinal changes in smell identification," *Journals of Gerontology - Series A Biological Sciences and Medical Sciences*. Oxford University Press, 51(2). doi: 10.1093/gerona/51A.2.M86.

Shou, Y. *et al.* (2019) "A review of the possible associations between ambient PM2.5 exposures and the development of Alzheimer's disease," *Ecotoxicology and Environmental Safety*. Academic Press, 174, pp. 344–352. doi: 10.1016/j.ecoenv.2019.02.086.

Singh-Manoux, A. *et al.* (2012) "Timing of onset of cognitive decline: Results from Whitehall II prospective cohort study," *BMJ (Online)*, 344(7840). doi: 10.1136/bmj.d7622.

Snowdon, D. A. *et al.* (1996) "Linguistic ability in early life and cognitive function and Alzheimer's disease in late life: Findings from the Nun Study," *Journal of the American Medical Association*, 275(7), pp. 528–532. doi: 10.1001/jama.275.7.528.

Stevenson, R. J. (2009) "An initial evaluation of the functions of human olfaction," *Chemical Senses*, pp. 3–20. doi: 10.1093/chemse/bjp083.

Suglia, S. F. *et al.* (2007) "Association of Black Carbon with Cognition among Children in a Prospective Birth Cohort Study," *American Journal of Epidemiology*, 167(3), pp. 280–286. doi: 10.1093/aje/kwm308.

Sunderman, F. W. (2001) "Nasal toxicity, carcinogenicity, and olfactory uptake of metals," *Annals of Clinical and Laboratory Science*, 31(1), pp. 3–24. Available at: http://www.ncbi.nlm.nih.gov/pubmed/11314863.

Tan, S. Y. *et al.* (2016) "A review of heavy metals in indoor dust and its human health-risk implications," *Reviews on Environmental Health*. Walter de Gruyter GmbH, pp. 447–456. doi: 10.1515/reveh-2016-0026.

Teichert, T. *et al.* (2013) "Association between Traffic-Related Air Pollution, Subclinical Inflammation and Impaired Glucose Metabolism: Results from the SALIA Study," *PLoS ONE*. Edited by G. Targher, 8(12), p. e83042. doi: 10.1371/journal.pone.0083042.

Tin-Tin-Win-Shwe *et al.* (2006) "Brain cytokine and chemokine mRNA expression in mice induced by intranasal instillation with ultrafine carbon black," *Toxicology Letters*, 163(2),

pp. 153–160. doi: 10.1016/j.toxlet.2005.10.006.

Tingley, D. *et al.* (2014) “mediation: R Package for Causal Mediation Analysis,” *Journal of Statistical Software*, 59(5), pp. 1–38. doi: 10.18637/jss.v059.i05.

Tjälve, H. and Henriksson, J. (1999) “Uptake of metals in the brain via olfactory pathways,” *Neurotoxicology*, 20(2–3), pp. 181–95. Available at: http://www.ncbi.nlm.nih.gov/pubmed/10385882.

Tonne, C. *et al.* (2014) “Traffic-related air pollution in relation to cognitive function in older adults,” *Epidemiology (Cambridge, Mass.)*, 25(5), pp. 674–81. doi: 10.1097/EDE.0000000000000144.

Upadhyay, U. D. and Holbrook, E. H. (2004) “Olfactory loss as a result of toxic exposure,” *Otolaryngologic Clinics of North America*. W.B. Saunders, pp. 1185–1207. doi: 10.1016/j.otc.2004.05.003.

Valeri, L. and VanderWeele, T. J. (2013) “Mediation analysis allowing for exposure–mediator interactions and causal interpretation: Theoretical assumptions and implementation with SAS and SPSS macros,” *Psychological Methods*. NIH Public Access, 18(2), pp. 137–150. doi: 10.1037/a0031034.

VanderWeele, T. J. (2013) “A Three-way Decomposition of a Total Effect into Direct, Indirect, and Interactive Effects,” *Epidemiology*, 24(2), pp. 224–232. doi: 10.1097/EDE.0b013e318281a64e.

Vent, J. *et al.* (2003) “The impact of ethanol and tobacco smoke on intranasal epithelium in the rat,” *American Journal of Rhinology*, 17(4), pp. 241–247. doi: 10.1177/194589240301700411.

Vent, J. *et al.* (2004) “Pathology of the olfactory epithelium: Smoking and ethanol exposure,” *Laryngoscope*, 114(8), pp. 1383–1388. doi: 10.1097/00005537-200408000-00012.

Veronesi, B. *et al.* (2005) “Effects of Subchronic Exposures to Concentrated Ambient Particles: VII. Degeneration of Dopaminergic Neurons in Apo E −/− Mice,” *Inhalation Toxicology*, 17(4–5), pp. 235–241. doi: 10.1080/08958370590912888.

Vierkötter, A. *et al.* (2010) “Airborne particle exposure and extrinsic skin aging,” *Journal of Investigative Dermatology*, 130(12), pp. 2719–2726. doi: 10.1038/jid.2010.204.

Visser, P. J. and Verhey, F. J. (2008) “Mild cognitive impairment as predictor for Alzheimer’s disease in clinical practice: Effect of age and diagnostic criteria,” *Psychological Medicine*, 38(1), pp. 113–122. doi: 10.1017/S0033291707000554.

Vossoughi, M. *et al.* (2014) “Air pollution and subclinical airway inflammation in the SALIA cohort study,” *Immunity & Ageing*. BioMed Central Ltd., 11(1), pp. 1–5. doi: 10.1186/1742-4933-11-5.

Waller, R. E., Brooks, A. G. and Adler, M. W. (1973) “The 1952 fog cohort study,” *British journal of preventive & social medicine*, 27(1), pp. 68–69. doi: 10.1136/jech.27.1.68-c.

Wang, M. *et al.* (2013) “Evaluation of land use regression models for NO2 and particulate matter in 20 European study areas: The ESCAPE project,” *Environmental Science and Technology*, 47(9), pp. 4357–4364. doi: 10.1021/es305129t.

Wehling, E. I. *et al.* (2016) “Longitudinal changes in odor identification performance and neuropsychological measures in aging individuals,” *Neuropsychology*. American Psychological Association Inc., 30(1), pp. 87–97. doi: 10.1037/neu0000212.

Weis, S. *et al.* (2019) “Normal Aging Brain,” in *Imaging Brain Diseases*. Springer Vienna, pp. 871–895. doi: 10.1007/978-3-7091-1544-2_31.

Wellenius, G. A. *et al.* (2012) “Residential proximity to nearest major roadway and cognitive function in community-dwelling seniors: Results from the mobilize Boston

Study," *Journal of the American Geriatrics Society*, 60(11), pp. 2075–80. doi: 10.1111/j.1532-5415.2012.04195.x.

Wellenius, G. A., Schwartz, J. and Mittleman, M. A. (2005) "Air Pollution and Hospital Admissions for Ischemic and Hemorrhagic Stroke Among Medicare Beneficiaries," *Stroke*, 36(12), pp. 2549–2553. doi: 10.1161/01.STR.0000189687.78760.47.

Welsh, K. A. *et al.* (1994) "The Consortium to Establish a Registry for Alzheimer's Disease (CERAD). Part V. A normative study of the neuropsychological battery," *Neurology*, 44(4), pp. 609–609. doi: 10.1212/WNL.44.4.609.

Weuve, J. (2012) "Exposure to Particulate Air Pollution and Cognitive Decline in Older Women," *Archives of Internal Medicine*, 172(3), pp. 219–227. doi: 10.1001/archinternmed.2011.683.

WHO (2019) "Risk reduction for cognitive decline and dementia: WHO guidelines, World Health Organization 2019," *Geneva: World Health Organization.* Available at: https://www.who.int/mental_health/neurology/dementia/Dementia_Guidelines_Evidence_Profiles.pdf?ua=1 (Accessed: February 6, 2020).

WHO Health Data (2020) "Bevölkerung im Jahresdurchschnitt," Available at: http://www.gbe-bund.de/gbe10/ergebnisse.prc_tab?fid=9051&suchstring=&query_id=&sprache=D&fund_typ=TAB&methode=&vt=&verwandte=1&page_ret=0&seite=1&p_lfd_nr=3&p_news=&p_sprachkz=D&p_uid=gast&p_aid=76149453&hlp_nr=2&p_janein=J (Accessed: February 4, 2020).

World Health Organization, By: Henschel, S. and Chan, G. (2013) "Health risks of air pollution in Europe – HRAPIE project New emerging risks to health from air pollution – results from the survey of experts," *WHO*, p. 65. Available at: http://www.euro.who.int/__data/assets/pdf_file/0017/234026/e96933.pdf?ua=1. (Accessed: February 4, 2020).

Wilhelm, M. and Ritz, B. (2005) "Local variations in CO and particulate air pollution and adverse birth outcomes in Los Angeles County, California, USA," *Environmental Health Perspectives*, 113(9), pp. 1212–1221. doi: 10.1289/ehp.7751.

Wilson, R. S. *et al.* (2006) "Odor identification and decline in different cognitive domains in old age," *Neuroepidemiology*, 26(2), pp. 61–67. doi: 10.1159/000090250.

Wilson, R. S., Schneider, J. A., *et al.* (2007) "Olfactory identification and incidence of mild cognitive impairment in older age," *Archives of General Psychiatry*, 64(7), pp. 802–808. doi: 10.1001/archpsyc.64.7.802.

Wilson, R. S., Arnold, S. E., *et al.* (2007) "The relationship between cerebral Alzheimer's disease pathology and odour identification in old age," *Journal of Neurology, Neurosurgery and Psychiatry*. BMJ Publishing Group, 78(1), pp. 30–35. doi: 10.1136/jnnp.2006.099721.

Wilson, R. S., Yu, L. and Bennett, D. A. (2011) "Odor Identification and Mortality in Old Age," *Chemical Senses*, 36(1), pp. 63–67. doi: 10.1093/chemse/bjq098.

World Health Organization (2005) "WHO Air quality guidelines for particulate matter, ozone, nitrogen dioxide and sulfur dioxide: Global update 2005," *WHO.* doi: 10.1016/0004-6981(88)90109-6.

World Health Organization (2016) "Ambient Air Pollution: A global assessment of exposure and burden of disease," *WHO.* doi: 9789241511353.

Yahiaoui-Doktor, M. *et al.* (2019) "Olfactory function is associated with cognitive performance: Results from the population-based LIFE-Adult-Study," *Alzheimer's Research and Therapy*. BioMed Central Ltd., 11(1), p. 43. doi: 10.1186/s13195-019-0494-z.

Yitshak-Sade, M. *et al.* (2019) “Estimating the causal effect of annual PM2.5 exposure on mortality rates in the Northeastern and mid-Atlantic states,” *Environmental epidemiology (Philadelphia, Pa.)*, 3(4), p. e052. doi: 10.1097/EE9.0000000000000052.

Yu, Q. *et al.* (2018) “Olfactory dysfunction and its relationship with clinical symptoms of Alzheimer disease,” *Aging and Disease*. International Society on Aging and Disease, 9(6), pp. 1084–1095. doi: 10.14336/AD.2018.0819.

Zaudig, M. (1996) “Assessing behavioral symptoms of dementia of the Alzheimer type: Categorical and quantitative approaches,” *International Psychogeriatrics*. Cambridge University Press, 8(SUPPL. 2), pp. 183–200. doi: 10.1017/S1041610297003347.

Zhang, Z. F., Yu, S. Z. and Zhou, G. D. (1988) “Indoor air pollution of coal fumes as a risk factor of stroke, Shanghai,” *American Journal of Public Health*, 78(8), pp. 975–977. doi: 10.2105/AJPH.78.8.975.

Zhu, Y. *et al.* (2002) “Concentration and size distribution of ultrafine particles near a major highway,” *Journal of the Air and Waste Management Association*, 52(9), pp. 1032–1042. doi: 10.1080/10473289.2002.10470842.

Zou, Y. M. *et al.* (2016) “Olfactory dysfunction in Alzheimer’s disease,” *Neuropsychiatric Disease and Treatment*. Dove Medical Press Ltd., pp. 869–875. doi: 10.2147/NDT.S104886.

6 Appendix

Appendix A: Selection of mediation analyses for further CERAD subtests.

Table A1: Mediation analysis between air pollution and the CERAD subtest **"Word list discriminability"** mediated by odor identification (follow-up investigations in A) 2008/2009 (N=733) and B) 2012/2013 (N=542)).

	Average indirect effect			**Average direct effect**			**Total effect**			**Estimated proportion mediated**	
	β-estimate	**95%-CI**	**p-value**	**β-estimate**	**95%-CI**	**p-value**	**β-estimate**	**95%-CI**	**p-value**	**Proportion mediated %[a]**	**p-value**
A) Word list discriminability 2008/2009.											
PM_{10} [µg/m³]	<0.01	−0.01; 0.01	0.884	−0.02	−0.12; 0.08	0.728	−0.02	−0.12; 0.08	0.742	0.1	0.986
$PM_{2.5}$ [µg/m³]	<0.01	−0.02; 0.01	0.688	−0.07	−0.21; 0.07	0.394	−0.07	−0.21; 0.07	0.370	1.3	0.742
$PM_{2.5}$ abs [10^{-5}/m]	<0.01	−0.01; 0.01	0.908	−0.04	−0.13; 0.05	0.428	−0.04	−0.13; 0.05	0.442	−0.2	0.974
NO_2 [µg/m³]	<0.01	−0.01; 0.01	0.774	−0.12	−0.26; 0.01	0.062	−0.12	−0.26; 0.01	0.064	0.8	0.790
B) Word list discriminability 2012/2013.											
PM_{10} [µg/m³]	<0.01	−0.01; 0.01	0.964	0.09	−0.03; 0.21	0.174	0.09	−0.03; 0.21	0.166	<0.1	0.998
$PM_{2.5}$ [µg/m³]	<0.01	−0.02; 0.01	0.602	0.08	−0.09; 0.25	0.370	0.08	−0.09; 0.24	0.396	−1.2	0.790
$PM_{2.5}$ abs [10^{-5}/m]	<0.01	−0.01; 0.01	0.980	0.05	−0.07; 0.17	0.462	0.05	−0.07; 0.16	0.460	0.2	0.952
NO_2 [µg/m³]	<0.01	−0.02; 0.01	0.754	−0.01	−0.17; 0.16	0.936	−0.01	−0.17; 0.16	0.920	0.6	0.874

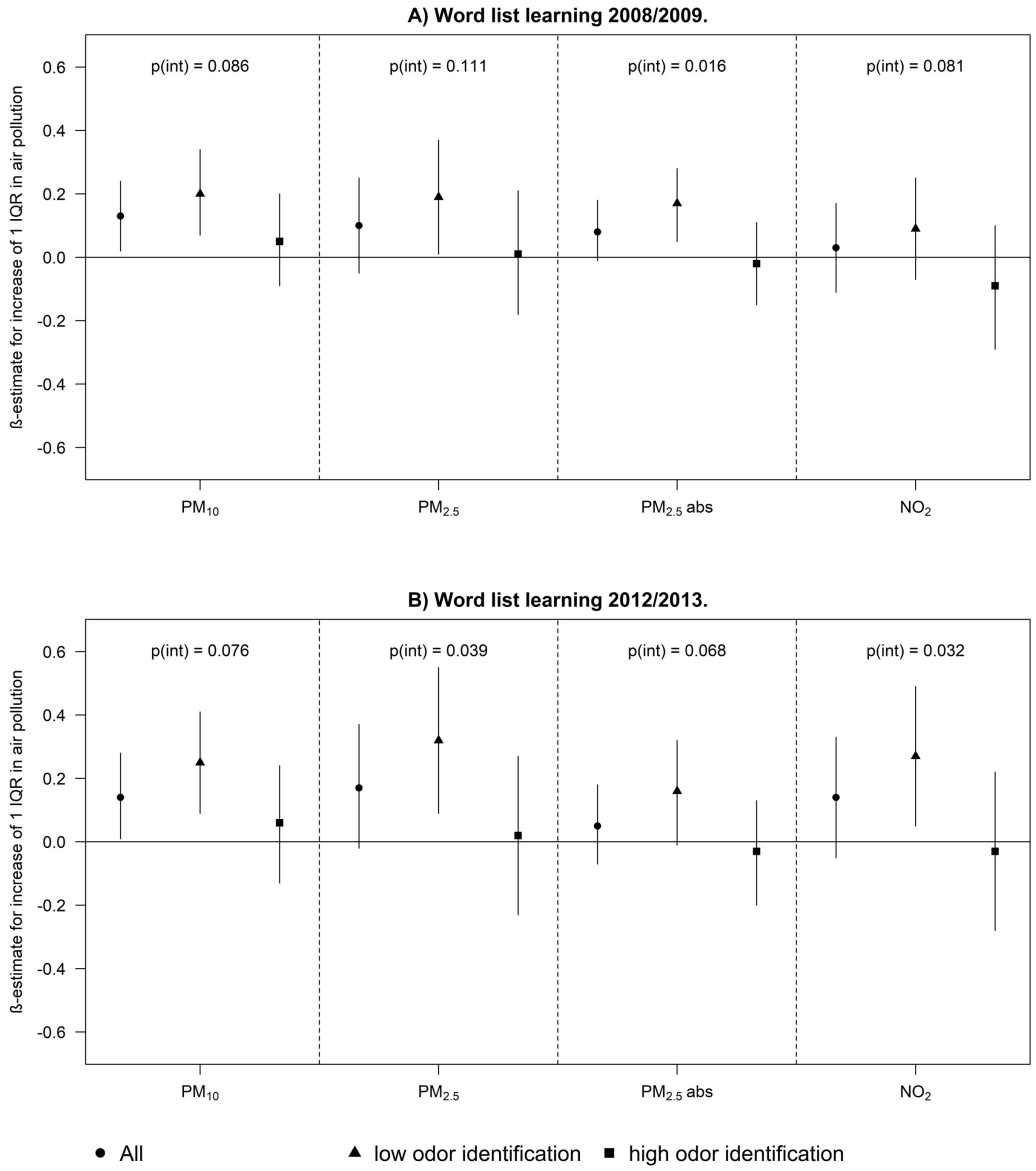
Estimated percent of the association between long-term air pollution exposure and cognitive impairment that was mediated by a decrease in odor identification. Approach utilizing exposure and mediator averages with interaction. Exposure: air pollution; mediator: odor identification; outcome: **CERAD subtest "world list discriminability"**. Presented are indirect, direct, and total effects (β-estimates, 95%-confidence intervals (95%-CI), and p-values). All models were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide. [a]: Proportion mediated = mediated effect / (direct effect + mediated effect) x 100.

Table A2: Mediation analysis between air pollution and the CERAD subtest of **visuo-construction performance** mediated by odor identification (follow-up investigations in A) 2008/2009 (N=733) and B) 2012/2013 (N=542)).

	Average indirect effect			Average direct effect			Total effect			Estimated proportion mediated	
	β-estimate	95%-CI	p-value	β-estimate	95%-CI	p-value	β-estimate	95%-CI	p-value	Proportion mediated %[a]	p-value
A) Visuo-construction performance 2008/2009.											
PM_{10} [µg/m³]	<0.01	−0.01; 0.01	0.878	−0.01	−0.22; 0.01	0.096	−0.10	−0.22; 0.01	0.108	−0.3	0.886
$PM_{2.5}$ [µg/m³]	<0.01	−0.02; 0.01	0.708	−0.13	−0.30; 0.04	0.140	−0.13	−0.30; 0.04	0.130	0.9	0.726
$PM_{2.5}$ abs [10^{-5}/m]	<0.01	−0.01; 0.01	0.912	−0.08	−0.19; 0.02	0.142	−0.08	−0.19; 0.02	0.144	−0.2	0.948
NO_2 [µg/m³]	<0.01	−0.02; 0.01	0.804	−0.26	−0.41; −0.10	<0.001	−0.26	−0.41; −0.11	<0.001	0.3	0.804
B) Visuo-construction performance 2012/2013.											
PM_{10} [µg/m³]	<0.01	−0.02; 0.02	0.984	0.04	−0.10; 0.17	0.618	0.04	−0.10; 0.17	0.624	0.9	0.940
$PM_{2.5}$ [µg/m³]	−0.01	−0.04; 0.02	0.544	−0.06	−0.26; 0.13	0.602	−0.07	−0.26; 0.13	0.544	3.8	0.728
$PM_{2.5}$ abs [10^{-5}/m]	<0.01	−0.02; 0.02	0.984	0.04	−0.10; 0.18	0.542	0.04	−0.09; 0.18	0.548	0.8	0.940
NO_2 [µg/m³]	−0.01	−0.04; 0.02	0.710	−0.22	−0.42; −0.02	0.028	−0.22	−0.42; −0.03	0.018	2.0	0.716

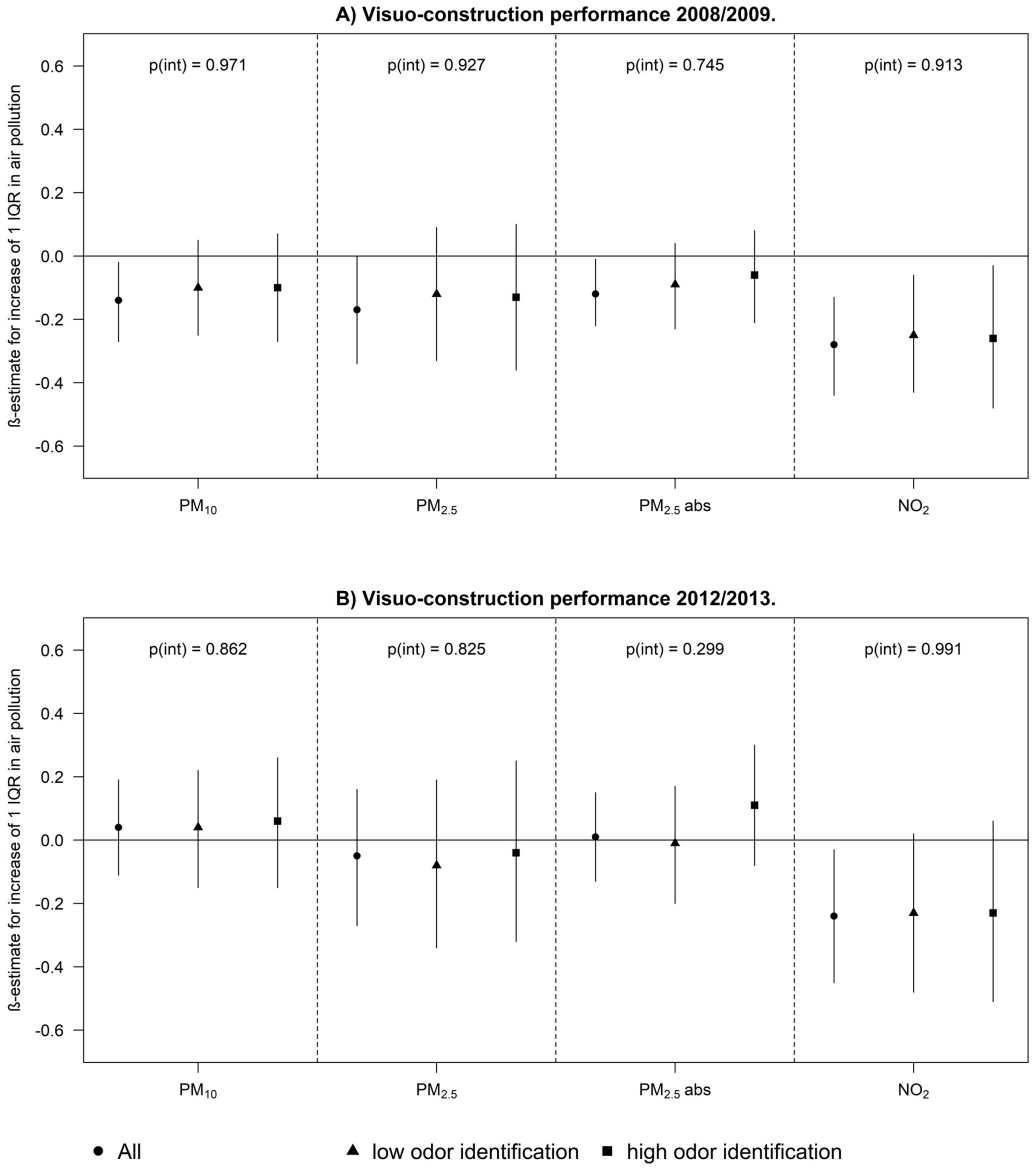
Estimated percent of the association between long-term air pollution exposure and cognitive impairment that was mediated by a decrease in odor identification. Approach utilizing exposure and mediator averages with interaction. Exposure: air pollution; mediator: odor identification; outcome: **CERAD subtest of visuo-construction performance**. Presented are indirect, direct, and total effects (β-estimates, 95%-confidence intervals (95%-CI), and p-values). All models were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide. [a]: Proportion mediated = mediated effect / (direct effect + mediated effect) x 100.

Appendix B: Selection of interaction analyses for further CERAD subtests.



Presented are β-estimates with corresponding 95%-confidence intervals (95%-CI) for the association between air pollution and impairment in the CERAD subtest "**Word list learning"** in subgroups defined by low (<median of total number of correctly identified Sniffin' Sticks) vs. high odor identification (≥median of total number of correctly identified Sniffin' Sticks) for an increase of 1 interquartile range (IQR) in air pollution. All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide.

Figure B1: Interaction analysis between air pollution and the CERAD subtest "**Word list learning"** with odor identification as a moderator (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).



Presented are β-estimates with corresponding 95%-confidence intervals (95%-CI) for the association between air pollution and impairment in the CERAD subtest of **visuo-construction performance** in subgroups defined by low (<median of total number of correctly identified Sniffin' Sticks) vs. high odor identification (≥median of total number of correctly identified Sniffin' Sticks) for an increase of 1 interquartile range (IQR) in air pollution. P-values are given for the interaction terms and defined at significant as 0.05 or lower. All associations were adjusted for age, educational level, body mass index (BMI), smoking status, living in an urban vs. rural area, cold, and hay fever. PM_{10} = particulate matter with an aerodynamic diameter of 10 µm or less; $PM_{2.5}$ = particulate matter with an aerodynamic diameter of 2.5 µm or less; $PM_{2.5}$abs = filter absorbance of particulate matter with an aerodynamic diameter of 2.5 µm or less; NO_2 = nitrogen dioxide.

Figure B2: Interaction analysis between air pollution and the CERAD subtest of **visuo-construction performance** with odor identification as a moderator (follow-up investigations in A) 2008/2009 (N = 733) and B) 2012/2013 (N = 542)).

7 Acknowledgments

I would like to express my thanks to Prof. Dr. Barbara Hoffmann for kindly providing the interesting topic and for her further suggestions whilst working on my analysis. I would also like to thank to Prof. Dr. Jean Krutmann as Scientific Manager and Dr. Alexander Beaucamp as the Economic and Legal Manager of the Leibniz Research Institute for Environmental Medicine for allowing the opportunity to perform this study at the IUF.

Special appreciation goes to Junior Prof. Dr. Tamara Schikowski for her active support from start to finish. Her enthusiasm, kindness, humanity, and foresight have contributed significantly to the success of this dissertation.

My special thanks go to Dr. Anke Hüls for her invaluable and motivating support, as well as for her comprehensive explanations regarding the statistical analysis of the data, and for the fact that her door and her ears were always open to my possible and impossible questions.

I am very grateful to the employees of the Research Group on Environmental Epidemiology of Lung, Brain, and Skin aging at the Leibniz Research Institute for Environmental Medicine for their support and advice. Prof. Dr. Ursula Krämer, Dr. Andrea Vierkötter, Sabine Stolz, Elke Link, and Dorothea Sugiri deserve special mention.

Many thanks also to the study participants for their consent to the use of their data.

Last but not least, I would like to thank my family and all my friends from the bottom of my heart.