Title: The association between ambient air pollution and cardiorespiratory health outcomes of adults residing in informal settlements of the Western Cape in a 12 month period

Mini Dissertation submitted in partial fulfilment of the requirements for the degree:

Masters of Public Health

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PREFACE

DECLARATION

I, TASNEEM ALLIE, hereby declare that the work on which this dissertation/thesis is based is my original work (except where acknowledgements indicate otherwise) and that neither the whole work nor any part of it has been, is being, or is to be submitted for another degree in this or any other university.

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Date: ……30/10/2024…….

Acknowledgements:

I would firstly like acknowledge my creator and give thanks for being blessed with the ability to further my studies.
A special thanks my supervisor and co-supervisor Professor Dalvie and Dr Olaniyan for their expert knowledge, advice, guidance and patience throughout the process.
I am grateful and thankful to my family, especially my parents AG and D. Allie, as well as my sister, brother, husband and daughter for their unwavering encouragement, support and motivation.

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PART A: JOURNAL MANUSCRIPT

PREPARATION FOR SUBMISSION

Journal: This manuscript has been prepared for the publication in the peer-reviewed journal Environmental Pollution. The author instructions have been adhered to apart from the cover page and co-author details which have been excluded from the dissertation.

Title: The association between ambient air pollution and cardiorespiratory health outcomes of adults residing in informal settlements of the Western Cape in a 12 month period.

Competing Interests Declaration: None

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Highlights

- What we know about the topic:
  - there is limited data on the association between ambient air pollution cardiorespiratory outcomes among adults in low-and middle income countries.

- What knowledge does the study add?

  Annual median concentration of $PM_{2,5}$ was 9.8 µg/m^3 and NO_2 was 16.86 µg/m^3, below the National Ambient Air Quality Standards (NAAQS) in informal settlements in the Western Cape

  There was a positive association between NO_2 and shortness of breath on exertion (OR:4.02, 95% CI:2.21-7.65) adjusting for $PM_{2,5}$ and other confounders.

  The concentration curve shows that there was an increase in the risk of self-reported shortness of breath at annual NO_2 levels above approximately 10 µg/m^3

- How does the current study impact on current guidelines?
  - The NAAQS should be more in keeping with the WHO Ambient Air Quality guidelines for NO_2 at 10 µg/m^3 and lower in low- and middle income countries where underlying susceptibility may increase the observed risk.

Acronym list

$PM_{2.5}$ = particulate matter with diameter of 2.5 microns

PM_{10} = particulate matter with diameter of 10 microns

NO_2 = nitrogen dioxide

$\mu g/m^3$ =microgram per cubic metre

DALY = disability adjusted life years

FEV_1 = forced - expiratory volume in 1 second

FVC = forced vital capacity

GIS = geographic information system

IQR = interquartile range

LMI = low- and middle-income countries

LUR = land-use regression

NAAQS = National Ambient Air Quality Standards

OR = odds ratio

PEF = peak expiratory flow

UCT = University of Cape Town

WHO = World Health Organization

Abstract

Background: There is limited data on the association between ambient air pollution cardiorespiratory outcomes among adults especially among populations with low socio-economic status in low- and middle income countries.

Objectives: A cross-sectional cohort study was conducted in four informal settlements of the Western Cape to determine the association between annual NO_2 and $PM_{2.5}$ air pollution exposure and cardiorespiratory outcomes in adults.

Methods: A questionnaire was administered to 506 participants who were guardians of learners in a primary study, to collect information on cardio-respiratory symptoms and relevant covariates. Land use regression modelling in R was used to estimate the annual average $PM_{2.5}$ and NO_2 exposure at each household. Cardio-respiratory outcomes included wheezing in the last 12 months, doctor diagnosed asthma, asthma symptoms score (calculated as the sum of positive responses to 3 asthma questions), chest pain, hypertension and hypercholesterolaemia. The quantile g computation package in R was used to assess the joint-effect of $PM_{2.5}$ and NO_2 on the outcome of interest. A concentration curve was determined, while adjusting for confounders (paraffin exposure, mould exposure, smoke exposure, low education, unemployed, sex and age).

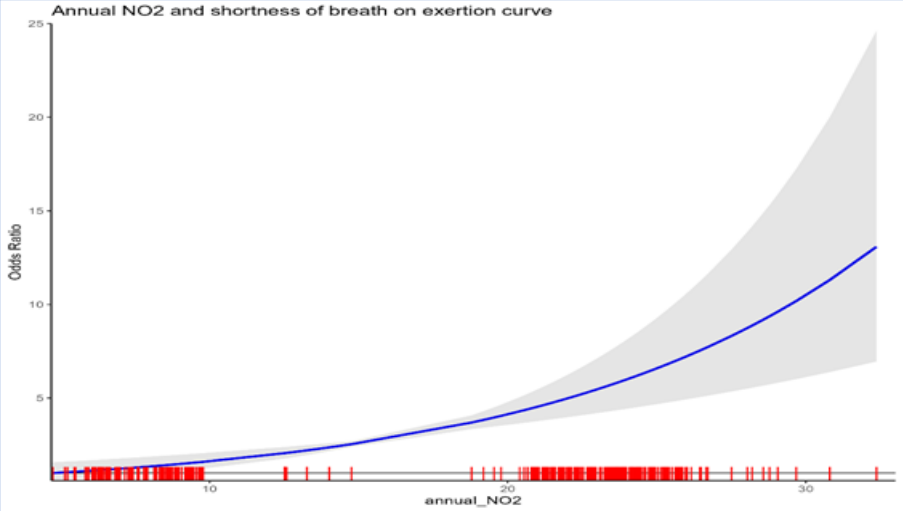
Results: Most of the participants were female (89.9%), with a median age of 40 (IQR=12.8). The annual median concentration of $PM_{2,5}$ 9.8 µg/m^3 and NO_2 was 16.86 µg/m^3, below the National Ambient Air Quality Standards (NAAQS) There was positive association between NO_2 and shortness of breath on exertion (OR:4.02, 95% CI:2.21-7.65) adjusting for $PM_{2,5}$ and other confounders. There were no other significant associations found between $PM_{2.5}$ and NO_2 levels and other cardiorespiratory outcomes including indoor air pollution. The concentration curve indicates an increased risk in self-reported shortness of breath on exertion with increasing NO_2 concentrations above 10µg/m^3.

Conclusion: The study found evidence of an association between annual NO_2 and shortness of breath on exertion. The study also provides policy-relevant information from the concentration-response curve in demonstrating elevated risk at a level four-times lower than the current National Ambient Air Quality Standard (NAAQS) of 40µg/m^3. This indicates that the NAAQS should be more in keeping with the WHO Ambient Air Quality guidelines for NO_2 at 10 µg/m^3 and lower in low- and middle-income countries where underlying susceptibility may increase the observed risk.

Keywords: air pollution, NO_2, $PM_{2.5}$, cardiorespiratory outcomes, low and middle-income countries, adults

Graphical Abstract

- Annual median concentration of $PM_{2,5}$ and NO_2 below the National Ambient Air Quality Standards (NAAQS) in informal settlements in the Western Cape ($PM_{2.5}$= 9.8 µg/m^3, NO_2= 16.86 µg/m^3)
  positive association between NO_2 and shortness of breath on exertion (OR:4.02, 95% CI:2.21-7.65) adjusting for $PM_{2,5}$ and other confounders
- 89.9% female
- Median age 40
- the concentration curve indicates an increased risk of self-reported shortness of breath on exertion with increasing NO_2 concentrations above 10µg/m^3



1. Introduction

Cardiorespiratory mortality makes up the top 4 causes of morbidity in 2019 worldwide and it ranks within the top 10 causes of disability adjusted life-years according to the WHO. (WHO, 2024) In 2019, 8.9 million (16%) deaths were attributed to Ischaemic Heart Disease alone making it the top cause of death, with lower respiratory tract infections ranking as the highest communicable cause of death in low-, middle- and high-income countries. Chronic obstructive pulmonary disease accounting for 6% of global mortality, more so in high income countries. Respiratory system cancers also rank in 6th place causing approximately 1.8 million deaths worldwide in 2019. (WHO, 2024)

Cardiovascular mortality accounts for approximately 60 – 80% of deaths due to air pollution (Boudrel et al, 2017). According to the WHO, about 20 % of cardiovascular deaths, 16% of lung cancer deaths, 26% lower respiratory tract infection deaths and 25% of chronic obstructive pulmonary disease (COPD) deaths are caused by air pollution. Globally, an estimated 7 million deaths occurred in adults and children in 2012 due to cardiorespiratory causes. Approximately 36% of these deaths were attributable to ischaemic heart disease, 33% due to cerebrovascular events, 17% chronic obstructive pulmonary disease, 8% acute lower respiratory tract infections and 6% lung cancer. About 75% of these deaths occur in low to middle income countries (WHO, 2018). Long term exposure to ambient air pollution may be associated with an elevated blood pressure, increased risk of cardiovascular morbidity and mortality (Hoek et al, 2013).

A search was conducted in PUBMED, Google scholar and Science Direct using keywords “ambient air pollution, cardiorespiratory, cardiovascular, hypertension, respiratory, particulate matter and nitrogen dioxide.” Studies conducted among adults from both urban and rural areas were included from 2014 - 2023. Although several epidemiological studies have been conducted investigating the association between long term ambient air pollution exposure and cardiorespiratory outcomes, (Chen et al 2015, Cai et al 2015, Crouse et al 2015, Wu et al 2016, Hartiala et al 2016, Pitchika et al 2017, Kim et al 2017, Weichenthal et al 2017, Cai et al 2017, Bai et al 2019, Yang et al 2019, Arku et al 2020, Liu et al 2021, Olaniyan et al 2022), the results are inconsistent, varies in outcome and exposure instruments used, with limited data in low- and middle income countries studied and on the joined effect of air pollutants.

According to Wichmann et al, 2012 a study conducted within the private sector in Cape Town, demonstrated an associated increase in cardiovascular and respiratory mortality with exposure to ambient air pollution. Cardiovascular mortality increased 3% with each increased interquartile range (IQR) of NO_2 and SO_2. However,

a study by Adebayo-Ojo et al, 2022 did not find a significant association between NO_2 and cardiovascular admissions for respiratory disease in private hospitals in Cape Town. A study by Bagula et al,(2021) conducted on adults in informal settlements in South Africa, found an association between annual ambient $PM_{2.5}$ and self-reported chest pain at levels below the South African National Ambient Air Quality Standards.

Particulate matter (PM) is made up of a mixture of solid and liquid particles found in the air. PM varies in particle size, with coarse particulate matter depicted as PM_{10} and fine particulate matter depicted as $PM_{2.5}$. Its constituents are from a range of organic natural and chemicals, metals such as copper, iron, potassium, nickel, sulphur, silicon, vanadium, and zinc as well as acids. Within the South African context, $PM_{2.5}$ was best predicted by a combination of the population density and presence of grills and waste burning sites within the studied area. (Saucy A et al, 2018). Particulate matter has been shown to affect cardiorespiratory morbidity and long-term exposure has been found to be associated with an 11% (95% CI 5, 16%) increase in cardiovascular mortality and 3% (95% CI −6, 13%) increase in mortality from non-malignant respiratory diseases (Hoek et al, 2013).

Exposure to $PM_{2.5}$ in the short term was associated with a 0·47% (95% CI 0·34 – 0·61) increase in cardiovascular mortality and a 0·57% (0·28 – 0·86) increase in respiratory mortality for each 10 $\mu g/m^3$ increase in same-day $PM_{2\cdot5}$. (Newell K, et al 2017)

One of the gaseous air pollutants which occur through the oxidation of nitrogen include nitrogen dioxide, nitric oxide, ozone, volatile organic compounds and carbon monoxide. Phytochemical reactions with ammonia often from agriculture react with nitrogen dioxide, which is toxic, to form particles that further contribute to inhaled air pollution. In the South African context and specifically within the informal settlement areas studied, NO_2 levels were best predicted by the presence of traffic related exposure within close proximity to the geographical region. (Saucy A et al, 2018) Both short- and long-term exposure to nitrogen dioxide has been found to increase cardiovascular mortality (Hoek et al, 2013) and exposure has been positively associated with COPD mortality relative risks (RR) = 1.03 (1.01 - 1.04) per 10 $\mu g/m^3$ (Huangfu et al, 2020)

This study investigated the association between 12 months of exposure to ambient air pollution ($PM_{2.5}$ and NO_2) and self – reported cardiorespiratory outcomes in adults in informal settlements within the Western Cape.

2. Methodology

2.1 Study design and sampling

The main study was a 12-month cohort study conducted on school children and their parents residing in informal settlements of 4 areas in the Western Cape including an urbanised industrialised area in Milnerton, Marconi-Beam; a peri-urbanised area namely Khayelitsha, a semi-arid rural area, Oudtshoorn and Masiphumele in Noordhoek, an urban non-industrialised area. The baseline study was conducted from February 2015 to September 2015, and the follow – up study from February 2016 to September 2016. This study is a cross-sectional analysis at 12-month follow-up of the parents of the children investigating the association between air pollution exposure and cardiorespiratory outcomes in adults.

The sampling was of participants is described elsewhere. (Olaniyan et al, 2017). In total, 506 guardians of students in the primary study were interviewed via questionnaires. Recruitment of the students took place when the Western Cape Education department provided a list of schools within the study areas. One or two schools closest to the City of Cape Town air monitoring stations were selected. The caregivers of school students recruited in the primary study were included in this study. Investigators met with the principal of grade 4 students in the study areas and obtained home addresses of these students. Grade 4 students were chosen as they would remain at the same school for the study period. Trained field investigators visited the homes to obtain consent for the students' participation as well as that of the caregivers in the current study. About 150 students were selected by random sampling from each area in schools where the number who consented exceeded this number. Those whose guardians consented were included in the study. Children were excluded if they had an operation in the last 12 months; if the child had vomiting or pain; if they were receiving tuberculosis treatment, if they had epilepsy or a respiratory infection in the prior 3 weeks.

2.2 Data Collection

2.2.1 Questionnaire

Air pollution measurements were done over a period of one year during 2015 -2016 prior to questionnaires being administered in the follow up study. Trained field staff conducted interviews at the homes of the guardian or parents of the children. Interviews were conducted in participants home language, either English, isiXhosa or Afrikaans. Capturing of the results was done on mobile phones. Items included in the questionnaire (Appendix 1) were: demographic characteristics, respiratory symptom history and health, cardiovascular disease, blood pressure, presence of hypercholesterolemia (diagnosed by a doctor, an explanation was also given) , occupational exposure, indoor pollutant exposure, outdoor pollutant exposure, physical activity history, stressors and smoking status and history. (Table 2) The questionnaires were based on a validated

questionnaire with questions included from the European Community Respiratory Health Survey (Jarvis et al, 2002) and the National Health and Nutrition Examination Survey Questionnaire (Curtin et al, 2013).

2.2.2 Air pollution measurement

Land use regression (LUR) models in R were developed for the primary study between 2015 – 2016 in which the annual average outdoor concentration of $PM_{2.5}$ and NO_2 were estimated at the home addresses of each participant (Saucy et al, 2018). The estimated annual air pollution concentrations were based on weekly measurements of $PM_{2,5}$ and NO_2 over a period of 1 year at 140 of the children's homes including 20 in Masiphumelele and 40 in each of the other study areas. Seasonal and annual averages of concentrations of air pollution were determined incorporating predictors of exposure such as the number of household members, major traffic sources close to the home (such as bus routes, train stations and highways), waste burning sites. Spatial variation in annual average concentrations that was determined by land – use from a geographic information system (GIS). The annual average concentration estimates were done prior to the questionnaires being administered on the participants in this follow up study.

2.3 Data Analysis

2.3.1 Outcome variable

Asthma outcome variables included individual asthma questions (dichotomous) and an asthma symptom score. The individual asthma questions included: "wheezing or whistling in the chest in the last 12 months" and "doctor confirmed asthma". The asthma symptom score was calculated as the sum of positive responses to the questions (1 point for a positive response):

- woken up with a feeling of tightness in the chest at any time in the last 12 months?,
- woken up by shortness of breath in the last 12 months?,
- woken by heavy coughing at any time in the last 12 months?

The 3 dichotomous cardiovascular outcomes (Yes/No) variables included the following :

- Self – reported chest pain: "Do you get pain or discomfort in your chest?",
- self- reported hypertension: "Have you ever been told by a doctor or other health professional that you had hypertension (hy-per-tenshun), also called high blood pressure?" and
- self – reported hypercholesterolaemia: "Did a doctor tell you that your blood cholesterol level was high?"

2.3.2 Statistical Analysis

Descriptive statistics was used to examine the characteristics of the study population, cardiorespiratory outcomes and air pollution variables. Simple regression analysis was implored to determine the associations between various potential confounders and cardiorespiratory outcomes. R version 4.3.1 was used to perform multiple logistic regression analysis for the association between ambient air pollution exposure ($PM_{2.5}$ and

NO_2) and cardiorespiratory outcomes controlling for confounders. Confounders adjusted for included those known in the literature and assessed by Direct Acyclic Graphs (DAG) and/or those statistically significantly associated in the bivariate analysis (the confounders included age, gender, smoke exposure, mould exposure, paraffin exposure, low education and being unemployed). A subset analysis was done on the female participants as the study population consisted of 89.9% female participants and too few males. The quantile g computation package in R was used to assess the joint-effect of $PM_{2.5}$ and NO_2 on the outcome of interest. A concentration curve was determined while adjusting for confounders (paraffin exposure, mould exposure, smoke exposure, low education, unemployed, sex and age). The two-pollutant model assessed the independent association of one-pollutant i.e., $PM_{2.5}$, while adjusting for the other pollutant i.e. NO_2. The joint-effect assessed both the contribution of the 2 pollutants ($PM_{2.5}$ and NO_2) on the outcome, simultaneously. For example, the two-pollutant model is the probability of an outcome due to $PM_{2.5}$ on NO_2, while the joint effect is the probability of an outcome due to $PM_{2.5}$ and NO_2. The model adjusted for all the variables in the logistic regression model in the main analysis. The counterfactual (i.e. theoretical minimum) was defined as the minimum assigned annual NO_2 exposure in the study. There was no simulation done due to the small size and thus the model included the study participants in the study. The concentration-response curve used the fitted predicted risk across the observed exposure levels in the study. The lowest observed exposure level was used as the counterfactual, from which the subsequent exposure-response relationship were interpolated.

2.4 Ethical consideration

The primary study received approval by the University of Cape Town's Research Ethics Committee (ethics number: 697/2014) and this sub-study received ethics approval with number: HREC REF 769/2023 from the same committee.

3. Results

3.1 Demographics Information

The median age across the study population was 40.3 years and was slightly higher in Khayelitsha and Oudtshoorn compared to the other two areas (Table 1). Female participants accounted for 89.1% of the study population overall and this was similar for all areas. isiXhosa was the most spoken language across all study areas except Oudtshoorn, where Afrikaans was the majority at 79.3%. English is least spoken in all informal settlements. 14.6% of the participants only completed primary school, while 85.4% completed high school. More than 80% of the study participants did not report to be physically active in all the areas. Almost a third of the participants in all areas were exposed to second-hand smoke as they reported a smoker in their home apart from Marconi Beam where the prevalence of second-hand smoke was about 16%. The prevalence of mould exposure varied between 17.5% in Masiphumelele and 3% in Marconi-Beam. Paraffin use varied in the different areas with the highest reported paraffin use in Marconi-Beam (75.2%) and the lowest reported in Khayelitsha (28.4%). Approximately half of the study population reported being unemployed (47.4%), with the highest unemployment prevalence in Khayelitsha (71.65) and Masiphumelele (71.8%) (Table1) and lower levels of employment in Marconi-Beam (47.3%) and Oudtshoorn (35.2%)

Table 1: Demographics and selected other characteristics of the study population in the four informal settlements within the Western Cape

	Khayelitsha	**Oudtshoorn**	**Marconi-Beam**	**Masiphumelele**	**Total Areas**
	n=109 (%)	n=165 (%)	n=129 (%)	n-103 (%)	n=506(%)
Age median (IQR)	42 (14)	43 (14)	38 (12)	38 (10)	40.3 (12.8)
Gender					
- **male**	17(15.6)	9(5.5)	12(9.3)	13(12.6)	51(10.1)
- **female**	92(84.4)	156(94.5)	117(90.7)	90(87.4)	455(89.9)
Language					
- **isiXhosa**	107(98.1)	34(20.6)	112(86.8)	98(95.1)	351(69.4)
- **English/Afrikaans/other**	2(1.9)	131(79.3)	17(13.1)	5(4.9)	155(30.6)
Education Level					
- **low education: primary school**	13 (11.9)	42(25.5)	9(7)	10(9.7)	74(14.6)
- **higher education**	96(88.1)	123(74.5)	120(93)	93(90.3)	432(85.4)
No Physical activity	88(80.7)	140(84.8)	126(97.7)	102(99)	456(90.1)
Smoker in the house	35(32.1)	53(32.1)	21(16.3)	33(32)	142(28.1)
Mould exposure	8(7.3)	19(11.5)	4(3.1)	18(17.5)	49(9.7)
Paraffin use at home	31(28.4)	87(52.7)	97(75.2)	64(62.1)	279(69.7)
Employed	78(71.6)	58(35.2)	61(47.3)	74(71.8)	271(53.6)

3.2 Air Pollution levels

The highest annual concentration of $PM_{2.5}$ and NO_2 was found in Oudtshoorn (62.9 µg/m^3) and Khayelitsha (39.9 µg/m^3) respectively. The lowest annual concentrations of both $PM_{2.5}$ and NO_2 were found in Oudtshoorn (0.75 µg/m^3 and 0.13). The median annual levels of $PM_{2.5}$ and NO_2 in all study areas (9.8 µg/m^3 and 16.86 µg/m^{3}respectively) was above the latest WHO air quality guideline (2021) of 5 µg/m^3 for $PM_{2.5}$ and 10 µg/m^3 for NO_2, which is below the South African Ambient Air Quality Standards for NO_2 of 40µg/m^3 (WHO, 2024). (figure 1a and 1b)

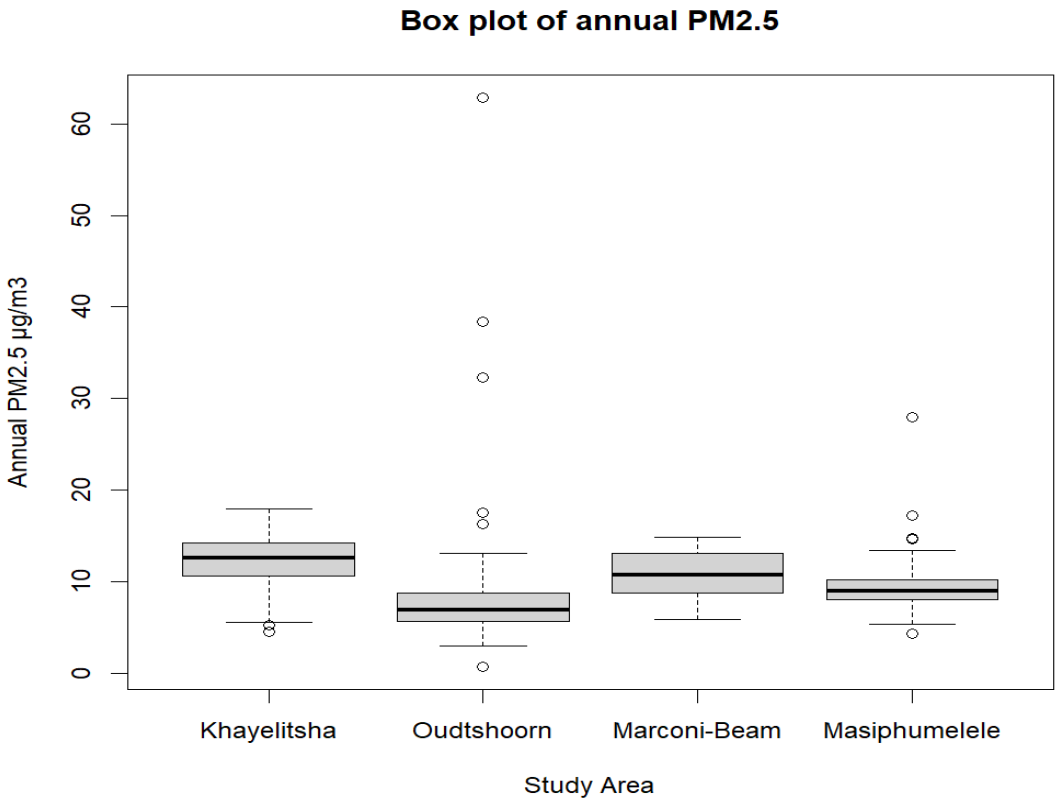


Figure 1a: The distribution of $PM_{2.5}$ across the four informal settlements in the Western Cape ($PM_{2.5}$ IQR = 4.96 µg/m^3)

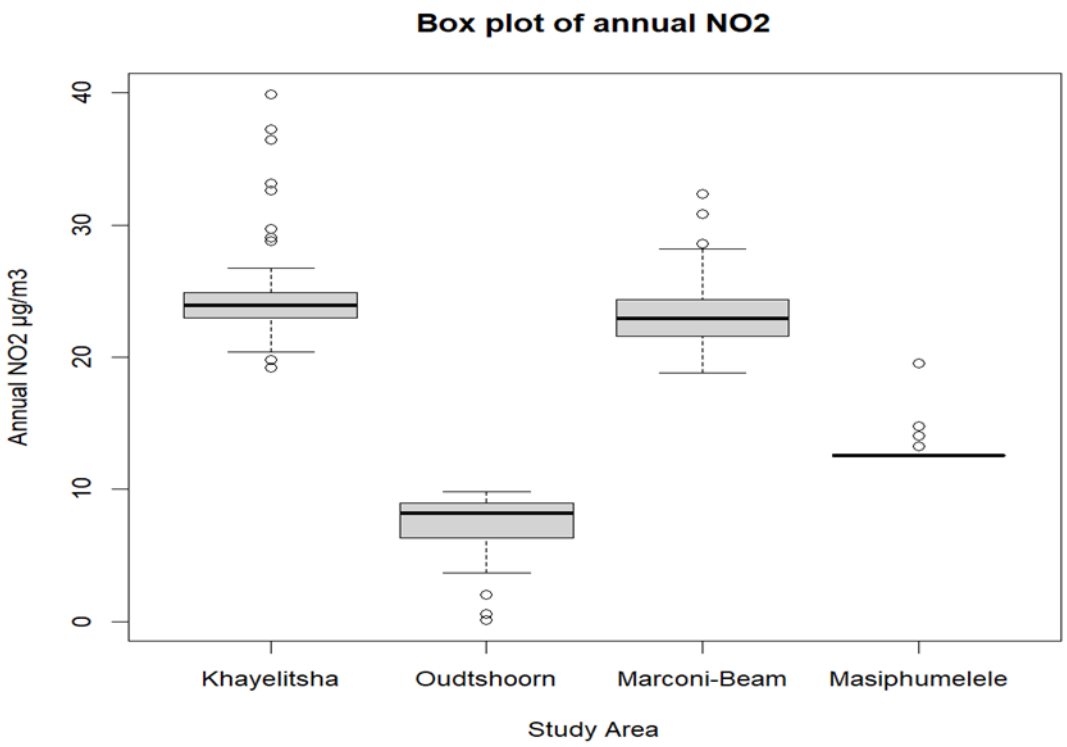


Figure 1b: The distribution of NO_2 across the four informal settlements in the Western Cape (NO_2 IQR = 14.52 µg/m^3)

3.3 Cardiorespiratory Outcomes

Table 1 summarises the cardiorespiratory health of the participants. Self- reported asthma symptom prevalences in all areas varied between 1% -12.5 %. About 6% (5.5%) of participants across the study population had an asthma symptom score >= 2., with the highest prevalence in Khayelitsha at (8.3%) and lowest in Oudtshoorn at (2.4%). The prevalence of self – reported chest pain among participants was 7.5%, with the highest prevalence reported in Masiphumelele (11.7%) and the lowest in Oudtshoorn (2.4%). Self - reported hypertension had the highest prevalence amongst cardiorespiratory outcomes (17%) across the study population with the highest prevalence in Oudtshoorn (23%) and the lowest in Marconi-Beam at (10.9%). Less than 2% of participants reported hypercholesterolemia (1.9%) with the highest prevalence in Khayelitsha at (5.5%) and the lowest in Oudtshoorn (0.6%).

Table 2: Cardiorespiratory outcomes of participants in the four study areas

	Khayelitsha	Oudtshoorn	Marconi-Beam	Masiphumelele	Total
	n=109 (%)	**n=165(%)**	**n=129(%)**	**n=103(%)**	**n=506(%)**
Have you had wheezing or whistling in your chest at any time in the last 12 months?	3(2.8)	4(2.4)	3(2.3)	2(1.9)	12(2.4)
Have you been out of breath when the wheezing noise was present?*	2(1.8)	4(2.4)	1(0.7)	2(1.9)	9(1.8)
Do you have to shortness of breath when walking at your own pace on level ground?	1(0.9)	4(2.4)	0(0)	0(0)	5(1)
In the last 12 months, have you had shortness of breath after heavy exercise or work?	17(15.6)	4(2.4)	31(24)	13(12.6)	65(12.8)
Was your asthma confirmed by a doctor?	3(2.6)	4(2.4)	2(1.6)	2(1.9)	11(2.2)
Are you currently taking any medicines including inhalers, aerosols or tablets for asthma	2(1.8)	4(2.4)	2(1.6)	1(0.9)	9(1.8)
Have you woken up with a feeling of tightness in your chest at any time in the last 12 months?*	10(9.2)	4(2.4)	6(4.7)	6(5.8)	26(5.1)
In the last 12 months, have you been woken-up by shortness of breath?*	6(5.5)	3(1.8)	5(3.9)	6(5.8)	20(4)
Have you been woken by heavy coughing at any time in the last 12 months?	2(1.8)	3(1.8)	5(3.9)	4(3.9)	14(2.8)
Asthma symptom score:#					
>=2	9(8.3)	4(2.4)	8(6.1)	7(6.8)	28(5.5)
self - reported chest pain	9(8.3)	4(2.4)	13(10.1)	12(11.7)	38(7.5)
self - reported hypertension	19(17.4)	38(23)	14(10.9)	15(14.6)	86(17)
self - reported hypercholesterolaemia	6(5.5)	1(0.6)	1(0.8)	2(1.9)	10(1.9)

#Asthma symptom is score calculated as the sum of positive responses to these questions and one point was given for any of the questions with an asterisk *

3.4 Association between cardiorespiratory outcomes and and indoor exposure and sociodemographic factors

Bivariate analysis to determine the association between cardiorespiratory outcomes and indoor exposure and sociodemographic factors at follow-up found age and unemployment to be significantly positively associated covariates. ($p<0.05$) (Table S1). As indicated before, the adjusted model to assess the association between air pollutants and cardiorespiratory outcomes included paraffin exposure, mould exposure, smoke exposure, low education and unemployed (Figure S1) as determined by the direct acyclic graph (DAG). Mould exposure, smoke exposure and paraffin use at home that were used as markers for indoor air pollution exposure were found to be not significantly associated with any outcomes in the bivariate analysis. (Table S1)

3.5 Associations between air pollution and cardiorespiratory outcomes

In the adjusted single-pollutant logistic regression model for the association between $PM_{2.5}$ and NO_2 ($\mu g/m^3$) and self-reported cardio-respiratory outcomes (Table 3) there was a significant positive association between NO_2 and shortness of breath on exertion (OR: 3.86, IQR: 2.21-6.97) and with reported chest pain (OR: 1.94, IQR: 1.00-3.85). In the adjusted two pollutant model (Table 4), there was a slightly increased significant positive association between NO_2 and self-reported shortness of breath on exertion (OR: 4.02, IQR: 2.21-7.65) compared to the single pollutant model. There were no other significant or near significant associations found between $PM_{2.5}$ and NO_2 levels and cardio-respiratory outcomes. The upper limit of NO_2 was approximately 35 ($\mu g/m^3$).

Sensitivity analysis (Table S2) excluding males in the multivariate analysis found a significant positive association between NO_2 and shortness of breath on exertion (OR: 3.45, IQR: 2.05-6.02) in the adjusted single pollutant model. A subset analysis was done on females due to the majority of the study population being female (89.9%) and few male participants. This subset analysis was compared with the overall results. The associations among the female subset analysis were slightly reduced, but like the finding which included both genders.

The cohort design was retained because a longitudinal analysis was conducted, but the number of new cases of outcomes were too few to determine an association

Table 3: Single-pollutant logistic regression model for the association between $PM_{2.5}$ and NO_2 ($\mu g/m^3$) and self-reported cardiorespiratory outcomes among adults in four informal settlements of the Western Cape of South Africa

	$PM_{2.5}$		**NO_2**	
	OR 95% CI			
Outcomes	**unadjusted**	**adjusted**	**unadjusted**	**adjusted**
Asthma Symptoms score >= 2 (n=28)	1.17 (0.79-1.56)	1.21 (0.80-1.64)	1.87 (0.91- 3.93)	1.94 (0.90-4.27)
Chest pain (n=38)	1.16 (0.82- 1.52)	1.14 (0.79-1.52)	**1.89 (1.01- 3.60)**	**1.94 (1.00-3.85)**
Hypercholesterolaemia (n=10)	1.17 (0.59- 1.69)	1.20 (0.58-1.86)	2.92 (0.88- 10.51)	3.17 (0.94-11.42)
Doctor-diagnosed asthma (n=12)	1.13 (0.56- 1.64)	1.23 (0.62-1.79)	0.85 (0.26- 2.64)	1.11 (0.33- 3.67)
Wheezing (n=12)	1.17 (0.63- 1.66)	1.29 (0.69-1.87)	1.02 (0.34- 3.03)	1.20 (0.38-3.79)
Shortness of breath on exertion (n=65)	1.18 (0.91- 1.51)	1.18 (0.89-1.52)	**3.45 (2.05- 6.02)**	**3.86 (2.21-6.97)**

Adjusted model includes age, sex, paraffin exposure, mould exposure, smoke exposure, low education, unemployed
Bold text denotes significant at P <0.05
Risk per IQR increase: $PM_{2.5}$ IQR = 4.96 $\mu g/m^3$, NO_2 IQR = 14.52 $\mu g/m^3$

Table 4: Two pollutant logistic regression model and joint-effect for the association between $PM_{2.5}$ and NO_2 ($\mu g/m^3$) and self - reported cardiorespiratory outcomes among adults in four informal settlements of the Western Cape Province of South Africa

	$PM_{2.5}$		NO_2		$PM_{2.5}$ + NO_2
	OR 95% CI				
Outcomes	**unadjusted**	**adjusted**	**unadjusted**	**adjusted**	**Joint-effect**
Asthma Symptoms score >= 2	1.11	1.14	1.78	1.81	1.33
(n=28)	(0.67-1.53)	(0.66-1.61)	(0.85-3.83)	(0.83-4.14)	(0.84-2.13)
Chest pain	1.09	1.06	1.82	1.89	1.36
(n=38)	(0.69-1.47)	(0.65-1.47)	(0.95-3.56)	(0.95-3.87)	(0.92-2.01)
Hypercholesterolaemia	1.08	1.00	2.83	3.17	1.29
(n=10)	(0.40-1.74)	(0.37-1.87)	(0.83-10.52)	(0.88-11.86)	(0.59-2.85)
Doctor-diagnosed asthma	1.15	1.23	0.80	1.01	1.04
(n=12)	(0.56-1.64)	(0.58-1.79)	(0.24-2.54)	(0.30-3.41)	(0.51-2.13)
Wheezing	1.17	1.23	0.95	1.01	1.04
(n=12)	(0.60-1.66)	(0.58-1.79)	(0.31-2.88)	(0.30-3.41)	(0.51- 2.13)
Shortness of breath on exertion	0.99	0.93	**3.47**	**4.02**	**1.92**
(n=65)	(0.65-1.35)	(0.58-1.33)	**(2.00-6.28)**	**(2.21-7.65)**	**(1.39-2.66)**

adjusted model includes: age, sex, paraffin exposure, mould exposure, smoke exposure, low education, unemployed
Bold text denotes significant at P <0.05
Risk per IQR increase: $PM_{2.5}$ IQR = 4.96 µg/m^3 , NO_2 IQR = 14.52 µg/m^3

Figure 2 shows that the concentration curve suggests an increase in the risk of self-reported shortness of breath on exertion at NO_2 concentrations above 15 µg/m^3. There are too few values in the upper range. It is therefore unclear if this supports an exponential curve.

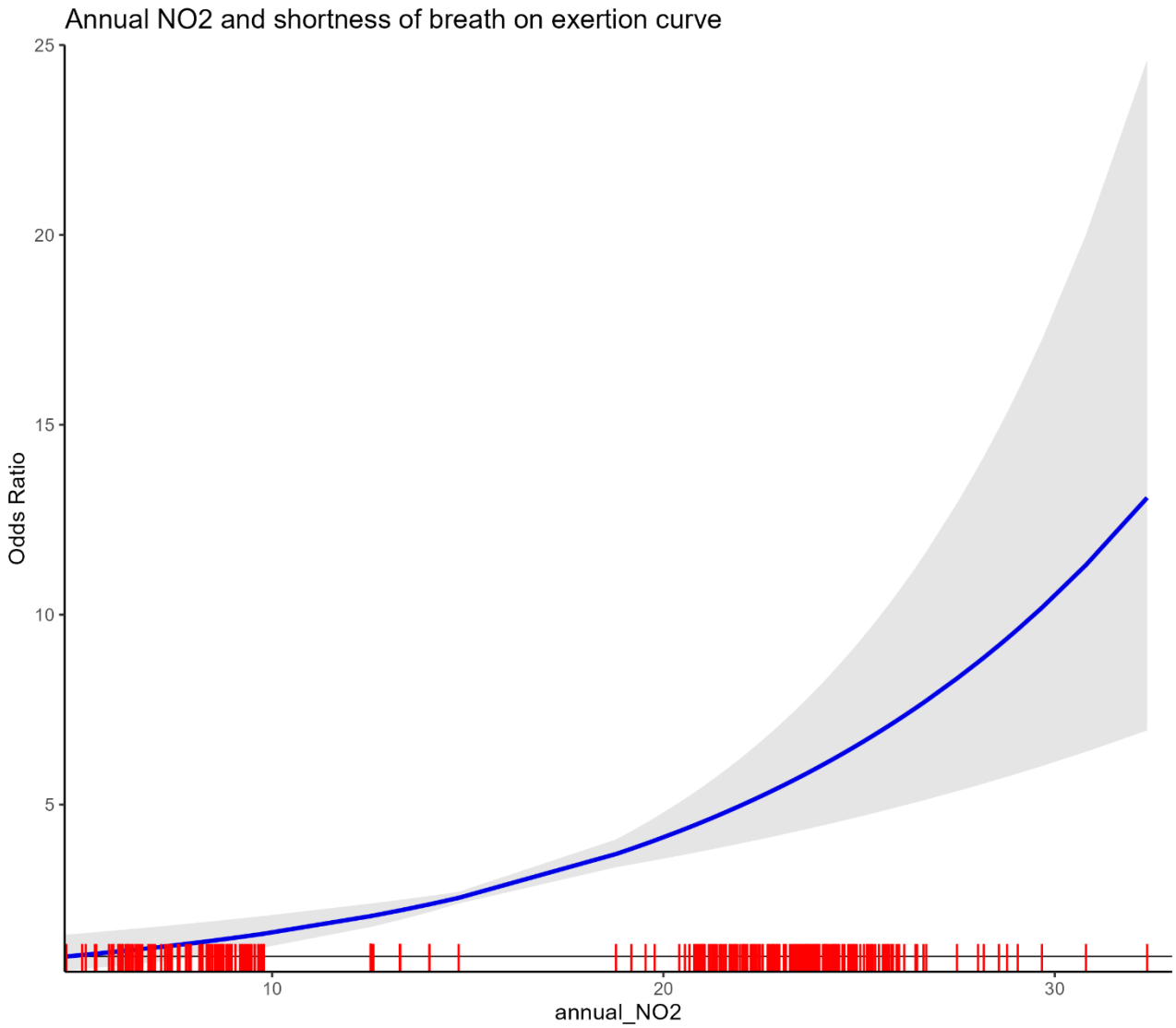


Figure 2: Concentration curve of the relationship between annual NO_2 and shortness of breath on exertion

4. Discussion

This study found a consistent statistically significant positive association between NO_2 and self-reported shortness of breath on exertion. In the two-pollutant model when taking $PM_{2.5}$ into account, there was an increased risk of self – reported shortness of breath on exertion for each IQR increase in NO_2 of 14.52 µg/m^3. Albeit slightly attenuated, the risk remained at an OR of 1.92 following the assessment of the joint effect of NO_2 and $PM_{2.5.}$ Using the quantile g – computation model the concentration response curve shows that with an increase in annual NO_2 levels above approximately 10 µg/m^3 there was an increase in the risk of self-reported shortness of breath. These associations were observed to be stronger among females in a subset analyses. No other positive associations were found between annual

NO_2 exposure and reported cardiorespiratory outcomes nor between annual $PM_{2.5}$ exposure and any cardio-respiratory outcomes. The positive association observed between NO_2, and shortness of breath is consistent with four previous studies identified that had investigated the association between NO_2 and cardio-respiratory outcomes in adults (Wang et al, 2023, Wang et 2019, Brunekreef et al 2021, Adam et.al. 2015). However, comparisons to the current study are difficult due to varying outcomes measures. For example, the outcomes in previous studies included mortality and hospital admissions, self-reported respiratory illness, CT scans for emphysema and spirometry as well as lung function. Interestingly, the study by Brunekreef et al 2021 found cardiorespiratory mortality relating to Acute Coronary Syndrome asthma and COPD were associated with NO_2, and not associated with $PM_{2.5}$ in their two-pollutant model with low concentrations of NO_2 and $PM_{2.5}$ of less than that specified by the EPA, National Ambient Air Quality Standards (NAAQS) and World Health Organization (WHO) Air Quality Guideline. Another study by Adebayo-Ojo et al (2022), did not find a significant association between NO_2 and cardiovascular admissions for respiratory disease to private hospitals in Cape Town. However, in a study by Adam et al (2015), long term exposure to NO_2 was associated with decreased lung function (FEV_1 and FVC) with every 10µg/m^3 increase in NO_2.

The biological plausibility underpinning the association of long-term exposure to NO_2, and poor cardiorespiratory outcome may be due to the fact that NO_2 may result in a decrease in immune cells such as lymphocytes and increase inflammation in the airways (Lee et al, 2021). A decrease in haemoglobin and haematocrit has also been found by Framptom et al, 2002 and direct injury from NO_2 to epithelial cells. Reactive nitrogen species (RNS) which originates from environmental exposure and inflammatory cells such as macrophages, eosinophils, and neutrophils, interacts with reactive oxygen species (ROS) which are derived from the process of mitochondrial respiration, the xanthine oxidase system and nicotinamide adenine dinucleotide phosphate (NADPH) oxidase. The consequence of this exposure causes an overwhelming oxidative imbalance resulting in asthma symptoms (Liu K et al, 2022). NO_2 inhalation has also been found to trigger allergen sensitization and the alteration of gene expression in the antioxidant immune defence pathway (Gruzieva et al, 2017).

Although long term $PM_{2.5}$ was not found to be associated with respiratory outcomes in this study, Ai et al, 2019 found $PM_{2.5}$ to be a risk factor for asthma development. There is also limited evidence on the association between NO_2 and $PM_{2.5}$ with cardiovascular outcomes. There were no previous studies conducted in Africa apart from the positive association between $PM_{2.5}$ and chest pain reported in the baseline study (Bagula et al 2021). In this follow-up study, there was no significant positive association found between $PM_{2.5}$ and chest pain. It should be noted that the outcome of "shortness of breath on exertion" was not explored in the baseline study in isolation, as was done in the follow up study. Furthermore, a longitudinal analysis between air pollution levels and cardio-respiratory outcomes was not possible due to insufficient number of new cases in the follow-up study. However, three studies outside Africa found positive associations between NO_2 and/or PM or ambient air pollution exposure and cardiovascular outcomes. Weichenthal et al, 2017, found an increased risk of admissions to hospitals for myocardial infarction with increasing

$PM_{2.5}$ amongst participants aged 65 years and older, but no association was found in participants younger than 65 years. Self-reported physician diagnosed cardiovascular disease was found to be associated with long term exposure to NO_2 in a study by Yang et al, 2019, and long-term ambient air pollution exposure was positively associated with cardiovascular outcomes by Kim et al, 2017.

Also worth noting are the air pollution levels in the current study. The highest average annual $PM_{2.5}$ and NO_2 levels was found in Khayelitsha, a peri-urban areas (12.2µg/m^3 and 24.5µg/m^3 respectively) and the lowest annual $PM_{2.5}$ and NO_2 levels were observed in Oudtshoorn, a rural area with limited traffic exposure (8.11µg/m^3 and 7.6µg/m^3). The low annual $PM_{2.5}$ and NO_2 levels in Oudtshoorn is consistent with that of rural areas in low- and middle-income countries according to the World Health Organization ambient air quality database update 2021 version 6.1. ($PM_{2.5}$ concentration and NO_2 concentration column H and I: $PM_{2.5}$: 2.7 µg/m^3 – 29.1 µg/m^3, NO_2: 1.1 – 34.4)

The current study is strengthened analytically by the ability to assess the concentration-response curve and the joint effect of both NO_2 and $PM_{2.5}$. The concentration-response curve aided the ability to provide policy-relevant indications of the risk associated with various levels of air pollution. For example, in the current study, after the WHO annual NO_2 guideline levels of 10µg/m^3, there was an increase in the risk of self-reported of shortness of breath. The concentration curve suggests an increase in the risk of self-reported shortness of breath on exertion at NO_2 concentrations above 10 µg/m^3. This risk was observed at a level four-times lower than the current South African National Ambient Air Quality Standard (NAAQS) of 40µg/m^3 (National Environmental Management: Air Quality Act 39 OF 2004). Thus, such a concentration-curve is relevant for local policy on NO_2 regulation in South Africa. Similarly, the current study was able to estimate the joint effect of both NO_2 and $PM_{2.5}$ using nuanced analytical approaches such as the quantile g-computation. This allowed the estimation of the combined effect from both NO_2 and $PM_{2.5}$ as opposed to a two-pollutant model that would have controlled for the effect of the other pollutant.

Nonetheless, there are some limitations to the current study that should be highlighted. The self-reporting of symptoms may have led to outcome misclassification and recall bias in participants as outcomes were not objectively measured via spirometry to determine shortness of breath at rest or on exertion. Participants may be more likely to over-report symptoms if they are aware of the potential association with air pollution, and conversely less likely to report their symptoms due to recall bias. We hypothesized the latter outcome misclassification to be potentially non-differential, and if at all would skew the reported association would be drawn towards the null. Further studies could include possible at home or tested peak flow measurements for increased accuracy. Certain terms such as the question regarding hypercholesterolaemia may not have been understood by participants or not known since it requires a clinical test to be done to know if one has the condition, as it may remain asymptomatic. This may have also been misclassified and under – reported.

Likewise, one cannot deduce causality based on the cross-sectional study design. As indicated before, there was an insufficient number of new cases in the follow-up study for a longitudinal analysis. The lack of an association between

$PM_{2.5}$ and cardiorespiratory outcomes may have been due to the low spatial variability and contrast with an IQR for $PM_{2.5}$ of 4.96 µg/m^3 in the four study areas, which resulted from the land use -regression model's low explanatory power of $R^2 \approx 29\%$ (Saucy et al, 2018). Individuals may have had different time - activity exposure levels as they did not all follow the same daily pattern as that of the school children in the baseline and with half the study population being employed, significant travel or exposure in areas outside of estimated study areas may have occurred. Thus, there is potential for exposure misclassification due to the use of air pollution estimates at residential addresses. It should also be noted that the limited proportion of male participants precludes generalizability as males potentially have different covariate exposures as compared to females and may have had more significant findings with regard to $PM_{2.5}$ exposure as demonstrated in Ai et al, 2019.

5. Conclusion

This study found evidence of an association between long term annual NO_2 exposure and shortness of breath at mean levels of 17.03 µg/m^3. The study provides policy-relevant information from the concentration-response curve in demonstrating elevated risk at a level four-times lower than the current South African National Ambient Air Quality Standard (NAAQS) of 40µg/m^3. This indicates that the NAAQS should be more in keeping with the WHO Ambient Air Quality guidelines (2021) for NO_2 at 10 µg/m^3 and lower in low- and middle-income countries where underlying susceptibility may increase the observed risk.

Strategies may be implemented to reduce vehicular environmental exposure for adults such as improving or encouraging alternative transport modes or the use of electric vehicles to decrease vehicle emissions.

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6. References

1. Adam M, Schikowski T, Carsin AE, Cai Y, Jacquemin B, Sanchez M, Vierkötter A, Marcon A, Keidel D, Sugiri D, Al Kanani Z, Nadif R, Siroux V, Hardy R, Kuh D, Rochat T, Bridevaux PO, Eeftens M, Tsai MY, Villani S, Phuleria HC, Birk M, Cyrys J, Cirach M, de Nazelle A, Nieuwenhuijsen MJ, Forsberg B, de Hoog

2. Adebayo-Ojo TC, Wichmann J, Arowosegbe OO, Probst-Hensch N, Schindler C, Künzli N. Short-Term Joint Effects of PM_{10}, NO_2 and SO_2 on Cardio-Respiratory Disease Hospital Admissions in Cape Town, South Africa. Int J Environ Res Public Health. 2022 Jan 3;19(1):495. doi: 10.3390/ijerph19010495. PMID: 35010755; PMCID: PMC8744938.

3. Ai, S., Qian, Z. M., Guo, Y., Yang, Y., Rolling, C. A., Liu, E., Wu, F., & Lin, H. (2019). Long-term exposure to ambient fine particles associated with asthma: A cross-sectional study among older adults in six low- and middle-income countries. Environmental Research, 168, 141-145. https://doi.org/10.1016/j.envres.2018.09.02

4. Arku RE, Brauer M, Ahmed SH, et al. Long-term exposure to outdoor and household air pollution and blood pressure in the Prospective Urban and Rural Epidemiological (PURE) study. Environ Pollution. 2020; 262:114197

5. Bagula H, Olaniyan T, de Hoogh K, Saucy A, Parker B, Leaner J, Röösli M and Dalvie MA. Ambient Air Pollution and Cardiorespiratory Outcomes amongst Adults Residing in Four Informal Settlements in the Western Province of South Africa. International Journal of Environmental and Public Health. 2021

6. Bai, L., Shin, S., Burnett, R. T., Kwong, J. C., Hystad, P., van Donkelaar, A., Goldberg, M. S., Lavigne, E., Copes, R., Martin, R. v., Kopp, A., & Chen, H. (2019). Exposure to ambient air pollution and the incidence of congestive heart failure and acute myocardial infarction: A population-based study of 5.1 million Canadian adults living in Ontario. Environment International, 132. https://doi.org/10.1016/j.envint.2019.105004

7. Beelen R, Raaschou-Nielsen O, Stafoggia M […] Hoek G. The Effects of long-term exposure to air pollution on natural-cause mortality: An analysis of 22 European cohorts within the multicentre ESCAPE project. Lancet (2014) 383(9919) 785-795

8. Benzton J, Otsuka F, Virmani R. Mechanisms of plaque rupture and formation. Circulation Research. 2014 114:1852–1866

9. Boudrel T, Bind MA, Béjot Y, Morel O and Argacha JF. Cardiovascular effects of air pollution. Arch Cardiovasc. Disease. 2017

10. Brunekreef B, Strak M, Chen J, Andersen ZJ, Atkinson R, Bauwelinck M, Bellander T, Boutron MC, Brandt J, Carey I, Cesaroni G, Forastiere F, Fecht D, Gulliver J, Hertel O, Hoffmann B, de Hoogh K, Houthuijs D, Hvidtfeldt U, Janssen N, Jorgensen J, Katsouyanni K, Ketzel M, Klompmaker J, Hjertager Krog N, Liu S, Ljungman P, Mehta A, Nagel G, Oftedal B, Pershagen G, Peters A, Raaschou-Nielsen O, Renzi M, Rodopoulou S, Samoli E, Schwarze P, Sigsgaard T, Stafoggia M, Vienneau D, Weinmayr G, Wolf K, Hoek G. Mortality and Morbidity Effects of Long-Term Exposure to Low-Level $PM_{2.5}$, BC, NO_2, and O_3:

An Analysis of European Cohorts in the ELAPSE Project. Res Rep Health Eff Inst. 2021 Sep;2021(208):1-127. PMID: 36106702; PMCID: PMC9476567.

11. Cai Y, Zijlema WL, Doiron D, Blangiardo M, Burton PR, Fortier I, Gaye A, Gulliver J, de Hoogh K, Hveem K, Mbatchou S, Morley DW, Stolk RP, Elliott P, Hansell AL, Hodgson S. Ambient air pollution, traffic noise and adult asthma prevalence: a BioSHaRE approach. Eur Respir J. 2017 Jan 11;49(1):1502127. doi: 10.1183/13993003.02127-2015. PMID: 27824608.

12. Cai Y, Hansell AL, Blangiardo M, Burton PR; BioSHaRE, de Hoogh K, Doiron D, Fortier I, Gulliver J, Hveem K, Mbatchou S, Morley DW, Stolk RP, Zijlema WL, Elliott P, Hodgson S. Long-term exposure to road traffic noise, ambient air pollution, and cardiovascular risk factors in the HUNT and lifelines cohorts. Eur Heart J. 2017 Aug 1;38(29):2290- 2296. doi: 10.1093/eurheartj/ehx263. PMID: 28575405; PMCID: PMC5837618.

13. Carey IM, Atkinson RW, Kent AJ, van Staa T, Cook DG, Anderson HR. Mortality associations with long-term exposure to outdoor air pollution in a national English cohort. Am J Respir Crit Care Med. 2013 Jun 1;187(11):1226-33. doi: 10.1164/rccm.201210-1758OC. PMID: 23590261; PMCID: PMC3734610.

14. Cesaroni, G, Forastiere, F, Stafoggia, M, Andersen, Z. J., Badaloni, C, Beelen, R., Caracciolo, B, de Faire, U, Erbel, R, Eriksen, K. T, Fratiglioni, L, Galassi, C, Hampel, R, Heier, M., Hennig, F., Hilding, A., Hoffmann, B., Houthuijs, D., Jöckel, K. H, Peters, A. (2014). Long term exposure to ambient air pollution and incidence of acute coronary events: Prospective cohort study and meta-analysis in 11 European cohorts from the escape project. BMJ (Online), 348. https://doi.org/10.1136/bmj.f7412 Cardiac Evaluation. Journal of the American Heart Association, 5(8). https://doi.org/10.1161/JAHA.116.003947

15. Chen SY, Wu CF, Lee JH, Hoffmann B, Peters A, Brunekreef B, Chu DC, Chan CC. Associations between Long-Term Air Pollutant Exposures and Blood Pressure in Elderly Residents of Taipei City: A Cross-Sectional Study. Environ Health Perspect. 2015 Aug;123(8):779-84. doi: 10.1289/ehp.1408771. Epub 2015 Mar 20. PMID: 25793646; PMCID: PMC4529013.

16. Crouse D, Peters P, Villeneuve P, Proux M, Shin H, Goldberg M, Johnson M, Wheeler A. […] Within- and between-city contrasts in nitrogen dioxide and mortality in 10 Canadian cities; A subset of the Canadian Census Health and Environment Cohort (CanCHEC) Burnett R. Journal of Exposure Science and Environmental Epidemiology (2015) 25(5) 482-489

17. Curtin, L.R.; Mohadjer, L.K.; Dohrmann, S.M.; Kruszon-Moran, D.; Mirel, L.B.; Carroll, M.D.; Hirsch, R.; Burt, V.L.; Johnson, C.L. National Health and Nutrition Examination Survey: Sample design, 2007–2010. Vital Health Stat. 2013, 2, 1–23.

18. Fisher, J. A., Puett, R. C., Hart, J. E., Camargo, C. A., Varraso, R., Yanosky, J. D., & Laden, F. (2016). Particulate matter exposures and adult-onset asthma and COPD in the Nurses' Health Study. In European Respiratory Journal (Vol. 48, Issue 3, pp. 921– 924). European Respiratory Society. https://doi.org/10.1183/13993003.00845-2015

19. Fisher F, Cropper ML, Kumar P, Binagwaho A, Koudenoukpo JB, Park Y, Taghian G, Landrigan PJ. Air pollution and development in Africa: impacts on health, the economy, and human capital. Lancet.Open Access Published:October, 2021DOI:https://doi.org/10.1016/S2542-5196(21)00201-1

20. Gruzieva O., Xu C.-J., Breton C.V., Annesi-Maesano I., Antó J.M., Auffray C., Ballereau S., Bellander T., Bousquet J., Bustamante M., et al. Epigenome-Wide Meta-Analysis of Methylation in Children Related to Prenatal NO_2 Air Pollution Exposure. Environ. Health Perspect. 2017;125:104–110. doi: 10.1289/EHP36.\

21. Hart JE, Puett RC, Rexrode KM, albert CM, Laden F. Effect modification of long-term air pollution exposures and the risk of incident cardiovascular disease in US women. Journal of the American Heart Association, 4(12). https://doi.org/10.1161/JAHA.115.002301

22. Hartiala, J., Breton, C. v., Tang, W. H. W., Lurmann, F, Hazen, S. L., Gilliland, F. D, & Allayee, H. (2016). Ambient Air Pollution Is Associated with the Severity of Coronary Atherosclerosis and Incident Myocardial Infarction in Patients Undergoing Elective

23. Hoek G, Ranjini M Krishnan RM, Rob Beelen R, Annette Peters A, Bart Ostro B, Bert Brunekreef B, Joel D Kaufman JD. Long-term air pollution exposure and cardiorespiratory mortality: a review. Environmental Health. 2013

24. Hystad, P., Larkin, A., Rangarajan, S., AlHabib, K. F., Avezum, Á., Calik, K. B. T, Chifamba, J., Dans, A., Diaz, R., du Plessis, J. L, Gupta, R, Iqbal, R., Khatib, R, Kelishadi, R., Lanas, F, Liu, Z., Lopez-Jaramillo, P, Nair, S, Poirier, P., … Brauer, M. (2020). Associations of outdoor fine particulate air pollution and cardiovascular disease in 157 436 individuals from 21 high-income, middle-income, and low-income countries (PURE): a prospective cohort study. The Lancet Planetary Health, 4(6), e235–e245. https://doi.org/10.1016/S2542-5196(20)30103-0

25. Huang K, Liang F, Yang X, Liu F, Li J, Xiao Q, Chen J, Liu X, Cao J, Shen C, Yu L, Lu F, Wu X, Zhao L, Wu X, Li Y, Hu D, Huang J, Liu Y, Lu X, Gu D. Long term exposure to ambient fine particulate matter and incidence of stroke: prospective cohort study from the China-PAR project. BMJ. 2019 Dec 30;367: l6720. doi: 10.1136/bmj. l6720. PMID: 31888885; PMCID: PMC7190010

26. Jarvis, D.; Knox, J.; Burney, P.; Chinn, S.; Luczynska, C.; Anto, J.M.; Cerveri, I.; de Marco, R.; Gislasson, T.; Heinrich, J.; et al. The European Community Respiratory Health Survey II. Steer. Comm. Eur. Respir. J. 2002, 20, 1071–1079.

27. Jacquemin B, Siroux V, Sanchez M, Carsin AE, Schikowski T, Adam M, Bellisario V, Buschka A, Bono R, Brunekreef B, Cai Y, Cirach M, Clavel-Chapelon F, Declercq C, de Marco R, Nazelle A, Ducret-Stich RE, Ferretti VV, Gerbase MW, Hardy R, Heinrich J, Janson C, Jarvis D, Al Kanaani Z, Keidel D, Kuh D, Le Moual N, Nieuwenhuijsen MJ, Marcon A, Modig L.et al. Ambient Air Pollution and Adult Asthma Incidence in Six European Cohorts (ESCAPE). Environmental Health Perspectives. 2015

28. Karimi, B., Samadi, S. Long-term exposure to air pollution on cardio-respiratory, and lung cancer mortality: a systematic review and meta-analysis. J Environ Health Sci Engineer 22, 75–95 (2024). https://doi.org/10.1007/s40201-024-00900-6

29. Kauffman J, Adar SA, Barr RG, Budoff M, Burke GL, Curl CL, Daviglus ML, Roux AVD, Gassett AJ, Jacobs Jr DR, Kronmal R, Larson TV, Navas-Acien A, Olives C, Sampson PD, Sheppard L, Siscovick DS, Stein JH, Szpiro AA, Watson KE. Association between air pollution and coronary artery calcification within six metropolitan areas in the USA (the Multi-Ethnic Study of Atherosclerosis and Air Pollution): a longitudinal cohort study. Volume 388, Issue 10045, P696-704, 13 August 2016.

30. Kim H, Kim J, Kim S, Kang SH, Kim HJ, Kim H, Heo J, Yi SM, Kim K, Youn TJ, Chae IH. Cardiovascular Effects of Long-Term Exposure to Air Pollution: A Population-Based Study With 900 845 Person-Years of Follow-up. J Am Heart Assoc. 2017 Nov 8;6(11):e007170. doi: 10.1161/JAHA.117.007170. PMID: 29118034; PMCID: PMC5721790.

31. K, Declerq C, Bono R, Piccioni P, Quass U, Heinrich J, Jarvis D, Pin I, Beelen R, Hoek G, Brunekreef B, Schindler C, Sunyer J, Krämer U, Kauffmann F, Hansell AL, Künzli N, Probst-Hensch N. Adult lung function and long-term air pollution exposure. ESCAPE: a multicentre cohort study and meta-analysis. Eur Respir J. 2015 Jan;45(1):38-50. doi: 10.1183/09031936.00130014. Epub 2014 Sep 5. PMID: 25193994; PMCID: PMC4318659.

32. Lee YG, Lee PH, Choi SM, An MH, Jang AS. Effects of Air Pollutants on Airway Diseases. Int J Environ Res Public Health. 2021 Sep 20;18(18):9905. doi: 10.3390/ijerph18189905. PMID: 34574829; PMCID: PMC8465980

33. Liu K, Hua S, Song L. PM2.5 Exposure and Asthma Development: The Key Role of Oxidative Stress. Oxid Med Cell Longev. 2022 Apr 4;2022:3618806. doi: 10.1155/2022/3618806. PMID: 35419163; PMCID: PMC9001082.

34. Liu, Y., Yang, S., Li, C. *et al.* Changes in years of life lost and life expectancy by reducing air pollution. Air Qual Atmos Health 17, 89–102 (2024). https://doi.org/10.1007/s11869-023-01430-0

35. Ljungman, P. L. S, Andersson, N, Stockfelt, L., Andersson, E. M, Sommar, J. N, Eneroth, K., Gidhagen, L., Johansson, C, Lager, A., Leander, K., Molnar, P., Pedersen, N. L., Rizzuto, D., Rosengren, A., Segersson, D., Wennberg, P., Barregard, L., Forsberg, B., Sallsten, G., … Pershagen, G. (2019). Long-term exposure to particulate air pollution, black carbon, and their source components in relation to ischemic heart disease and stroke. Environmental Health Perspectives, 127(10). https://doi.org/10.1289/EHP4757

36. Mark W. Frampton, Joseph Boscia, Norbert J. Roberts, Jr., Mitra Azadniv, Alfonso Torres, Christopher Cox, Paul E. Morrow, Joan Nichols, David Chalupa, Lauren M. Frasier, F. Raymond Gibb, Donna M. Speers, Ying Tsai, and Mark J. Utell. Nitrogen dioxide exposure: effects on airway and blood cells. American Journal of Physiology-Lung Cellular and Molecular Physiology 2002 282:1, L155-L165

37. National Environmental Management: Air Quality Act 39 OF 2004 (Gazette No. 27318, Notice No. 163. Commencement date: 11 September 2005 – save for sections 21, 22, 36 to 49, 51(1)(e), 51(1)(f), 51(3), 60 and 61 [Government Notice R898, Gazette No. 28016]) (Commencement date of Sections 21, 22, 36 to 49, 51(1)(e), 51(1)(f), 51(3), 60 and 61: 1 April 2010 [Proc. No. 220, Gazette No. 33041]) NATIONAL AMBIENT AIR QUALITY STANDARDS Published under Government Notice 1210 in Government Gazette 32816 dated 24 December 2009. Commencement date: 24 December 2009

38. Newell, K. Kartsonaki , Lam KBH, Kurm OP Cardiorespiratory health effects of particulate ambient air pollution exposure in low-income and middle-income countries: a systematic review and meta-analysis. Lancet Planet Health 2017

39. Olaniyan T, Jeebhay M, Röösli M, Naidoo RN, Künzlib N, de Hoogh K, Saucy A, Badpa M, Baatjies R, Parker B, Leanerg J, Dalvie MA. The association between ambient NO_2 and $PM_{2.5}$ with the respiratory health of school children residing in informal settlements: A prospective cohort study. Environmental research. 2020

40. Olaniyan T, Jeebhay M, Roosli M, Naidoo R, Baatjjies R, Kunzil N, Tsai M, Davey M, de Hoogh K, Berman D, Parker B, Leaner J, Dalvie MA. A prospective cohort study on ambient air pollution and respiratory morbidities including childhood asthma in adolescents from the western Cape Province: study protocol. BMC Public Health. 2017.

41. Olaniyan T, Dalvie MA, Roosli M, Naidoo RN, Künzli N, Hoogh K, Berman D, Parker B, Leaner J, Jeebhay MF. Short term seasonal effects of airborne fungal spores on lung function in a panel study of schoolchildren residing in informal settlements of the Western Cape of South Africa. Environmental Pollution. 2020

42. Pitchika A, Hampel R, Wolf K, Kraus U, Cyrys J, Babisch W, Peters A, Schneider A. Long term associations of modelled and self-reported measures of exposure to air pollution and noise at residence on prevalent hypertension and blood pressure. Sci Total Environ. 2017 Sep 1;593-594:337-346. doi: 10.1016/j.scitotenv.2017.03.156. Epub 2017 Mar 27. PMID: 28346907.

43. Santos UP, Arbex MA, Braga ALF, Mizutani RF, Cançado JED, Terra-Filho M, Chatkin JM. Environmental air pollution: respiratory effects. J Bras Pneumol. 2021 Feb 8;47(1):e20200267. doi: 10.36416/1806-3756/e20200267. PMID: 33567063; PMCID: PMC7889311

44. Saucy, A.; Röösli, M.; Künzli, N.; Tsai, M.-Y.; Sieber, C.; Olaniyan, T.; Baatjies, R.; Jeebhay, M.; Davey, M.; Flückiger, B.; et al. Land Use Regression Modelling of Outdoor NO_2 and $PM_{2.5}$ Concentrations in Three Low Income Areas in the Western Cape Province, South *Africa.* Int. J. Environ. Res. Public Health *2018,* 15*, 1452.* https://doi.org/10.3390/ijerph15071452

45. Wang X et al. Air pollution associated with incidence and progression trajectory of chronic lung diseases: a population-based cohort study. Thorax 2023. Doi:10.1136/thorax-2022-219489.

46. Wang M, Aaron CP, Madrigano J, Hoffman EA, Angelini E, Yang J, Laine A, Vetterli TM, Kinney PL, Sampson PD, Sheppard LE, Szpiro AA, Adar SD, Kirwa K, Smith B, Lederer DJ, Diez-Roux AV, Vedal S, Kaufman JD, Barr RG. Association Between

Long-term Exposure to Ambient Air Pollution and Change in Quantitatively Assessed Emphysema and Lung Function. JAMA. 2019 Aug 13;322(6):546-556. doi: 10.1001/jama.2019.10255. PMID: 31408135; PMCID: PMC6692674.

47. Weichenthal S, Kulka R, Lavigne E, van Rijswijk D, Brauer M, Villeneuve PJ, Stieb D, Joseph L, Burnett RT. Biomass Burning as a Source of Ambient Fine Particulate Air Pollution and Acute Myocardial Infarction. Epidemiology. 2017 May;28(3):329-337. doi: 10.1097/EDE.0000000000000636. PMID: 28177951; PMCID: PMC5389593.

48. Wichmann J, Voyi K. Ambient Air Pollution Exposure and Respiratory, Cardiovascular and Cerebrovascular Mortality in Cape Town, South Africa: 2001–2006. Int. J. Environ. Res. Public Health 2012, 9, 3978-4016

49. World Health Organization ambient air quality database update 2021 version 6.1

50. World Health Organization. Ambient air pollution: A global assessment of exposure and burden of disease. WHO. World Health Organization; 2016.

51. World Health Organization. Ambient Air Pollution Attributable DALYs. 2018. Available online: https://www.who.int/data/gho/data/indicators/indicator-details/GHO/mbd-aap-ambient-air-pollution-attributable-dalys

52. WHO Global Ambient Air Quality Database (Update 2018). Available online: https://www.who.int/news-room/fact-sheets/detail/ambient-(outdoor)-air-quality-and-health

53. WHO (2014) Burden of disease from the joint effects of household and ambient air pollution for 2012. Summary of results. Available at: www.who.int/phe/health_topics/outdoorair/databases/HAP_BoD_results_March2014.pdf

54. Wu TJ, Wu CF, Chen BY, Lee YL, Guo YL. Age of asthma onset and vulnerability to ambient air pollution: an observational population-based study of adults from Southern Taiwan. BMC Pulm Med. 2016 Apr 19;16(1):54. doi: 10.1186/s12890-016-0218-0. PMID: 27094099; PMCID: PMC4837625.

55. Xu et al, 2023. Joint effects of Long Term exposure to ambient fine particulate matter and ozone on asthmatic symptoms: Prospective Cohot Study.

56. Yang BY, Guo Y, Markevych I, Qian ZM, Bloom MS, Heinrich J, Dharmage SC, Rolling CA, Jordan SS, Komppula M, Leskinen A, Bowatte G, Li S, Chen G, Liu KK, Zeng XW, Hu LW, Dong GH. Association of Long-term Exposure to Ambient Air Pollutants With Risk Factors for Cardiovascular Disease in China. JAMA Netw Open. 2019 Mar 1;2(3):e190318. doi: 10.1001/jamanetworkopen.2019.0318. PMID: 30848806; PMCID: PMC6484675.

Supplementary Information

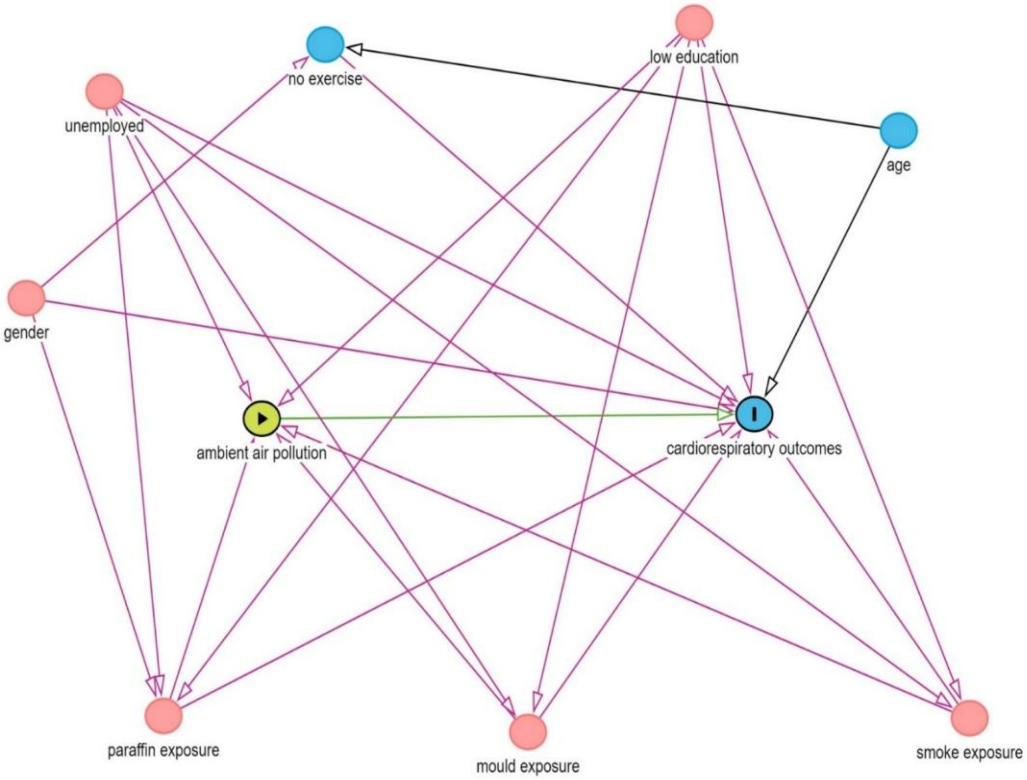

Figure S1: DAG

Table S 1: Bivariate analysis of covariates and cardiorespiratory outcomes

Outcomes	Age (years)	Gender (male) (n=51)	smoke exposure (n=142)	mould exposure (n=49)	paraffin exposure (n=279)	low education (n=74)	unemployed (n=235)
asthma symptom score >=2 (n=28)	**1.08 (1.04-1.11)**	0.49 (0.19-1.51)	0.85 (0.33-1.95)	0.71 (0.11-2.46)	0.80 (0.37-1.74)	**4.26 (1.86-9.43)**	1.05 (0.49-2.29)
self - reported chest pain (n=38)	**1.06 (1.03-1.09)**	0.72 (0.29-2.18)	0.56 (0.22-1.23)	0.79 (0.18-2.30)	0.90 (0.46-1.75)	**2.62 (1.19-5.42)**	1.01 (0.52-1.98)
self - reported hypertension (n=86)	**1.09 (1.06-1.11)**	1.99 (0.84 -5.88)	0.92 (0.54-1.54)	1.68 (0.81-3.29)	1.09 (0.69-1.76)	**5.33 (3.10-9.17)**	**1.33 (1.19-1.49)**
self- reported hypercholesterolemia (n=10)	**1.07 (1.01-1.12)**	0.44 (0.11-2.96)	2.62 (0.72-9.56)	2.39 (0.35-9.87)	0.81 (0.22-2.95)	1.47 (0.22-6.02)	0.83 (0.50-1.19)
doctor diagnosed asthma (n=12)	**1.1 (1.04-1.15)**	1.12 (0.21-20.83)	0.56 (0.09-2.22)	0.93 (0.05-5.02)	0.67 (0.19-2.26)	**11.18 (3.29-43.64)**	1.27 (0.96-1.63)
Wheezing (n=12)	**1.09 (1.04-1.15)**	0.55 (0.14-3.65)	0.51 (0.08-1.95)	0.84 (0.05-4.49)	0.81 (0.25-2.62)	**12.97 (3.97-49.71)**	1.22 (0.92-1.55)
shortness of breath on normal activity (n=5)	**1.12 (1.05-1.21)**	NC	0.64 (0.03-4.36)	2.36 (0.12-16.35)	0.20 (0.01-1.37)	NC	NC
shortness of breath on exertion (n=65)	**1.04 (1.02-1.07)**	1.12 (0.49-30.2)	0.67 (0.35-1.22)	0.27 (0.14-0.89)	1.26 (0.74-2.16)	1.56 (0.77-2.95)	0.97 (0.83-1.11)

*NC = *denotes not computable due to lack of model convergence*

Table S 2: A single - pollutant model demonstrating the association between $PM_{2.5}$ or NO_2 ($\mu g/m^3$) with self -reported cardiorespiratory outcomes among female adult participants in four informal settlements of the Western Cape Province of South Africa

	$PM_{2.5}$		**NO_2**	
	OR 95% CI			
Outcomes	**unadjusted**	**adjusted**	**unadjusted**	**adjusted**
Asthma Symptoms score	1.17	1.08	1.87	1.59
>= 2 (n=28)	(0.79-1.56)	(0.64-1.52)	(0.91- 3.93)	(0.68-3.80)
Chest pain	1.16	1.11	**1.89**	1.68
(n=38)	(0.82- 1.52)	(0.74-1.50)	**(1.01- 3.60)**	(0.82-3.50)
Hypercholesterolaemia	1.17	1.03	2.92	2.17
(n=10)	(0.59- 1.69)	(0.40-1.82)	(0.88- 10.51)	(0.51-9.13)
Doctor-diagnosed asthma	1.13	1.21	0.85	0.97
(n=12)	(0.56- 1.64)	(0.56-1.80)	(0.26- 2.64)	(0.26- 3.51)
Wheezing	1.17	1.21	1.02	0.97
(n=12)	(0.63- 1.66)	(0.56-1.80)	(0.34- 3.03)	(0.26-3.51)
Shortness of breath	1.18	1.17	**3.45**	**4.20**
(n=65)	(0.91- 1.51)	(0.87-1.52)	**(2.05- 6.02)**	**(2.32-7.90)**

adjusted model includes: age, sex, paraffin exposure, mould exposure, smoke exposure, low education, unemployed
NC denotes not computable due to lack of model convergence
Bold text denotes significant at P <0.05

PART B: APPENDICES

Appendix 1: Research Proposal

Title: The association between ambient air pollution and cardiorespiratory health outcomes of adults residing in informal settlements of the Western Cape in a 12 month period.

Student: Tasneem Allie Student Number: ALLTAS007

Supervisor: Professor Aqiel Dalvie

Co-supervisor: Dr Toyib Olaniyan

Degree: Master of Public Health

1. Literature Review

A literature review was conducted to provide an understanding on the current evidence regarding the association between ambient air pollution and cardiorespiratory health outcomes.

A search was conducted in PUBMED, Google scholar and Science Direct using keywords “ambient air pollution, cardiorespiratory, cardiovascular, hypertension, respiratory, particulate matter and nitrogen dioxide.” Studies conducted among adults from both urban and rural areas were included from 2014 - 2023. Studies which involved children or adolescents and studies with short term exposure to particulate matter and nitrogen oxide for less than 12 months were excluded. Studies involving the pathophysiological mechanisms involved in the development of cardiorespiratory outcomes when exposed to air pollution were also explored. The outcomes researched in terms of cardiovascular morbidity and mortality include hypertension, ischaemic heart disease, myocardial infarctions and stroke.

Background information regarding the epidemiological evidence relating to the impact of air pollution on cardiorespiratory outcomes will be discussed. A description will be given on the types of air pollution and the pathophysiology as to the mechanisms involved in the development of cardiorespiratory outcomes after long term exposure to ambient air pollution.

1.1 Introduction

According to the World Health Organization, in 2019 ambient air pollution was associated with the premature death of approximately 4.2 million people globally. Low- and middle income countries are especially burdened with the harmful effects, and it is estimated that approximately 89% of these deaths occur in these countries. Studies in these areas have been limited as is evidenced by the paucity of data within these regions in the 2016 WHO report on trends on exposure to ambient air pollution. In Africa, air pollution caused by a combination of household and ambient air pollution is estimated to cause approximately 1.1 million deaths in 2019 (Fisher et al, 2021)

Cardiorespiratory illnesses encompass a range of conditions that affect the cardiovascular and respiratory systems. These non-communicable diseases include Ischaemic Heart Disease, hypertension, rheumatic heart disease which affect the cardiovascular system as well as cerebrovascular disease such as stroke. The pulmonary or respiratory system is affected by chronic conditions such as asthma, chronic obstructive pulmonary disease and lung cancer, as well as those conditions of infective origin such as pneumonia and chronic bronchitis. WHO, 2019)

While cardiovascular disease itself can be influenced by a range of risk factors such as obesity, tobacco use, genetic predisposition, high sodium intake, hypertension, hypercholesterolaemia and physical inactivity, it has become increasingly evident that air pollution is a contributory component. Studies have indicated that long term exposure to ambient air pollution may be associated with an elevated blood pressure, increased risk of cardiovascular morbidity and mortality. (Hoek et al, 2013)

Cardiovascular disease accounts for the highest number of deaths worldwide resulting in 17,9 million deaths in 2016 according to the World Health Organization. Cardiovascular mortality accounts for approximately 60 – 80% of deaths due to air pollution (Boudrel et al, 2017). High blood pressure remains the leading cause for cardiovascular deaths worldwide and accounts for an estimated 7.6 million deaths per year. About 75% of these deaths occur in low to middle income countries (WHO,

2018). According to the global disease burden survey 2016, elevated systolic blood pressure accounted for approximately 80 million disability adjusted life-years.

In South Africa, cardiovascular disease is the second leading cause of death after HIV/AIDS.
According to the South African Medical Research Council's second burden of disease survey, 23.4 % of deaths in the Western Cape are due to cardiovascular disease (South African Medical Research Council, Burden of Disease Study, 2016).

Research into the role of ambient air pollution contributing to this non-communicable burden of disease is becoming increasingly evident, however research, especially in low to middle income countries is lacking. There is a great burden of environmental air pollution in low to middle income countries and evidence has shown that urbanisation may lead to higher levels of exposure to air pollution.

According to Wichmann et al, 2012 a study conducted in Cape Town demonstrated an associated increase in cardiovascular and respiratory mortality with exposure to ambient air pollution. Cardiovascular mortality increased 3% with each increased interquartile range (IQR) of NO_2 and SO_2.

In a study conducted by Olaniyan et al, 2020 in informal settlements within the Western Cape, there was an increase in the incidence of asthma -associated outcomes with long - term exposure to ambient NO_2 in children. These results were found at NO_2 levels below that which is expected by both local and international air quality standards at an annual average concentration of 16.62µg/m3. Other biological components of particulate matter such as fungal spores were also shown to decrease peak expiratory flow when assessing lung function in children within South Africa. (Olaniyan et al, 2020)

1.2 Ambient air pollution

Air pollution is defined as the presence of toxic compounds in the air which impact negatively on the quality of air resulting in harmful health and environmental outcomes. (Environmental pollution Centres, 2021).

Ambient air pollution can be caused by natural phenomena such as wildfires or volcanoes known as inorganic particulates, or it may be formed in the active production of organic compounds through industrial and chemical mechanisms. These chemical processes can occur both inside the home in the form of combustion caused by creating fires for food and oils that are burned, or cleaning agents that result in aerosolized chemical exposure, however the focus on external factors in the environment that may contribute to air pollution include those of an industrial nature and vehicle emissions.

Primary air pollutants are formed and emitted directly into the air in the form of air pollution from the particle source. These include sulphur dioxide (SO_2), oxides of nitrogen (NO_2), carbon monoxide (CO), volatile organic compounds as well as carbonaceous and non - carbonaceous particles. Secondary air pollutants such as ozone are formed from chemical reactions in the lower atmosphere. The term "criteria air pollutants" is used to measure whether geographical areas are able attain air quality standards by adhering to certain levels of air pollution. (Vallero D et al, 2014)

The Environmental Protection Agency (EPA) has identified six "criteria air pollutants" as they are the most common hazardous emissions including 1) ozone (O_3), a naturally formed gas composed of O_2 and UV radiation, particulate matter which comprises a mixture of solid and liquid particles of varying sizes in the air, 2) carbon monoxide (CO), an odourless and colourless gas formed in combustion processes especially from motor vehicle exhaust fumes 3) lead (Pb) which becomes airborne via processes involving aircraft fuel, metal and ore production 4) sulphur dioxide formed in the combustion process of sulphur notable on fossil fuel production 5) particulate matter , which

comprises of a combination of solid particles and liquid droplets of varying sizes which exist in the air and 6) nitrogen dioxide formed in the production of fuel. (EPA, 2021). In this study we will focus on particulate matter and nitrogen dioxide.

1.2.1 Particulate Matter

Particulate matter (PM) is made up of a mixture of solid and liquid particles found in the air. PM varies in particle size, with coarse particulate matter depicted as PM_{10} and fine particulate matter depicted as $PM_{2.5.}$ Its constituents are from a range of organic natural and chemicals, metals such as copper, iron, potassium, nickel, sulphur, silicon, vanadium, and zinc as well as acids. Within the South African context, $PM_{2.5}$ was best predicted by a combination of the population density and presence of grills and waste burning sites within the studied area. (Saucy A et al, 2018) Particulate matter has been shown to affect cardiorespiratory morbidity and long-term exposure has been found to be associated with an 11% (95% CI 5, 16%) increase in cardiovascular mortality and 3% (95% CI −6, 13%) increase in mortality from non-malignant respiratory diseases (Hoek et al, 2013).

Exposure to $PM_{2.5}$ in the short term was associated with a 0·47% (95% CI 0·34 – 0·61) increase in cardiovascular mortality and a 0·57% (0·28 – 0·86) increase in respiratory mortality for each 10 µg/m^3 increase in same-day $PM_{2·5}$. (Newell K, et al 2017)

1.2.2 Nitrogen dioxide

One of the gaseous air pollutants which occur through the oxidation of nitrogen include nitrogen dioxide, nitric oxide, ozone, volatile organic compounds and carbon monoxide. Phytochemical reactions with ammonia often from agriculture react with nitrogen dioxide, which is toxic, to form particles that further contribute to inhaled air pollution. In the South African context and specifically within the informal settlement areas studied, NO_2 levels were best predicted by the presence of traffic related exposure within close proximity to the geographical region. (Saucy A et al, 2018) Both short- and long-term exposure to nitrogen dioxide has been found to increase cardiovascular mortality (Hoek et al, 2013) and exposure has been positively associated with COPD mortality relative risks (RR) = 1.03 (1.01 - 1.04) per 10 µg/m^3 (Huangfu et al, 2020)

1.3 Pathophysiology of ambient air pollution and cardiorespiratory disease

Acute and chronic exposure to ambient air pollution may lead to inflammation of multiple organ systems most notably the cardiovascular, respiratory and autonomic nervous systems. An underlying genetic susceptibility and an abnormal response of gene expression in response to air pollution results in a sequence of events that lead to inflammatory responses with resultant respiratory and cardiovascular presentations such as asthma and ischaemic heart disease and the associated increase in morbidity and mortality relating to these outcomes. (Brooke, 2010)

1.3.1 Inflammation

Exposure to ambient air pollution may result in local and systemic inflammation. Local inflammation within the pulmonary system consequently results in constriction of the airways also known as bronchospasm. Systemic inflammation results in a cascade of events. Systemic oxidant stress, particularly in the vascular system, endothelial dysfunction, leukocyte platelet activation and the acute phase response and coagulation factors all contribute to the development of atherosclerosis. The progression of atherosclerotic plaques and the resultant plaque instability may progress to enable the plaque to rupture. The resultant thrombus may lead to obstruction within the vasculature. Evidence

of premature aortic and coronary calcification along with atherosclerosis may lead to ischaemic heart disease (IHD) and result in a myocardial infarction (MI). (Brooke et al, 2010)

1.3.2 Autonomic Nervous System

Another mechanism which may result in bronchospasm is the activation of pulmonary reflexes which is triggered by the autonomic nervous system when one is exposed to ambient air pollution. Activation of the autonomic nervous system may stimulate an increase or alteration in the heart rate which may result in arrhythmias. Arrhythmias may further contribute to the development of plaque instability and potential rupture of this may be associated with an increased risk of myocardial infarctions due to its atherothrombotic effect. Atherosclerotic plaque is a prominent feature of cardiovascular disease. In the Multi-Ethnic Study of Atherosclerosis Air Pollution Study (MESA Air), a healthy cohort of adults aged 45 – 84 years of age was followed for 10 years. The exposure to fine particles less the 2,5 micrometres in diameter and nitrogen oxides in the air and the development of atherosclerotic plaque was found to be −0·9 µm per year (95% CI −3·0 to 1·3 with exposure of 5 µg/m^3 higher long-term exposure to $PM_{2·5}$ and an in intima-media thickness or plaque formation of 0·2 µm per year (−1·9 to 2·4) with exposures of 40 ppb higher NO_2. (Kauffman et al, 2016) The development of atherosclerotic plaque is a major contributor to thrombi formation associated with coronary heart disease. (Bentzon et al, 2014)

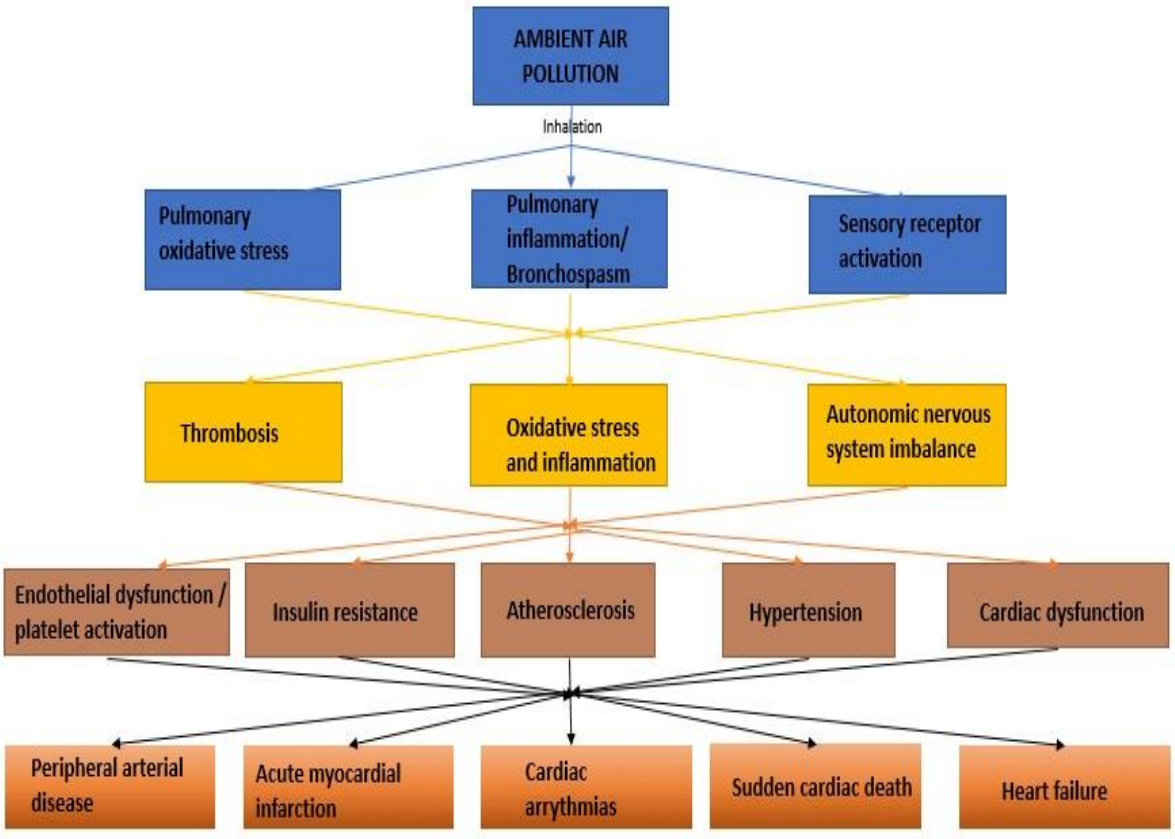

Figure 1. Schematic representation of the cardiorespiratory effects of ambient air pollution. (Adapted from Braunwald Heart Disease, 11th Edition, Chapter 52 Air Pollution and cardiovascular disease, 2018)

1.4 A review of the evidence on Ambient Air Pollution ($PM_{2.5}$ and PM_{10}) and NO_2 and its association with Cardiorespiratory Disease

A systematic review article by Hoek et al (2013) on the association between air pollution (particulate matter and NO_2) and cardiorespiratory outcomes included ten (10) publications relating to cardiovascular outcomes and eighteen (18) publications regarding respiratory outcomes. The results of the studies were divided into cardiovascular and respiratory categories based on the outcomes in order to represent the association to air pollution according to outcome.

Long term exposure to $PM_{2.5}$ was found to be associated with both cardiovascular and respiratory mortality. An increase in all – cause mortality associated with exposure to long term $PM_{2.5}$ was found

in most of the studies consolidated within the review, with significant heterogeneity across the studies. The excess mortality for cardiovascular disease per 10 ug/m^3 $PM_{2.5}$ was 11% (95% CI 5, 16%). The mortality due to respiratory disease was less than that of cardiovascular disease (Hoek et al, 2013). The systematic review lacked data on middle and low - income countries and also differed in terms of outcomes measures. Cardiovascular mortality was broadly measured as an outcome and it did not describe hypertension incidence in isolation as an outcome. A review of the literature was conducted for studies published between 2014 - 2023 globally in both urban and rural areas on short- and long-term exposure to ambient air pollution, particulate matter, nitrogen dioxide and sulphur dioxide.

1.4.1 Evidence on the association between cardiorespiratory mortality and morbidity with long term ambient air pollution exposure

Twenty studies were reviewed since the systematic review by Hoek et al, 2013 within the period 2014 – 2023. The majority (n = 8) of the studies were conducted in Europe, followed by Asia (n = 5) and North America (n = 5). One study was found in South Africa and one study was conducted in 21 countries which included low, middle and high income - settings. African countries that were included in the multinational study include South Africa, Tanzania and Zimbabwe. These studies were conducted in both the urban and rural geographical areas.

Participants' ages varied in the studies. Most of the studies consisted of adults above the age of 35, however 5 studies included a younger adult population from the age of 18 in Huang et al 2019, Kim et al 2017, Cai et al 2015, 2017 and Crouse et al 2015.

Six studies conducted research using a cross - sectional study design with the rest and majority (13) conducting research over a prospective period of time. The prospective cohort studies would be expected to yield more accurate results based on the pathophysiology of the outcomes and due to the variability in air pollution levels over seasons, environmental changes in urbanisation and exposure levels varying depending on the movement of participants.

Most of the studies (n = 8) gathered outcome data from information obtained from ICD codes or death certificates which made inferences on cardiorespiratory outcomes. (Wang et al 2014, Crouse et al, 2015, Hartiala et al 2016, Kim et al 2017, Bai et al 2019, Ljungman et al 2019, Liu et al 2021, Olaniyan et al 2022) Five (5) of the studies had the participants answer questionnaires which relied on participants recalling and reporting on their own health outcomes. (Cai et al 2015, Jacquemin et al 2015, Hart et al 2015, Wu et al 2016, Fisher et al 2016, Bagula et al 2021) Only 3 (three) of the studies made use of clinical measurements to ascertain health outcomes studied. (Pitchika et al 2017, Cai et al 2017, Arku et al 2020) Four (4) of the studies used a combination of either death certificates/ICD codes, questionnaires and clinical data, a combination which may have led to the obtained of more accurate outcome measures. (Chen et al 2015, Liu et al 2017)

The majority (n = 15) of the study outcomes related to cardiovascular mortality or morbidity while the remainder reported respiratory or a combination of cardiorespiratory outcomes. Significant associations were found between long term exposure to ambient air pollution and cardiorespiratory outcomes in 15 of the studies reviewed.

Positive associations were found for hypertension and long - term ambient air pollution exposure in four studies. (Chen et al 2015, Pitchika et al 2017, Liu et al 2017, Arku at al, Chen et al 2015). A discrepancy in systolic versus diastolic blood pressure elevation was found amongst the studies. The inclusion of an elderly cohort sample above the age of 65 in the study by Chen et al 2015 may account for the absence of association in increased diastolic blood pressure due to the resultant decrease in arterial compliance that presents with advanced age and isolated systolic blood pressure of often found in people above the age of 60. Each interquartile increase in annual mean $PM_{2.5}$ (1 $\mu g/m^3$) was significantly associated with a 15% higher prevalence of hypertension (95% CI: 0.7 - 30.8%). (Pitchika et al 2017) Among the participants in the study by Liu et al 2017, about 30 % reported being smokers, and due to the rural living environment made use of high unclean energy for heating and cooking purposes which may have confounded significant results.

These finding were consistent with the study by Cai et al 2016, which demonstrated that long-term exposure to NO_2 (OR=1.034, 95% CI: 1.005–1.063) was significantly associated with hypertension for every 10 µg/m^3 increase.

Positive associations for long term exposure to ambient air pollution and cardiovascular morbidity and mortality was found in nine of the studies. The study by Crouse et al, 2015 showed no association with cerebrovascular outcomes and long term NO_2 exposure and Hart et al 2015 only demonstrated an increased association of cardiovascular disease and stroke in diabetics with no statistically significant association being found in the non-diabetic study population. Long term exposure to NO_2 was associated with cardiovascular disease (HR per 5 p.p.b.: 1.04; 95% CI: 1.01–1.06), ischaemic heart disease (HR per 5 p.p.b.: 1.05; 95% CI: 1.02–
1.08) but not cerebrovascular disease (HR per 5 p.p.b.: 1.01; 95% CI: 0.96–1.06) (Crouse et al, 2015) Positive associations of borderline significance was found in the study conducted by Jacquemin et al, 2015.

Two studies demonstrated no association in long term ambient air pollution exposure and cardiovascular mortality or ischaemic heart disease (Wang et al 2014, Ljungman et al 2019) The study conducted by Wang et al 2014 took place among 19 European cohorts which differed significantly in terms of exposure levels among cohorts and participant demographics. This study also differed in that it measured exposure constituents which have not been studied in great detail. Hart et al 2015, found associations that were not statistically significant for cardiovascular disease, and stroke in terms of the general non-diabetic population. In diabetic women, long term exposure to $PM_{2.5}$ was associated with an increased risk of ischaemic heart disease for each 10 µg /m^3 increase in 12-month average (HR 1.44 95% CI: 1.23 to 1.68 (Hart et al, 2015). This finding may be due to the pathophysiology of diabetes and its subsequent complications and may not be reflective of an association with air pollution in isolation. More research is thus required on the diabetic population in order to ascertain whether the disease poses a vulnerability to air pollution in the individual and if there is a true association with diabetes and air pollution or whether the current evidence is indicative of a biased outcome due to the development of diabetic complications.

The study by Huang et al 2019, found an association between fine particulate matter exposure ($PM_{<2.5}$) and risk of stroke however the exposure differed from the $PM_{2.5}$ and NO_2 in the review studies. For each 10 µg/m^3 increase in $PM_{2.5}$, there was a significant association with a 13% increase in stroke incidence according to Huang et al, 2019. Time varying exposure was accounted for and exposure validated monthly to minimise bias relating to participants varying exposure levels away from their residential addresses.

One study Cai et al 2017, demonstrated an increase in biochemical markers which predispose to cardiovascular morbidity and found an association between changes in biochemistry markers and stroke. A study conducted by Hart et al 2015, did not demonstrate the same extent of association and found that for each 10 µg/m^3 increase in 12-month average $PM_{2.5}$, $PM_{2.5\ -\ 10}$, and PM_{10}, the multivariable adjusted hazard ratios were not significant. The study by Hart et al, 2015 may however not be generalisable to men, or socioeconomically and racially diverse populations as the study sample was conducted on a middle aged, female nursing population.

Three studies concluded an association between long term exposure to $PM_{2.5}$ and the development of acute myocardial infarction. (Bai et al 2019, Bagula et al 2021, Olaniyan et al 2022) The study by Bagula

et al 2021, found an association between self - reported chest pain, an indication of possible ischaemic heart disease or cardiovascular morbidity and long-term ambient air pollution exposure.

Results of the reviewed studies indicated that long term exposure to $PM_{2.5}$ is associated with an increased risk of asthma per 10 mcg/ m^3.

Long term exposure to NO_2 was associated with a 1.9% (95% CI 1.1–2.8%) higher prevalence of ever-had asthma (Cai et al 2015) In terms of respiratory outcomes, three studies found positive associations between long term ambient air pollution exposure and asthma. (Cai et al 2015, Wu et al 2016, Liu et al 2021) Two studies found no association between long term ambient air pollution exposure and asthma (Jacquemin et al, 2015, Fisher et al, 2016)

The study by Fisher et al 2016, produced evidence that found no consistent association between exposure to $PM_{2.5}$, PM_{10} and $PM_{2.5-10}$ on the incidence of asthma or COPD. A HR for a 10-μg·m−3 increase in 4-year average of 0.94 (95% CI 0.84–1.06), 0.90 (95% CI 0.73–
1.12) and 0.93 (95% CI 0.77–1.13) for PM_{10}, $PM_{2.5}$ or $PM_{10–2.5}$ respectively for asthma incidence. This study may not be generalisable to other populations as the sample studied included a female only population. Questionnaires used in the study may have resulted in outcome bias due to self - reporting and missed diagnoses or undiagnosed disease. Exposure misclassification may have occurred due to the differences in time-activity patterns of exposure.

Methods of measuring the level of exposure to ambient air pollution differed amongst the studies. Most of the studies (12) made use of air monitoring stations which included AEA tech dispersion models and spatio - temporal prediction models. EPA air quality monitoring stations were used in Wu et al 2016 and Hartiala et al 2016, global atmospheric chemistry transport model (GEOS-Chem CTM) was used in Olaniyan 2022 and Bai et al 2019, with Bai using it combined with a geographically weighted regression model grid with a spatial resolution of approximately 1 km × 1 km.

Four studies made use of land use regression modelling while two used a combination of land use regression models and air monitoring stations. One study questioned participants regarding self - reported exposure to vehicle traffic passing their homes but used this in conjunction with modelling data. Evidence demonstrated that self-reported measures of air pollution or noise exposure did not perform better than the objective measures. (Pitchika et al, 2017)

Huang et al 2019 used a satellite based spatiotemporal model with 1km x 1km resolution, as did Liu et al 2017. Time – varying exposure was accounted for in many of the studies making use of spatiotemporal models with resolution of 1 x 1 km.

1.4.2 Limitations and biases in the studies reviewed

Several limitations and biases were present in the studies reviewed. Amongst the studies that made use of questionnaires as the source of exposure or outcome data, recall bias may have been introduced. This would depend on the participants accuracy in recalling exposures, the date of occurrence, as well as their ability to accurately report outcomes.

A major limitation is exposure misclassification and information bias. Numerous studies made use of exposure data based on geographical areas distant from participants' homes or may not represent their actual exposure based on their daily geographical location and habits such as time spent indoors or outdoors. Home addresses were used as a point of measure which may not have accurately represented actual participant exposure and satellite estimation may have misrepresented individual exposure.

These studies may have misclassified the date of development of symptoms of cardiorespiratory illness as the outcome measure was ascertained from ICD codes or death certificates which made inferences about outcomes. Information bias regarding outcomes were present in some studies as outcome data was sourced from death certificates and medical records or ICD codes. Inferences regarding health outcomes could have been made or omitted based on a single diagnosis resulting in outcome bias and misclassification of the outcome and the duration of symptoms may not have been accurately accounted for due to the initiation of symptoms potentially preceding the diagnosis by months or even years.

Exposure misclassification could have been present in studies that used the mean exposure to air pollution as the exposure measurement as this level differed considerably across cities and even within cities as demonstrated in Crouse et al, 2015. Changes in exposure of outdoor air pollution may vary in individuals who spend much time indoors and the actual exposure may thus differ for individuals within the same study who have the same exposure misrepresented by the ambient air exposure measure used within the study. Although in environments with low levels of ambient air pollution exposure, which use large cohorts this bias may not be present.

The use of temporal land use regression models was used in numerous studies, in which current levels of ambient air pollution exposure was used to allude to health outcomes that have occurred in the years prior to exposure. The land use regression models give a stable representation of historic exposure, although this may not be true in geographical areas where rapid urbanisation has occurred. Thus, exposure misclassification may have occurred and generalisation to the context of informal settlements in a South African setting may not be possible.

Varying measurements of exposure have been used amongst the studies as well as a wide study period range. This may account for the differences in study outcomes. Comparing outcomes amongst studies with diverse levels of $PM_{2.5}$ and NO_2, remains difficult.

Some effect mediators were not considered in some studies such as the effect that different weather patterns could have on air pollution levels. This may have introduced information bias.

The study period and duration varied amongst studies and ranged from 12 months to 22 years, with the longer duration of study possibly detecting more cardiorespiratory outcomes due to a time – dependent relationship in the pathophysiology of health outcomes. Exposure misclassification is also more likely to be present in studies over a longer duration as participants may move or no longer reside in the areas studied.

Sampling bias may have occurred in certain studies with participants in the Fisher et al 2016 study who were all female nurses, in Wu et al 2016 study who were all known to be diagnosed with asthma and in Chen et al 2015 where all participants were over the age of 65.

Many confounders existed such as participants being smokers, being obese, having comorbid disease, indoor air pollution exposure and socioeconomic status which were adjusted for in some, but not all studies.

As most of the studies were conducted outside of Africa, generalisability within our context especially within the rural setting may be limited.

The differing methods of measuring exposure may account for the variability in effect estimates amongst the studies. Outdoor levels of particulate matter are used as a measure of individual exposure. This may result in exposure misclassification, and it has in previous studies resulted in

differences in effect estimates, as these levels are taken from a central site within the city if done via modelling.

1.5 Conclusion

In summary, evidence exists for the association between PM and NO_2 and cardiorespiratory health outcomes, however further evidence is required to reach conclusive results. One of the weaknesses in the evidence is the variable exposure data and more studies are required with strong exposure measures that estimate air pollution at high resolution. Exposure measure modelling methods of air pollution estimation include proximity modelling, interpolation models, air dispersion models and land use regression models. The resolution included in the studies ranged from 1 x 1 km, spatiotemporal exposure models with distances from primary or secondary roads of 0 – 50m, 50 – 200m and more than 200 m, coarse satellite derived estimates 10 x 10 km grid cells. Overall, the spatial resolution was limited to 10 km.

More accurate exposure measurement methods such as personal exposure monitoring due to the difference in indoor, outdoor and individual exposures levels are also required. Indoor – outdoor sampling comparisons and mobile air pollution monitoring studies are also required.

Another area in the evidence that requires improvement is the quality of outcome data where especially more objective measures are required. Many of the studies made use of data that inferred outcomes indirectly such as the use of ICD codes, death certificates, medical records and questionnaires that required participants to self- report outcomes. Studies which make use of more objective methods of measuring outcomes such as clinical methods of blood pressure monitoring for hypertension, electrocardiogram (ECG) changes or blood chemistry markers for ischaemic events and peak expiratory flow readings demonstrating reversibility for asthma diagnoses. More data is especially needed on respiratory health outcomes and on cerebrovascular health outcomes such as strokes.

Table 1: Review of epidemiological studies on long - term PM and NO_2 and cardiorespiratory outcomes

AUTHOR / YEAR	AIM	STUDY DESIGN, POPULATION	GEOGRAPHIC AREA	EXPOSURE	CVS / RESP OUTCOMES	RESULTS	BIAS / LIMITATIONS	CONCLUSION
Wang et al 2014	To determine the cardiovascular mortality associated with long term exposure to particulate matter	Cohort study 322,291 participants 4,551,184 person years at risk (follow-up 14.1 years	19 European cohorts	PM elemental constituents Land use regression modelling using satellite-derived and chemical transport modelling data residential	Cardiovascular mortality from registries	$PM_{2.5}$ HR1.17(95% CI: 0.93 – 1.47)	Exposure misclassification Variable demographics of study population, different exposure measures amongst cohorts Confounders such as weather not accounted for	No statistically significant association between particulate matter constituents and cardiovascular mortality
Chen et al 2015	To study the association between long term air pollutant exposure and BP in elderly residents	Cross sectional study 27752 participants > 65 years of age	Taipei	PM, NO_2 Land use regression modelling from ESCAPE project. 40 spatially distributed sites	Questionnaire and clinical blood pressure measurement	$PM_{2.5}$ 0.62 (95% CI: 0.24, 0.99) NO_2 0.34 (95% CI: 0.19, 0.50),	Sampling bias – elderly cohort. Socio- economic state not considered, Exposure misclassification	long term air pollution exposure was associated with an increased diastolic BP in elderly, nut not associated with systolic BP
Cai et al 2015	To investigate the effects of both ambient air pollution and traffic noise on adult asthma prevalence	Cross sectional study 50 805 participants > 20 years of age	Europe Norway Netherlands	PM_{10}, $PM_{2.5}$, NO_2 Land use regression modelling	Asthma prevalence, questionnaire	NO_2 higher by 10 μg·m−3 was associated with 1.9% (95% CI 1.1–2.8%) higher prevalence of ever-had asthma, but not current asthma prevalence	Misclassification of exposure	PM_{10} exposure is associated with asthma prevalence. NO_2 was significantly associated with ever-asthma but not current asthma
Crouse et al, 2015	To study the association between NO_2 exposure between and in cities and mortality	Cohort study, 10 cities in Canada 1984- 2006 (22year period) 735600 participants 25 years of age and older	Canada	NO_2 Ambient measurement and land use regression modelling coarse satellite derived estimates (i.e., 10 × 10 km grid cells	All-cause mortality Respiratory disease, cardiovascular disease, IHD	cardiovascular disease (HR per 5 p.p.b.: 1.04; 95% CI: 1.01–1.06), ischaemic heart disease (HR per 5 p.p.b.: 1.05; 95% CI: 1.02–1.08) and respiratory disease (HR per 5 p.p.b.: 1.04; 95% CI: 0.99–1.08), but not from cerebrovascular disease (HR per 5 p.p.b.: 1.01; 95% CI: 0.96–1.06)	Confounders: exposure misclassification	positive associations between overall NO_2 exposures and mortality from cardiovascular disease, respiratory disease IHD but not cerebrovascular disease

Hart et al 2015	To study the effect modification of long-term exposure to ambient air pollution and incident cardiovascular disease	Cohort study 114 537 women in the Nurses' Health Study, 1988 - 2006 30 - 55 years of age	United States	$PM_{2.5}$, PM_{10}, and $PM_{2.5-10}$ spatio-temporal prediction models for residential address time – varying exposure	biennial questionnaire diagnosed CVD or stroke medical records/ICD codes	For each 10 µg /m^3 increase in 12-month average $PM_{2.5}$, $PM_{2.5-10}$, and PM_{10}, the multivariable adjusted hazard ratios were 1.44 (95% CI: 1.23 to 1.68), 1.17 (95% CI: 1.05 to 1.30), and 1.19 (95% CI: 1.10 to 1.28) among women with diabetes	sampling bias: female nurses results not generalisable to men, racially or socioeconomically diverse population, information bias – exposure measurement error possible misclassification of exposure – lack of monitoring data before 1999, thus estimated misclassification of outcomes -	exposures to PM were associated with small, but non - statistically significant, elevations in incident CVD, CHD, and stroke, statistically significant increased risk in diabetics

							diagnosis required for outcome confounders present	
Jacquemin et al 2015	To study the association between ambient Air Pollution and Adult Asthma Incidence in Six European Cohorts	Prospective cohort study 23,704 participants 10-year period	6 European cohorts	PM_{10}, NO_2, passive samplers in the land use regression (LUR) models were used as primary exposure covariates ESCAPE annual mean concentration	Questionnaire	Positive associations of borderline significance were observed for nitrogen dioxide [adjusted odds ratio (OR) = 1.10; 95% CI: 0.99, 1.21 per 10 µg/m3; p = 0.10] and nitrogen oxides (adjusted OR = 1.04; 95% CI: 0.99, 1.08 per 20 µg/m3; p = 0.08). Nonsignificant positive associations were estimated for PM_{10} (adjusted OR = 1.04; 95% CI: 0.88, 1.23 per 10 µg/m3), $PM_{2.5}$ (adjusted OR = 1.04; 95% CI: 0.88, 1.23 per 5 µg/m3), $PM_{2.5absorbance}$ (adjusted OR = 1.06; 95% CI: 0.95, 1.19 per 10–5/m),	exposure misclassification information bias / outcome misclassification missed diagnosis	no significant effect of ambient air pollution on asthma incidence in adults
Wu et al 2016	To study the age of asthma onset and vulnerability to ambient air pollution	Cross - sectional cohort study 35682 participants 26-50 years of age	Taiwan	PM_{10}; air monitoring data collected from 26 Environmental Protection Administration air quality monitor stations. Kriging statistical mapping technique	Asthma severity in late onset asthmatics via questionnaire	exposure to higher PM_{10} were more likely to have higher severity scores (OR = 1.74; 95 % CI, 1.13 – 2.70) with asthma onset at > 12 years.	Known asthmatics -selection bias, sampling bias exposure misclassification	exposure to PM_{10} has a greater effect on late-onset asthma severity

Fisher et al 2016	To determine the association between long term particulate matter exposures and adult-onset asthma and COPD	Cohort study 121701 female nurses ages: 30 - 55 years 1992 - 2000	United States	$PM_{2.5}$, PM_{10}, $PM_{2.5\text{-}10}$ nationwide spatiotemporal exposure models Time – varying exposure modelling of residential distance to primary or secondary roads (0–50, 50–200 and ⩾200 m)	Biennial questionnaire incidence of asthma, emphysema or chronic bronchitis, clinical diagnosis and initiation of treatment	HRs for a $10\mu g \cdot m^{-3}$ increase in 4-year average PM_{10}, $PM_{2.5}$ or $PM_{10-2.5}$ 0.94 (95% CI 0.84–1.06), 0.90 (95% CI 0.73–1.12) and 0.93 (95% CI 0.77–1.13) for asthma, and 0.91 (95% CI 0.76–1.10), 0.93 (95% CI 0.66–1.31) and 0.83 (95% CI 0.60–1.14) for COPD	sampling bias, only female population, not generalisable exposure misclassification date of diagnosis limited to self-reported year of first physician diagnosis on the supplemental questionnaires, differences in time-activity patterns of exposure, outcome bias due to undiagnosed disease	no consistent evidence of an association between exposure to PM and incident asthma or COPD
Hartiala et al 2016	To study whether ambient Air Pollution Is Associated with the Severity of Coronary Atherosclerosis and Incident Myocardial Infarction in Patients Undergoing Elective Cardiac Evaluation	Prospective longitudinal cohort study 6575 participants undergoing angiogram 3 year follow up	Ohio	$PM_{2.5}$, NO_2 Daily concentrations of PM2.5 and NO2 in the United States from 1998 through 2010 were downloaded from the US Environmental Protection Agency's (EPA) Air Quality System (AQS) database	MI, stroke, all-cause mortality	A 2-SD increase in $PM_{2.5}$ levels over 36 months increased likelihood of having coronary atherosclerosis that was mild (odds ratio 1.43, 95% CI 1.11–1.83, P=0.005) and severe (odds ratio 1.63, 95% CI 1.26–2.11, P<0.0001) Exposure to higher $PM_{2.5}$ levels was also significantly associated with increased risk of incident myocardial infarction (hazard ratio 1.33, 95% CI 1.02–1.73, P=0.03) but not stroke or all-cause mortality	sampling bias - high risk of cardiac disease as participants were known with IHD	Increase in exposure to ambient air pollution is associated with mild and severe coronary atherosclerosis higher levels of air pollution exposure are associated with an increased risk of MI

Pitchika et al 2017	To study the long-term associations of exposure to air pollution and noise at residence on prevalent hypertension and blood pressure	Cross sectional cohort study 2552 participants aged 31 – 72 years of age	Augsberg, Germany	$PM_{2.5}$, NO_2 ESCAPE project monitoring sites and land use regression modelling	Clinical: Blood pressure measurement	An interquartile increase in annual mean $PM_{2.5}$ (1 µg/m^3) was significantly associated with 15% higher prevalence of hypertension 95% CI: 0.7; 30.8%)	Recall bias, exposure misclassification	long term air pollution exposure was associated with an increased diastolic BP
Kim et al 2017	To study the cardiovascular effects of long-term air pollution exposure	Prospective cohort study of 196094 participants Population based followed for 7 years > 18 years of age	Korea	$PM_{2.5}$, NO_2, CO, O_3. Daily air pollution levels from monitoring stations	Cardiovascular disease incl. MI, stroke, CCF from death certificate records / ICD	every 1-lg/m3 increase of long-term exposure to fine particulate matter ≤2.5 in aerodynamic diameter was associated with a 36% increased risk of cardiovascular events 95% CI 1.32 (1.22–1.43)	Outdoor exposure measures. No indoor data confounders information bias. Home addresses used for exposure	Long term air pollution exposure increased the risk of cardiovascular disease
Cai et al 2017	To study the effect of road traffic noise and ambient air pollution on blood chemistry markers i.e., TG, glucose, CRP	Prospective cohort study 1442 participants population based >20 years of age	Norway, Netherlands	PM_{10}, NO_2, Traffic noise Land use regression modelling	Blood chemistry: hyperlipidaemia	An IQR higher PM_{10} (2.0 mg/m3) or NO_2 (7.4 mg/m3) was associated with higher triglycerides (1.9%, 95% CI: 1.5 – 2.4% and 2.2%, 95% CI: 1.6–2.7%	Misclassification of exposure	Long term PM_{10} or NO_2 exposure was associated with worse biochemistry markers
Liu et al 2017	To study the association between ambient fine particulate air pollution and hypertension	Cross – sectional study 13975 participants aged 35 – 100 years	China	$PM_{2.5}$ satellite based model with spatial resolution	Hypertension prevalence and BP Self -reported questionnaire and physical exam	$PM_{2.5}$ odds ratio of 1.11 [95% CI 1.05 - 1.17].	Reporting bias, Almost 30 % smokers, rural high unclean energy used for heating/cooking	long-term exposure to $PM_{2.5}$ was associated with increased prevalence of hypertension and higher systolic BP
Huang et al 2019	To study the effect of long-term exposure to ambient Fine $PM_{\leq 2.5}$ µm on the incidence of total, ischemic, and haemorrhagic stroke among adults	Prospective cohort study population based 900 214 person years follow up 2000 - 2015 117 575 participants >18 years	China	Fine $PM_{\leq 2.5}$ µm ($PM_{2.5}$) satellite based spatiotemporal model residential address 1 x 1 km resolution, validated monthly	Questionnaire on Ischaemic and haemorrhagic stroke, Clinical BP hospital records and death certificates	Each 10 µg/m^3 increase in $PM_{2.5}$ was significantly associated with a 13% increase in stroke incidence multivariate adjustment 95% CI (1.34 - 1.74)	Sampling bias, population level exposure. Limited information on confounders / indoor air pollution	Increased risk of stroke with long term high fine PM exposure

Bai et al 2019	To study the association between exposure to ambient air pollution and the incidence of congestive heart failure and acute myocardial infarction	Cohort study 5.1 million participants 35 - 85 years of age 1996 - 2015	Ontario, Canada	$PM_{2.5}$, NO_2, O_3 annual concentrations of pollutants to the residential postal codes of subjects y relating satellite retrievals of aerosol optical depth, a measure of light extinction by aerosols in the total atmospheric column, to $PM_{2.5}$ using a global atmospheric chemistry transport model (GEOS-Chem CTM), combined with a geographically weighted regression model grid with a spatial resolution of approximately 1 km × 1 km	incidence of Congestive cardiac failure, acute MI	each interquartile range increase in exposure were 1.05 (95% CI: 1.04–1.05) for PM2.5, 1.02 (95% CI: 1.01–1.04) for NO2	information bias - individual-level lifestyle information was unavailable	exposure to $PM_{2.5}$, O_3, were positively associated with AMI
Ljungman et al 2019	To study the long-Term Exposure to Particulate Air Pollution, Black Carbon, and Their Source Components in Relation to Ischemic Heart Disease and Stroke	Cohort study 114,758 participants from 4 cohorts 1990- 2011 25 - 64 years of age (PPS) cohort and the GOT-MONICA (Multinational Monitoring of Trends and Determinants in Cardiovascular Diseases	Gothenburg, Stockholm, and Umeå	PM_{10}, $PM_{2.5}$ detailed emission databases, monitoring data, and high-resolution dispersion models	IHD	an IQR (1:94 µg m^3) of total $PM_{2,5}$ at residency in the preceding 1- to 5-y time window was associated with a 6.5% (95% CI: −0:5, 14) higher risk of incident IHD	Exposure misclassification - unable to exclude or account for indoor sources of air pollution or other local sources such as shipping or industry information bias - missing covariate information on participants with possible confounding outcome misclassification - symptoms may	no associations between total levels of air pollutants and incident IHD
							have preceded date of diagnosis	

Arku et al 2020	To study the association of long-term exposure to Outdoor and Household Air Pollution and Blood Pressure	Prospective cohort study 3-year period 137 809 participants Aged 35 – 70 years	21 countries 640 urban and rural communities (PURE study) low-, middle- and high-income regions: Argentina, Bangladesh, Brazil, Canada, Chile, China, Colombia, Ecuador, India, Iran, Kazakhstan, Kyrgyzstan, Malaysia, Pakistan, Palestine, Peru, Philippines, Poland, Russia, Saudi Arabia, South Africa, Sweden, Tanzania, Turkey, United Arab Emirates, Uruguay, Zimbabwe.	$PM_{2.5}$ geographically weighted regression modelling using data from satellite, simulation, and ground monitor-based sources	Hypertension Clinical BP measurement	increased OR of 1.04 (95% CI: 1.01, 1.07) for hypertension, per 10 µg/m3 increase in $PM_{2.5.}$	Confounders Varying population and socio-economic groups	Chronic exposure to outdoor $PM_{2.5}$ was associate with increased blood pressure
Liu et al 2021	To study the association between long-term exposure to ambient air pollution and road traffic noise and asthma incidence in adults: The Danish Nurse cohort	Prospective cohort study 28,731 female nurses age > 44 years 1970 - 2014	Denmark	$PM_{2.5}$, NO_2 Danish DEHM/UBM/AirGIS modelling system and road traffic noise (Lden)	ICD code	(HR) and 95% confidence intervals for asthma incidence associated with 3-year moving average exposures were 1.29 (1.03, 1.61) per 6.3 µg/m3 for $PM_{2.5}$, 1.16 (1.07, 1.27) per 8.2 µg/m3 for NO_2	information bias - first hospital contact used to determine asthma incidence, may have missed symptomatic cases sampling bias - nurses only, female	Long-term exposure to air pollution was associated with adult-asthma incidence
Bagula, 2021	To study the association between ambient air pollution and cardiorespiratory outcomes in adults	Cross sectional study Parents/guardians of 590 school pupils	South Africa	$PM_{2.5,}$ NO_2 Land-Use Regression for Particulate Matter ($PM_{2.5}$) and Nitrogen Dioxide (NO_2) at participants' homes	Questionnaire	IQR increase of 5.12 µg/m3 in $PM_{2.5}$ was significantly associated with an increased prevalence of self-reported chest-pain, [Odds ratio: 1.38 (95% CI: 1.06–1.80)]	Recall bias in outcomes prevalence - misdiagnosis	circumstantial evidence of an association between annual ambient $PM_{2.5}$ exposure and self-reported chest pain

Olaniyan et al 2022	To study the association between ambient air pollution and the risk of acute myocardial	Cohort study 2.7 million participants from CanCHEC cohort 25 - 89 years of age 2006 - 2016	Canada	$PM_{2.5}$, NO_2, O_3 ground-level $PM_{2.5}$ were derived from annual surfaces at 1 km2 resolution by relating satellite aerosol optical depth to near-	diagnosis from health administrative database	each IQR increase in exposure was found to be associated with a hazard ratio of 1.026 (95% CI: 1.007–	outcome bias - missing symptoms from participants not formally diagnosed exposure misclassification -	positive associations between long-term exposures to $PM_{2.5}$ and to a lesser degree NO_2, with incident AMI
	infarction and stroke			surface $PM_{2.5}$ concentrations using a global atmospheric chemistry transport model (GEOS-Chem CTM)		1.046) for $PM_{2.5}$, 1.025 (95% CI: 1.001–1.050) for NO_2	residential addresses used as exposure thus not accounting for work or commute exposures	and stroke hospitalizations

The review also identified that studies with more comprehensive data on confounders such as age, individual habits, household air pollution exposure, smoking status, comorbid disease especially diabetes, dietary intake, obesity and time spent outdoors are required. Studies on female participants, those who are overweight and among smokers are also lacking.
Additionally, research data is required from LMIC especially in the context of Africa and South Africa both within the urban areas and rural areas undergoing rapid urbanisation. Limited data exists on long term exposure to ambient air pollution and its effects in adults within the South African context.

Table 1 summarises the epidemiological evidence on the association between long-term PM and NO_2 exposure and cardiorespiratory outcomes for the period 2014 -2023

2. Problem Statement

Ambient air pollution has been associated with premature death of approximately 4.2 million people globally and approximately 1,1 million people in Africa in 2019. (WHO, 2019) Cardiorespiratory outcomes have been associated with long term exposure to ambient air pollution, but paucity of data on the topic exists in Africa and South Africa. The association between ambient air pollution and cardiorespiratory outcomes within lower socio – economic settings has not been extensively studied.

What is the association between ambient air pollution and cardiorespiratory outcomes in informal settlements in the Western Cape, South Africa?

3. Study Aims and Objectives

This study aims to investigate the association between 12 months of exposure to ambient air pollution ($PM_{2.5}$ and NO_2) and self – reported cardiorespiratory outcomes in adults in informal settlements within the Western Cape.

Objectives:

3.1.1 To describe the levels of ambient air pollution ($PM_{2.5}$) and nitrogen dioxide (NO_2) of adults residing in four informal settlements of the Western Cape over the 12 month study period

3.1.2 To determine the self-reported incidence of cardiorespiratory health outcomes in the study population in the Western Cape at the end of the 12 month period.

3.1.3 To assess the association between exposure of ambient air pollution ($PM_{2.5}$ and NO_2) on self - reported cardiorespiratory outcomes in the study participants.

4. Hypothesis

There is an association between exposure to ambient air pollution over 12 months and cardiorespiratory outcomes in adults residing in informal settlements in the Western Cape.

5. Methodology

Study Site

The main study investigated was conducted on school children residing in informal settlements of 4 areas in the Western Cape Three exposed areas included an urbanised industrialised area in Milnerton, Marconi-Beam; a peri-urbanised area namely Khayelitsha which is largely made up of informal settlements and the semi-arid area of Oudtshoorn. Additionally, a control area which is of a similar socioeconomic status as the 3 exposed areas namely, Masiphumele in Noordhoek, was identified due to its low air pollution score ranking. In this study guardians or parents of the children in the main study who consented to participating were included.

Study Design

This is a prospective cohort to determine the association between ambient air pollution exposure and cardiorespiratory outcomes in the adults, the parents or guardians of the pupils in the main study. The baseline study was conducted from February 2015 to September 2015, and the follow – up study after a 12 month period from February 2016 to September 2016.

Data Collection

5.3.1 Questionnaire

Trained field staff conducted interviews at the homes of the guardian or parents of the children. Interviews were conducted in participants home language, either English, isiXhosa or Afrikaans. Capturing of the results was done with the assistance of mobile phones. Items which were included in the questionnaire were: demographic characteristics, respiratory symptom history and health, cardiovascular disease, blood pressure, presence of hypercholesterolemia, occupational exposure, indoor pollutant exposure, outdoor pollutant exposure, physical activity history, stressors and smoking status and history. (see Appendix 1)

Cardiorespiratory questions that were included in the questionnaire:

Cardiorespiratory related questions:

1. Have you had wheezing or whistling in your chest at any time in the last 12 months
2. Shortness of breath in the last 12 months
3. Woken up by feeling of tight chest in the last 12 months
4. Attack of shortness of breath at rest in the last 12 months
5. Woken up by attack of shortness of breath in the last 12 months
6. Attack of shortness of breath after exercise in the last 12 months
7. Self- reported asthma Medication for asthma control
8. Self-reported doctor diagnosis of asthma
9. Self-reported chest-pain
10. Self-reported hypertension
11. Self-reported high cholesterol

An asthma symptoms score was determined by assigning a score of 1 to each positive response to the eight (8) questions relating to asthma.

5.3.2 Exposure Description

Land use regression (LUR) models were developed for the primary study between 2015 – 2016 in which the annual average outdoor concentration of $PM_{2.5}$ and NO_2 were estimated at the home addresses of each participant.(Saucy et al, 2018) The estimated annual air pollution concentrations were based on weekly measurements of $PM_{2,5}$ and NO_2 over a period of 1 year at 140 of the children's homes including 40 in each of the study areas and 20 in Masiphumelele. Seasonal and annual averages were determined and concentrations of air pollution measured by predictors of exposure such as number of household members, major traffic sources close to the home such as bus routes, train stations and highways), waste burning sites and spatial variation in annual average concentrations was determined by land – use from a geographic information system (GIS).

Regression models were developed and validated using the leave – one – out – cross-validation (LOOCV). 76% of the spatial variability in the NO_2 adjusted concentrations were explained by the annual NO_2 LUR model. 62% for summer and 77% for winter. 29% of the spatial variability in the $PM_{2.5}$ adjusted concentrations were explained by the annual $PM_{2.5}$ LUR model. 36% for summer and 29% for winter.

5.4 Data Analysis

5.4.1 Sample size calculation

The sample size was calculated using the following formula:

$$\frac{N \approx 4P\,(1-P)\,Z_{a/2} + Z_{\beta})}{(RD)^2}$$

N = sample size

$Z_{a/2}$ = the alpha risk

Z_{β} = the power

$$P = \frac{P_0 + P_1}{2}$$

P_0 = the prevalence of cardiorespiratory illness in rural areas of South Africa estimated at 21% (Mphewgwana et al, 2020)

P_1 = the prevalence of cardiorespiratory illness in exposed to adults in South Africa at 33.4% (Bagula et al, 2021)

$Z_{a/2}$ = 0.05

Z_{β} = 80% or 0.20

$$4(0.272)(1-0{,}272)(1.96+0.842) \,/\, (0.334 - 0.21)^2$$

= 147

A sample size of 147 participants was calculated based on the above formula.

5.4.2 Outcome variable

The one outcome variable was doctor diagnosed asthma – if a participant answers "Yes" to the questions: Have you ever had asthma?" "Was this confirmed by a doctor?"

The second outcome variable was an asthma symptom score based on the answering of 8 asthma related questions.

The third outcome variable was a cardiovascular symptoms score was based on 3 cardiovascular related questions.

5.4.3 Statistical Analysis

Descriptive statistics will be used to examine the characteristics of the study population, cardiorespiratory outcomes and various potential confounders. Bivariate regression will be used to determine the associations between various potential confounders and cardiorespiratory outcomes.

R version 4.3.1 will be used to perform the random – effects models and multiple logistic regression analysis for the association between ambient air pollution exposure ($PM_{2.5}$ and NO_2) and cardiorespiratory outcomes controlling for confounders. Directed acyclic graphs will be used to identify confounding variables and causal inferences. Possible confounders include age, sex, education, smoking status, work air pollution exposure and home cooking fuel exposure.

5.5 Ethical consideration

The primary study received approval by the University of Cape Town's Research Ethics Committee (ethics number: 697/2014). (see Appendix 2) and this sub-study will also be submitted for ethical approval.

The ethical principles of beneficence, non-maleficence, autonomy and justice were considered in the study. The study would benefit the communities whose participants who were included. No one was harmed during the study.

5.5.1 Consent/Assent

Consent was obtained prior to conducting the study. The care-givers signed written consent and grade 4 students who were at the average age of 10 years old were asked for assent prior to being interviewed. Students were only interviewed once the care-givers consent was obtained. No one was penalised if they refused to participate in the study. All participants were interviewed with the same questionnaire.

5.5.2 Data Management Plan

A data management plan was submitted online. The data will only be available to the researchers and confidentiality will be maintained. Consent forms were stored separately to study data and these were kept in locked filing cabinets at the University of Cape Town.

5.5.3 Benefits

The schools involved will receive a copy of the final report. The evidence obtained may benefit society in the form of allowing for environmental health protocols to be put into place in order to mitigate risk, prevent morbidity as well as to identify individuals at risk and assist them in acquiring early treatment.

Community education will occur in the forms of disseminating information regarding the effects of air pollution to the teachers, students and care-givers.

No foreseeable harms whether physical, psychological, social or economic should occur to the study participants.

6. References

1. Arku RE, Brauer M, Ahmed SH, et al. Long-term exposure to outdoor and household air pollution and blood pressure in the Prospective Urban and Rural Epidemiological (PURE) study. Environ Pollution. 2020; 262:114197
2. Bagula H, Olaniyan T, de Hoogh K, Saucy A, Parker B, Leaner J, Röösli M and Dalvie MA. Ambient Air Pollution and Cardiorespiratory Outcomes amongst Adults Residing in Four Informal Settlements in the Western Province of South Africa. International Journal of Environmental and Public Health. 2021
3. Bai, L., Shin, S., Burnett, R. T., Kwong, J. C., Hystad, P., van Donkelaar, A., Goldberg, M. S., Lavigne, E., Copes, R., Martin, R. v., Kopp, A., & Chen, H. (2019). Exposure to ambient air pollution and the incidence of congestive heart failure and acute myocardial infarction: A population-based study of 5.1 million Canadian adults living in Ontario. Environment International, 132. https://doi.org/10.1016/j.envint.2019.105004
4. Beelen R, Raaschou-Nielsen O, Stafoggia M […] Hoek G. The Effects of long-term exposure to air pollution on natural-cause mortality: An analysis of 22 European cohorts within the multicentre ESCAPE project. Lancet (2014) 383(9919) 785-795
5. Benzton J, Otsuka F, Virmani R. Mechanisms of plaque rupture and formation. Circulation Research. 2014 114:1852–1866
6. Boudrel T, Bind MA, Béjot Y, Morel O and Argacha JF. Cardiovascular effects of air pollution. Arch Cardiovasc. Disease. 2017
7. Cai Y, Hansell AL, Blangiardo M, Burton PR; BioSHaRE, de Hoogh K, Doiron D, Fortier I, Cai Y, Hansell AL, Blangiardo M, Burton PR; BioSHaRE, de Hoogh K, Doiron D, Fortier I, Gulliver J, Hveem K, Mbatchou S, Morley DW, Stolk RP, Zijlema WL, Elliott P, Hodgson S. Long-term exposure to road traffic noise, ambient air pollution, and cardiovascular risk factors in the HUNT and lifelines cohorts. Eur Heart J. 2017 Aug 1;38(29):22902296. doi: 10.1093/eurheartj/ehx263. PMID: 28575405; PMCID: PMC5837618.

8. Cai Y, Zijlema WL, Doiron D, Blangiardo M, Burton PR, Fortier I, Gaye A, Gulliver J, de Hoogh K, Hveem K, Mbatchou S, Morley DW, Stolk RP, Elliott P, Hansell AL, Hodgson S. Ambient air pollution, traffic noise and adult asthma prevalence: a BioSHaRE approach. Eur Respir J. 2017 Jan 11;49(1):1502127. doi: 10.1183/13993003.021272015. PMID: 27824608.

10. Carey IM, Atkinson RW, Kent AJ, van Staa T, Cook DG, Anderson HR. Mortality associations with long-term exposure to outdoor air pollution in a national English cohort. Am J Respir Crit Care Med. 2013 Jun 1;187(11):1226-33. doi: 10.1164/rccm.201210-1758OC. PMID: 23590261; PMCID: PMC3734610.

9. Cesaroni, G., Forastiere, F., Stafoggia, M., Andersen, Z. J., Badaloni, C., Beelen, R., Caracciolo, B., de Faire, U., Erbel, R., Eriksen, K. T., Fratiglioni, L., Galassi, C., Hampel, R., Heier, M., Hennig, F., Hilding, A., Hoffmann, B., Houthuijs, D., Jöckel, K. H., … Peters, A. (2014). Long term exposure to ambient air pollution and incidence of acute coronary events: Prospective cohort study and meta-analysis in 11 European cohorts from the escape project. BMJ (Online), 348. https://doi.org/10.1136/bmj.f7412 Cardiac Evaluation. Journal of the American Heart Association, 5(8). https://doi.org/10.1161/JAHA.116.003947

10. Chen SY, Wu CF, Lee JH, Hoffmann B, Peters A, Brunekreef B, Chu DC, Chan CC. Associations between Long-Term Air Pollutant Exposures and Blood Pressure in Elderly Residents of Taipei City: A Cross-Sectional Study. Environ Health Perspect. 2015 Aug;123(8):779-84. doi: 10.1289/ehp.1408771. Epub 2015 Mar 20. PMID: 25793646; PMCID: PMC4529013.

11. Crouse D, Peters P, Villeneuve P, Proux M, Shin H, Goldberg M, Johnson M, Wheeler A. […] Within- and between-city contrasts in nitrogen dioxide and mortality in 10 Canadian cities; A subset of the Canadian Census Health and Environment Cohort (CanCHEC) Burnett R. Journal of Exposure Science and Environmental Epidemiology (2015) 25(5) 482-489

12. Douglas P. Zipes, Peter Libby, Robert O. Bonow, Douglas L. Mann, Gordon F. Tomaselli, Eugene Braunwald Braunwald Heart Disease, 11th Edition, Chapter 52 Air Pollution and cardiovascular disease. 2018

13. Fisher S, Bellinger D, Cropper M, Kumar P, Binagwaho A, Koudenoukpo J, Park, Y, Taghian G, Landrigan P. Air pollution and development in Africa: impacts on health, the economy, and human capital. The Lancet Planetary Health (2021) 5(10) e681e688

14. Fisher, J. A., Puett, R. C., Hart, J. E., Camargo, C. A., Varraso, R., Yanosky, J. D., & Laden, F. (2016). Particulate matter exposures and adult-onset asthma and COPD in the Nurses' Health Study. In European Respiratory Journal (Vol. 48, Issue 3, pp. 921–924). European Respiratory Society. https://doi.org/10.1183/13993003.00845-2015

Gulliver J, Hveem K, Mbatchou S, Morley DW, Stolk RP, Zijlema WL, Elliott P, Hodgson S. Long-term exposure to road traffic noise, ambient air pollution, and cardiovascular risk factors in the HUNT and lifelines cohorts. Eur Heart J. 2017 Aug 1;38(29):22902296. doi: 10.1093/eurheartj/ehx263. PMID: 28575405; PMCID: PMC5837618.

15. Hart JE, Puett RC, Rexrode KM, albert CM, Laden F. Effect modification of long-term air pollution exposures and the risk of incident cardiovascular disease in US women. Journal of the American Heart Association, 4(12).

16. Hartiala, J., Breton, C. v., Tang, W. H. W., Lurmann, F., Hazen, S. L., Gilliland, F. D., & Allayee, H. (2016). Ambient Air Pollution Is Associated with the Severity of Coronary Atherosclerosis and Incident Myocardial Infarction in Patients Undergoing Elective

17. Hoek G, Ranjini M Krishnan RM, Rob Beelen R, Annette Peters A, Bart Ostro B, Bert Brunekreef B, Joel D Kaufman JD. Long-term air pollution exposure and cardio- respiratory mortality: a review. Environmental Health. 2013
https://doi.org/10.1161/JAHA.115.002301

18. Huang K, Liang F, Yang X, Liu F, Li J, Xiao Q, Chen J, Liu X, Cao J, Shen C, Yu L, Lu F, Wu X, Zhao L, Wu X, Li Y, Hu D, Huang J, Liu Y, Lu X, Gu D. Long term exposure to ambient fine particulate matter and incidence of stroke: prospective cohort study from the China-PAR project. BMJ. 2019 Dec 30;367: l6720. doi: 10.1136/bmj. l6720. PMID: 31888885; PMCID: PMC7190010

19. Huangfu P, Atkinson R. Long - term exposure to NO_2 and O_3 and all – cause and respiratory mortality: A systematic review and meta-analysis. 2020

20. Hystad, P., Larkin, A., Rangarajan, S., AlHabib, K. F., Avezum, Á., Calik, K. B. T., Chifamba, J., Dans, A., Diaz, R., du Plessis, J. L., Gupta, R., Iqbal, R., Khatib, R., Kelishadi, R., Lanas, F., Liu, Z., Lopez-Jaramillo, P., Nair, S., Poirier, P., … Brauer, M. (2020). Associations of outdoor fine particulate air pollution and cardiovascular disease in 157 436 individuals from 21 high-income, middle-income, and low-income countries (PURE): a prospective cohort study. The Lancet Planetary Health, 4(6), e235–e245. https://doi.org/10.1016/S2542-5196(20)30103-0

21. Jacquemin B, Siroux V, Sanchez M, Carsin AE, Schikowski T, Adam M, Bellisario V, Buschka A, Bono R, Brunekreef B, Cai Y, Cirach M, Clavel-Chapelon F, Declercq C, de Marco R, Nazelle A, Ducret-Stich RE, Ferretti VV, Gerbase MW, Hardy R, Heinrich J, Janson C, Jarvis D, Al Kanaani Z, Keidel D, Kuh D, Le Moual N, Nieuwenhuijsen MJ, Marcon A, Modig L.et al. Ambient Air Pollution and Adult Asthma Incidence in Six European Cohorts (ESCAPE). Environmental Health Perspectives. 2015

22. Kauffman J, Adar SA, Barr RG, Budoff M, Burke GL, Curl CL, Daviglus ML, Roux AVD, Gassett AJ, Jacobs Jr DR, Kronmal R, Larson TV, Navas-Acien A, Olives C, Sampson PD, Sheppard L, Siscovick DS, Stein JH, Szpiro AA, Watson KE. Association between air pollution and coronary artery calcification within six metropolitan areas in the USA (the Multi-Ethnic Study of Atherosclerosis and Air Pollution): a longitudinal cohort study. Volume 388, Issue 10045, P696-704, 13 August 2016.

23. Kim H, Kim J, Kim S, Kang SH, Kim HJ, Kim H, Heo J, Yi SM, Kim K, Youn TJ, Chae IH. Cardiovascular Effects of Long-Term Exposure to Air Pollution: A Population-Based Study With 900 845 Person-Years of Follow- J Am Heart Assoc. 2017;6: e007170. DOI: 10.1161/JAHA.117.007170

24. Liu C, Chen R, Zhao Y, Ma Z, Bi J, Liu Y, Meng X, Wang Y, Chen X, Li W, Kan H. Associations between ambient fine particulate air pollution and hypertension: A nationwide cross-sectional study in China. Sci Total Environ. 2017 Apr 15;584585:869-874. doi: 10.1016/j.scitotenv.2017.01.133. Epub 2017 Jan 30. PMID: 28153400; PMCID: PMC8102884.

25. Liu S, Lim YH, Pedersen M, Jorgensen JT, Amini H, Cole – Hunter T, Mehta AJ, So R, Mortensen LH, Rudi G, Westerndorp J, Loft S ,Brauner EV, Ketzel M, Hertel O, Brandt J, Jensen SS, Christensen JH, Sigsgaard T, Geels C, Frohn LM, Brboric M, Radonic J, Sekulic MT, et al. Long-term exposure to ambient air pollution and road traffic noise and asthma incidence in adults: The Danish Nurse cohort. Environmental international.2021

26. Ljungman, P. L. S., Andersson, N., Stockfelt, L., Andersson, E. M., Sommar, J. N., Eneroth, K., Gidhagen, L., Johansson, C., Lager, A., Leander, K., Molnar, P., Pedersen, N. L., Rizzuto, D., Rosengren, A., Segersson, D., Wennberg, P., Barregard, L., Forsberg, B., Sallsten, G., … Pershagen, G. (2019). Long-term exposure to particulate air pollution, black carbon, and their source components in relation to ischemic heart disease and stroke. Environmental Health Perspectives, 127(10). https://doi.org/10.1289/EHP4757

27. Mpewkgwana PM, Malema N, Monyeki KD, Mothiba TM, Makgahlela M, Lgatla N, Makgato I, Sodi T. Hypertension Prevalence and determinants among Black South African Adults in Semi-urban and Rural Areas. Int J. Environ. Res Public Health. 2020

28. Newell, K. Kartsonaki , Lam KBH, Kurm OP Cardiorespiratory health effects of particulate ambient air pollution exposure in low-income and middle-income countries: a systematic review and meta-analysis. Lancet Planet Health 2017

29. Olaniyan T, Dalvie MA, Roosli M, Naidoo RN, Künzli N, Hoogh K, Berman D, Parker B, Leaner J, Jeebhay MF. Short term seasonal effects of airborne fungal spores on lung function in a panel study of schoolchildren residing in informal settlements of the Western Cape of South Africa. Environmental Pollution. 2020

30. Olaniyan T, Jeebhay M, Roosli M, Naidoo R, Baatjjies R, Kunzil N, Tsai M, Davey M, de Hoogh K, Berman D, Parker B, Leaner J, Dalvie MA. A prospective cohort study on ambient air pollution and respiratory morbidities including childhood asthma in adolescents from the western Cape Province: study protocol. BMC Public Health. 2017.

31. Olaniyan T, Jeebhay M, Röösli M, Naidoo RN, Künzlib N, de Hoogh K, Saucy A, Badpa M, Baatjies R, Parker B, Leanerg J, Dalvie MA. The association between ambient NO2 and PM2.5 with the respiratory health of school children residing in informal settlements: A prospective cohort study. Environmental research. 2020

32. Pitchika A, Hampel R, Wolf K, Kraus U, Cyrys J, Babisch W, Peters A, Schneider A. Longterm associations of modelled and self-reported measures of exposure to air pollution and noise at residence on prevalent hypertension and blood pressure. Sci Total Environ. 2017 Sep 1;593-594:337-346. doi: 10.1016/j.scitotenv.2017.03.156. Epub 2017 Mar 27. PMID: 28346907.

33. Saucy et al. Land Use Regression Modelling of Outdoor NO2 and PM2.5 Concentrations in Three Low Income Areas in the Western Cape Province, South Africa. International Journal of Environmental and Public Health.2018
34. Vallero, D. Fundamentals of Air Pollution (Fifth Edition) Chapter 7 - Air Pollutant Hazards. Pages 197-214. 2014
35. Wang M, Beelen R, […] Hoek G Long-term exposure to elemental constituents of particulate matter and cardiovascular mortality in 19 European cohorts: Results from the ESCAPE and TRANSPHORM projects. Environment International (2014) 66 97-106
36. Wichmann J, Voyi K. Ambient Air Pollution Exposure and Respiratory, Cardiovascular and Cerebrovascular Mortality in Cape Town, South Africa: 2001–2006. Int. J. Environ. Res. Public Health 2012, 9, 3978-4016
37. Wu TJ, Wu CF, Chen BY, Lee YL, Guo YL. Age of asthma onset and vulnerability to ambient air pollution: an observational population-based study of adults from Southern Taiwan. BMC Pulm Med. 2016 Apr 19;16(1):54. doi: 10.1186/s12890-0160218-0. PMID: 27094099; PMCID: PMC4837625.
38. www.epa.gov/pm-pollution
39. South African Medical Research Council, 2nd Burden of Disease Study, 2016
40. World Health Organization, Health Topics: Air pollution. 2018 http://www.who.int/airpollutionguidelines/en
41. www.environmentalpollutioncentres.org/air 2021

Appendix 2: Adult questionnaire

Do you have any question for me before I start?	Yes / No
What is the adult's study number?	
What is the study area?	Khayelitsha Oudtshoorn Phoenix Grabouw Noordhoek
Is respondent male or female?	Male Female
What is your date of birth?	
Are you the head of household?	Yes No
What is the highest grade or year of school you completed? [READ CHOICES]	Never attended school or only pre-school Primary school High school Higher Education (College/Technikon/University) Refused
In which town, province and country were you born?	
What is your home language?	IsiXhosa Afrikaans English Other
If other; Please specify language	
Have you lived anywhere else apart from your current home?	Yes No
In how many towns have you lived before?	

What is the name of the town you lived before?	
How many years did you live in the town mentioned above?	
Have you had wheezing or whistling in your chest at any time in the last 12 months?	Yes No
Have you been out of breath when the wheezing noise was present?	Yes No
Have you had this wheezing or whistling when you did not have a cold?	Yes No
Have you woken up with a feeling of tightness in your chest at any time in the last 12 months?	Yes No
In the last 12 months, have you had shortness of breath even when you are resting?	Yes No
In the last 12 months, have you had shortness of breath after heavy exercise or work?	Yes No
In the last 12 months, have you been woken-up by shortness of breath?	Yes No
Have you been woken by an attack of shortness of breath in the last 3 months?	Yes No
How many times a week on average have you been woken by shortness of breath in the last 3 months?	
Have you been woken by heavy coughing at any time in the last 12 months?	Yes No
Do you usually cough first thing in the morning in the winter?	Yes No
Do you usually cough during the day, or at night, in the winter?	Yes No
Do you cough like this for 3 months every year?	Yes No

Do you usually bring up any phlegm from your chest at anytime of the day, in the winter?	Yes No

Do you usually bring up any phlegm from your chest first thing in the morning in the winter?	Yes No
Do you bring up phlegm like this for more than 3 months every year?	Yes No
Are you disabled from walking?	Yes No
Due to what condition?	
Do you get shortness of breath when walking quickly on level ground or walking uphill?	Yes No
Do you have to shortness of breath when walking at your own pace on level ground?	Yes No
Do you get respiratory symptoms (such as wheeze, tightness in your chest or shortness of breath) during your monthly cycle?	yes No Does not apply to me (i.e. amenorrhea)
Have you ever had asthma?	Yes No
Was this confirmed by a doctor?	Yes No
How old were you when you first had asthma?	
How old were you when you last had asthma?	
How many times did you have asthma in the last 12 months?	
How many times did you have asthma in the last 3 months?	
Are you currently taking any medicines including inhalers, aerosols or tablets for asthma?	Yes No

Do you have a peak flow meter of your own?	Yes No
How many times have you used it over the last 3 months?	

Do you have written instructions from your doctor on how to manage your asthma if it gets worse or if you have an attack?	Yes No
Have you ever noticed that your asthma got worse with your monthly cycle?	Yes, in the week before my period Yes, during my period Yes, in the week after my period Yes, another time of the month Does not apply to me (i.e., amenorrhoeal) No
Have you been pregnant (at least 25 weeks) since your asthma started?	Yes No
What happened to your asthma during your pregnancies?	got better got worse stayed the same not the same for all pregnancies don't know Refused
Do you get pain or discomfort in your chest?	Yes No Refused Don't know
Do you get it when you walk uphill or hurry?	Yes No Never walks uphill or hurries Refused Don't know

Do you get it when you walk at an ordinary pace on level ground?	Yes No Refused Don't know

Please look at this card and show me where the pain or discomfort is located. CODE ALL THAT APPLY.PROBE FOR ADDITIONAL AREAS. HAND CARD CDQ1	1 2 3 4 5 6 7 8 Refused Don't know
Have you ever been told by a doctor or other health professional that you had hypertension (hy-per-tenshun), also called high blood pressure?	Yes No Refused Don't know
How old were you when you got hypertension or high blood pressure?	
Are you on medication for high blood pressure/hypertension?	Yes No Refused Don't know
Did a doctor tell you that your blood cholesterol level was high?	Yes No Refused Don't know

Has a doctor told you that you have any of the following (CLICK ALL THAT APPLY and REPEAT OPTIONS)	Heart attack Stroke Emphysema Thyroid Chronic bronchitis Tuberculosis None of the above

Are you currently employed?	Employed (including military service) Self employed Unemployed, looking for work Not working because of poor health Full-time house-person Full time student Retired Other
What is your current job title?	
Have you ever worked in any of the following? (CLICK ALL THAT APPLY)	Farm Factory Hospitals School Bar/Restaurant Transporter Market Mines Bakery Other
If other, please specify?	
How long have you been working/worked on this job?	

Have any of these jobs ever made your chest tight or wheezy?	Yes No
If YES above, please specify the job(s) that made your chest tight or wheezy. (CLICK ALL THAT APPLY)	Farm Factory Hospitals School Bar/Restaurant Transporter Market

	Mines Bakery Other
If other, please specify	
Have you had to leave any of these jobs because they affected your breathing?	Yes No
Describe which job(s) affected your breathing? (CLICK ALL THAT APPLY)	Farm Factory Hospitals School Bar/Restaurant Transporter Market Mines Bakery Other
If other, please specify	
When was your present home built? (years)	

Please select the type of structure/building:	Brick or concrete block house Semi-detached (double) house Brick house or room in backyard Wood house (not shack) Shack (main house, not in backyard) Shack in backyard Traditional dwelling/hut/structure made of traditional materials Caravan/tent Other
Please specify the other type of building you live in not specified above.	
How many years have you lived in your current home	

Does your home have air conditioning?	Yes No
What type of fuel do you mostly use for heating your house? (CLICK ALL THAT APPLY)	Electricity Wood Coal Gas Paraffin (Kerosene) Cattle manure / animal dung Solar (sun) I do not heat my house Charcoal Other
Please specify if you use any other heating appliance not specified	

What type of fuel do you mostly use for cooking? (CLICK ALL THAT APPLY)	Electricity Wood Coal Gas Paraffin (Kerosene) Cattle manure / animal dung Solar (sun) heat Charcoal Other
Please specify if you use any other kind of cooking fuel not specified above.	
When you were five years old, what fuel did you mostly use for cooking in the home you lived in then?	Electricity Wood Coal Gas Paraffin (kerosene) Cattle manure / animal dung

	Solar (sun) heat Charcoal Other
What other fuel did you use for cooking?	
Over the last four weeks, how many hours on average did you spent cooking each day?	
Over the last four weeks when you were cooking did you have a door or window to the outside air open	most of the time some of time rarely (or only occasionally) I do not have a door or window that opens to the outside in my kitchen
Do you have an extractor fan over the cooker?	Yes No

When cooking, do you use the fan?	all of the time some of the time none of the time
Do you have any of the following	have fitted carpets covering the whole floor contain rugs have double glazing windows None of the above
How old is the oldest carpet or rug? (years)	
Does your bedroom have....	have fitted carpets covering the whole floor? contain rugs? have double glazing? None of the above
How old is the oldest carpet or rug in your bedroom	less than one year 1-5 years old more than 5 years old
Do you sleep with the windows open	all of the time? sometimes?

	only occasionally? never?
Do you sleep with the windows open at night during winter?	Yes No
Has there been any water damage to the building or its contents, for example, from broken pipes, leaks or floods?	Yes No Don't know
Has there been any water damage in the last 12 months?	Yes No Don't know

Within the last 12 months have you had any damp spots inside your home?	Yes No
Did you do any of the following to reduce allergies in your house? (CLICK ALL THAT APPLY)	Remove carpet Bought a new carpet Use anti-dust mite sprays Got rid of a pets None of the above
Which of the following pets do you keep?	Cat Dog Birds No pets
Which places is the pet allowed to be in? (CLICK ALL THAT APPLY)	Inside the house In the bedroom Outside the house
How old were you when you had your first pet?	
Which of the following best describes the place you lived most when you were 5 years old?	farm village in a rural area small town suburb of a city inner city

Are you affected by animals, dust or pollen from trees or flowers?	Yes No
What time of the year do you sneeze a lot, or get itchy or watery eyes? (CLICK ALL THAT APPLY)	Winter Spring Summer Autumn

In the past month, how often did you walk a mile or more at a time without stopping?	Never More than once a day Twice a week Once a month
In the past month, did you do any of the following physical activity:	Jogging Cycling Swimming Aerobics Dancing Gardening Lift Weights None of the above
How many times in a month do you do the above physical activities?	
Ten years ago, were you more or less active than now?	
Do you have someone you can talk to about your problems or help in making decisions?	Yes No Don't need help Refused Don't know
Did you have enough emotional support in the last 12 months?	Yes No Refused Don't know
If you need extra cash, can you count on anyone to help; for example, by paying any bills, housing costs, hospital visits, or providing you with food and clothes?	Yes No Offered help but I wouldn't accept it Refused Don't know

In the last 12 months, did you feel stressed at work?	Yes No Refused Don't know
Have you smoked at least 100 cigarettes during your entire life (equivalent to about 5 packs)?	Yes No
How old were you when you first started smoking cigarettes fairly regularly?	
Do you smoke cigarettes now?	Yes No
About how many cigarettes do you smoke per day?	
For approximately how many years have you smoked this amount?	
Have you ever quit smoking because you had a health problem that was caused or made worse by smoking?	Yes No Don't know
Have you ever smoked a pipe regularly? YES means more than 350grams of tobacco in a lifetime	Yes No
How much pipe tobacco are you smoking now? (grams per week)	
Have you ever smoked cigars regularly? YES means more than 1 cigar a week for a year	Yes No
On the average, how many cigars per week do you smoke?	
You did a great job! Thank you for helping us! Do you have any questions for me?	Yes No

Appendix 3: Ethics approval of main study

UNIVERSITY OF CAPE TOWN
Faculty of Health Sciences
Human Research Ethics Committee

Room E52-24 Old Main Building
Groote Schuur Hospital
Observatory 7925
Telephone [021] 404 7682 • **Facsimile** [021] 406 6411
Email: nosi.tsama@uct.ac.za
Website: www.health.uct.ac.za/research/humanethics/forms

18 November 2014

HREC REF: 697/2014

A/Prof A Dalvie
CIDER
Public Health & Family Medicine

Dear A/Prof Dalvie

PROJECT TITLE: AN EPIDEMIOLOGICAL COHORT STUDY OF SCHOOL CHILDREN INVESTIGATING ASTHMA AND OF ADULTS INVESTIGATING CARDIOPULMONARY OUTCOMES

Thank you for submitting your study to the Faculty of Health Sciences Human Research Ethics Committee dated for review.

It is a pleasure to inform you that the HREC has **formally approved** the above-mentioned study.

Approval is granted for one year until the 30TH November 2015.

Please submit a progress form, using the standardised Annual Report Form if the study continues beyond the approval period. Please submit a Standard Closure form if the study is completed within the approval period.
(Forms can be found on our website: www.health.uct.ac.za/research/humanethics/forms)

Please note that the ongoing ethical conduct of the study remains the responsibility of the principal investigator.

Please quote the HREC reference no in all your correspondence.

Yours sincerely

PROFESSOR M BLOCKMAN
CHAIRPERSON, FHS HUMAN ETHICS
Federal Wide Assurance Number: FWA00001637.
Institutional Review Board (IRB) number: IRB00001938

This serves to confirm that the University of Cape Town Human Research Ethics Committee complies to the Ethics Standards for Clinical Research with a new drug in patients, based on the Medical Research Council (MRC-SA), Food and Drug Administration (FDA-USA), International Convention on Harmonisation Good Clinical Practice (ICH GCP) and Declaration of Helsinki guidelines.
The Human Research Ethics Committee granting this approval is in compliance with the ICH Harmonised Tripartite Guidelines E6: Note for Guidance on Good Clinical Practice (CPMP/ICH/135/95) and FDA Code Federal Regulation Part 50, 56 and 312.

Appendix 4: Approval letter from the Western Cape Education Department

Western Cape Government
Education

Directorate: Research

Audrey.wyngaard@westerncape.gov.za
tel: +27 021 467 9272
Fax: 0865902282
Private Bag x9114, Cape Town, 8000
wced.wcape.gov.za

REFERENCE: 20140917-36653
ENQUIRIES: Dr A T Wyngaard

Prof Mohamed Aqiel Dalvie
School of Public Health and Family Medicine
Health Sciences Faculty
Anzio Road
Observatory
7729

Dear Prof Mohamed Aqiel Dalvie

RESEARCH PROPOSAL: AN EPIDEMIOLOGICAL COHORT STUDY OF SCHOOL CHILDREN INVESTIGATING ASTHMA AND OF ADULTS INVESTIGATING CARDIOPULMONARY OUTCOMES

Your application to conduct the above-mentioned research in schools in the Western Cape has been approved subject to the following conditions:

1. Principals, educators and learners are under no obligation to assist you in your investigation.
2. Principals, educators, learners and schools should not be identifiable in any way from the results of the investigation.
3. You make all the arrangements concerning your investigation.
4. Educators' programmes are not to be interrupted.
5. The Study is to be conducted from **01 April 2014 till 30 April 2017**
6. No research can be conducted during the fourth term as schools are preparing and finalizing syllabi for examinations (October to December).
7. Should you wish to extend the period of your survey, please contact Dr A.T Wyngaard at the contact numbers above quoting the reference number?
8. A photocopy of this letter is submitted to the principal where the intended research is to be conducted.
9. Your research will be limited to the list of schools as forwarded to the Western Cape Education Department.
10. A brief summary of the content, findings and recommendations is provided to the Director: Research Services.
11. The Department receives a copy of the completed report/dissertation/thesis addressed to:

    The Director: Research Services
    Western Cape Education Department
    Private Bag X9114
    CAPE TOWN
    8000

We wish you success in your research.

Kind regards.
Signed: Dr Audrey T Wyngaard
Directorate: Research
DATE: 17 September 2014

Appendix 5: Consent form

Consent to participate in a survey investigating cardiopulmonary health effects in adults and asthma

in school children due to exposures to ambient air pollution and other environmental pollutants in the Western Cape

1. Title of research project

Epidemiological Cohort studies of Adults investigating Cardiopulmonary Outcomes and of School Children investigating Asthma Outcomes following Exposure to Ambient Air Pollution and Other Environmental Pollutions

2. Name of researchers

Mohamed Aqiel Dalvie (BSc, Honours, MSc, PhD)

Mohamed Jeebhay (MBChB, MMED, PhD)

Rajen Naidoo (MBChB, MMED, PhD)

Toyib Adedamola Olaniyan (BSc Hons., MSc)

Purpose of the research project

The Department of Environmental Health and Development Planning, is conducting this survey investigating the effect of air pollution and other environmental pollutants on cardiopulmonary health in adults and asthma in school children. This study is necessary and important because air pollution in the Western Cape is significant and could result in health effects in exposed persons. In order to effectively study the health effects from air pollution, it is important to follow participants for a long time. This study is therefore very important as no previous long term study following participants for three years has been done before in the Western Cape. We would like to interview you on your own health and on your child's health during this year (2015) and the following year (2016). The study will benefit residents in the Western Cape exposed to air pollution and other environmental pollutants.

3. Description of the research project

A member of our study team will interview you in privacy once at your home for about 30 minutes

during 2015 and 2016.

The questionnaires will include:

a) Child Questionnaires: You will be asked questions about your child's breathing problems, your child's medication and use of health services.

b) Adult Questionnaire: You will be asked questions about your breathing or chest problems; current and previous employment history; smoking habit (if any); home environment; and questions on your use of medications and health services.

4. Risks and discomforts of the research

There are minimal risks associated with completing the questionnaires. The only risk is a loss of confidentiality about personal information but the data will be seen only by study personnel. All reports will present aggregate data in which individuals will not be identifiable.

6. Expected benefits to you and to others

If any problems are found through the questionnaire, then you will be referred to your practitioner or local clinic for further management if any.

The results of the study would help you and others know the risks associated with various environmental pollutants for adults and children. This would further allow you to manage and/or reduce your and your child's risk. A copy of the final report of the study will be made available at the school that your child attends. Additionally, an information sheet on the risks of air pollution and how to manage these risks will be distributed to you. A seminar on the results of the study and the managing of the risks of air pollution will be held at the school after the completion of the study.

The results obtained from this questionnaire at large would help the government of the Western Cape know the degree to which health is affected by environmental pollution. This would help in further planning in reducing environmental exposures in residential areas.

7. Costs to you resulting from participation in the study

The study is offered at no cost to you. In the event a problem is discovered and you wish to be seen by a doctor for it, we can recommend to you who to see. However, the study cannot pay for these additional medical visits or treatments.

8. Confidentiality of information collected

Your and your child's name will not appear in any reports on this study. The records from the questionnaires will be kept completely confidential and will be seen only by members of the study team.

9. Documentation of the consent

One copy of this signed document will be kept together with our research records for this study. A copy of the information sheet about the study will be given to you to keep.

10. Contact person.

You may contact one of the following persons for answers to further questions about the research, your rights, or any injury you may feel is related to the study.

Name of person: Prof MA Dalvie (The principal investigator) telephone 021 4066610

Name of person: Prof. Marc Blockman (Chairperson of the Faculty of Health Sciences Human Research Ethics Committee at the University of Cape Town) telephone 021 4066496

You may contact Prof. Blockman if you have any questions or concerns about your and/or your child's rights or welfare as research participants.

11. Voluntary nature of participation

Your participation in this project is voluntary. Subsequent to your consent, you may refuse to participate in or withdraw from the study at any time without penalty or loss of benefits to which you may otherwise be entitled.

12. Consent of the participant

I have read the information given above, or it has been read to me. I understand the meaning of this information. By signing this form, I hereby consent to participate in the study. I also understand that I am free to withdraw from the study at any time without penalty.

_____ _____

Printed name of participant Signature, Mark, or Thumb Print

_____ _____

Interviewer's name (Print) Signature

_____ _____ Witness (Print) (If caregiver is illiterate) Signature

DATE: _____

Consent to participate in a survey investigating cardiopulmonary health effects in adults and asthma

in school children due to exposures to ambient air pollution and other environmental pollutants in the Western Cape

3. Title of research project

Epidemiological Cohort studies of Adults investigating Cardiopulmonary Outcomes and of School Children investigating Asthma Outcomes following Exposure to Ambient Air Pollution and Other Environmental Pollutions

4. Name of researchers

Mohamed Aqiel Dalvie (BSc, Honours, MSc, PhD)

Mohamed Jeebhay (MBChB, MMED, PhD)

Rajen Naidoo (MBChB, MMED, PhD)

Toyib Adedamola Olaniyan (BSc Hons., MSc)

Purpose of the research project

The Department of Environmental Health and Development Planning, is conducting this survey investigating the effect of air pollution and other environmental pollutants on cardiopulmonary health in adults and asthma in school children. This study is necessary and important because air pollution in the Western Cape is significant and could result in health effects in exposed persons. In order to effectively study the health effects from air pollution, it is important to follow participants for a long time. This study is therefore very important as no previous long term study following participants for three years has been done before in the Western Cape. We would like to interview you on your own health and on your child's health during this year (2015) and the following year (2016). The study will benefit residents in the Western Cape exposed to air pollution and other environmental pollutants.

3. Description of the research project

A member of our study team will interview you in privacy once at your home for about 30 minutes

during 2015 and 2016.

The questionnaires will include:

c) Child Questionnaires: You will be asked questions about your child's breathing problems, your child's medication and use of health services.

d) Adult Questionnaire: You will be asked questions about your breathing or chest problems; current and previous employment history; smoking habit (if any); home environment; and questions on your use of medications and health services.

4. Risks and discomforts of the research

There are minimal risks associated with completing the questionnaires. The only risk is a loss of confidentiality about personal information but the data will be seen only by study personnel. All reports will present aggregate data in which individuals will not be identifiable.

13. Expected benefits to you and to others

If any problems are found through the questionnaire, then you will be referred to your practitioner or local clinic for further management if any.

The results of the study would help you and others know the risks associated with various environmental pollutants for adults and children. This would further allow you to manage and/or reduce your and your child's risk. A copy of the final report of the study will be made available at the school that your child attends. Additionally, an information sheet on the risks of air pollution and how to manage these risks will be distributed to you. A seminar on the results of the study and the managing of the risks of air pollution will be held at the school after the completion of the study.

The results obtained from this questionnaire at large would help the government of the Western Cape know the degree to which health is affected by environmental pollution. This would help in further planning in reducing environmental exposures in residential areas.

14. Costs to you resulting from participation in the study

The study is offered at no cost to you. In the event a problem is discovered and you wish to be seen by a doctor for it, we can recommend to you who to see. However, the study cannot pay for these additional medical visits or treatments.

15. Confidentiality of information collected

Your and your child's name will not appear in any reports on this study. The records from the questionnaires will be kept completely confidential and will be seen only by members of the study team.

16. Documentation of the consent

One copy of this signed document will be kept together with our research records for this study. A copy of the information sheet about the study will be given to you to keep.

17. Contact person.

You may contact one of the following persons for answers to further questions about the research, your rights, or any injury you may feel is related to the study.

Name of person: Prof MA Dalvie (The principal investigator) telephone 021 4066610

Name of person: Prof. Marc Blockman (Chairperson of the Faculty of Health Sciences Human Research Ethics Committee at the University of Cape Town) telephone 021 4066496

You may contact Prof. Blockman if you have any questions or concerns about your and/or your child's rights or welfare as research participants.

18. Voluntary nature of participation

Your participation in this project is voluntary. Subsequent to your consent, you may refuse to participate in or withdraw from the study at any time without penalty or loss of benefits to which you may otherwise be entitled.

19. Consent of the participant

I have read the information given above, or it has been read to me. I understand the meaning of this information. By signing this form, I hereby consent to participate in the study. I also understand that I am free to withdraw from the study at any time without penalty.

_____ _____

Printed name of participant Signature, Mark, or Thumb Print

_____ _____

Interviewer's name (Print) Signature

_____ _____ Witness (Print) (If caregiver is illiterate) Signature

DATE: _____

Appendix 6: Ethics consent for the current study

UNIVERSITY OF CAPE TOWN
Faculty of Health Sciences
Human Research Ethics Committee

Room 45 E-52-E-Floor- Old Main Building
Groote Schuur Hospital
Observatory 7925
Telephone [021] 406 6492
Email: hrec-submissions@uct.ac.za
Website: www.health.uct.ac.za/home/human-research-ethics

20 October 2023

HREC REF: 769/2023

Prof A Dalvie
Centre for Environmental and Occupational Health Research
Falmouth Building -FHS
Email: aqiel.dalvie@uct.ac.za
Student: alltas007@gmail.com

Dear Prof Dalvie

PROJECT TITLE: THE ASSOCIATION BETWEEN AMBIENT AIR POLLUTION AND CARDIORESPIRATORY HEALTH OUTCOMES OF ADULTS RESIDING IN INFORMAL SETTLEMENTS OF THE WESTERN CAPE IN A 12 MONTH PERIOD-SUB-STUDY LINKED TO 697/2014- (MASTERS' CANDIDATE-DR TASNEEM ALLIE)

Thank you for submitting your study to the Faculty of Health Sciences Human Research Ethics Committee (HREC) for review.

It is a pleasure to inform you that the HREC has **formally approved** the above-mentioned study.

Approval is granted for one year until the 30 October 2024.

You are required to submit a progress report form, using the standardised Annual Report Form (FHS016) or (FHS017) if the study continues beyond the approval period. Please submit a Standard Closure form if the study is completed within the approval period.
(Forms can be found on our website: www.health.uct.ac.za/fhs/research/humanethics/forms)

The HREC acknowledge that the student: Dr Tasneem Allie will also be involved in this study.

Please quote HREC REF 769/2023 in all your correspondence.

Please note that the ongoing ethical conduct of the study remains the responsibility of the principal investigator.

Please note that for all studies approved by the HREC, the principal investigator **must** obtain appropriate institutional approval, where necessary, before the research may occur.

Yours sincerely

PROFESSOR M BLOCKMAN
CHAIRPERSON, FACULTY OF HEALTH SCIENCES HUMAN RESEARCH ETHICS COMMITTEE

Federal Wide Assurance Number: FWA00001637. Institutional Review Board (IRB) number: IRB00001938 NHREC-registration number: REC-210208-007
This serves to confirm that the University of Cape Town Human Research Ethics Committee complies to the Ethics Standards for Clinical Research with a new drug in patients, based on the Medical Research

Council (MRC-SA), Food and Drug Administration (FDA-USA), International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use: Good Clinical Practice (ICH GCP), South African Good Clinical Practice Guidelines (DoH 2020), based on the Association of the British Pharmaceutical Industry Guidelines (ABPI), and Declaration of Helsinki (2013) guidelines. The Human Research Ethics Committee granting this approval is in compliance with the ICH Harmonised Tripartite Guidelines E6: Note for Guidance on Good Clinical Practice (CPMP/ICH/135/95) and FDA Code Federal Regulation Part 50, 56 and 312.

Appendix 7: Author Instructions

Environmental Pollution

Supports open access

About the journal

Aims and scope

Environmental Pollution is an international peer-reviewed journal that publishes high quality research papers and review articles about all aspects of environmental pollution and its effects on ecosystems and human health. The journal welcomes high-quality process-oriented and hypothesis-based submissions that report results from original and novel research and contribute new knowledge to help address problems related to environmental pollution at a regional or global scale.

Subject areas include, but are not limited to:
• Sources and occurrences of pollutants that are clearly defined and measured in environmental compartments, food and food-related items, and human bodies;
• Interlinks between contaminant exposure and biological, ecological, and human health effects, including those of climate change;
• Contaminants of emerging concerns (including but not limited to antibiotic resistant microorganisms or genes, microplastics/nanoplastics, electronic wastes, light, and noise) and/or their biological, ecological, or human health effects;
• Laboratory and field studies on the remediation/mitigation of environmental pollution via new techniques and with clear links to biological, ecological, or human health effects;
• Modeling of pollution processes, patterns, or trends that is of clear environmental and/or human health interest;
• New techniques that measure and examine environmental occurrences, transport, behavior, and effects of pollutants within the environment or the laboratory, provided that they can be clearly used to address problems within regional or global environmental compartments.

Papers focusing on the following areas are likely to be returned to the authors without review:
• Routine surveys or monitoring programs primarily of local or regional interest;
• Descriptions of well-known contaminants, such as legacy pollutants, in yet another location;
• Studies relating to waste treatment that do not have specific relevance to pollution within the environment;
• Synthesis/fabrication of new materials solely for remediation and/or mitigation of pollution without any direct environmental relevance;
• Nitrogen or phosphorus deposition or biogeochemical processes with little or no relation to environmental consequences and/or climate change;
• Studies on eutrophication and secondary pollution by eutrophication without illuminating their governing mechanisms and factors;
• Studies within which the concentrations of toxicants used are higher than those that are typically found in an environmental pollution context. Authors of toxicology studies must justify the concentrations that they are using by reference to environmentally relevant concentrations that have been reported in the literature.

Please DO NOT ask the Editors-in-Chief for permission before submitting a manuscript. Kindly check the guidelines to determine whether your manuscript is within the scope of the journal; if yes, please go ahead and submit it.

Article types

A covering letter is required for all article types. This must explicitly express how the submission fits the Aims and Scope of *Environmental Pollution*, and should establish the ramifications of the research findings with regards to environmental quality, ecological health, and/or human health. Failure to include such justifications in the cover letter may result in returning the paper to the author.

Research Papers: Full Research Papers should not exceed 8000 words (including abstract, figures, and tables but excluding references). Please note that small tables and figures each count as 300 words, and large tables or figures with multiple panels may count for 600 or more words. There should be no more than nine figures and tables (e.g., 5 figures and 4 tables maximum) in the main text. Any additional figures and tables should be placed in Supplementary Material.

The abstract (up to 300 words), highlights and conclusions of papers in this journal must contain clear and concise statements. A graphical abstract is mandatory.

Review Articles: Authors may submit manuscripts that provide in-depth critical review of a special subject. These reviews must provide a Synthesis and Critical Evaluation of the state of the knowledge of the subject and indicate research directions. The Editors also periodically invite review articles. Manuscripts should not exceed 10,000 words, as defined above.

We also encourage Systematic Reviews that provide a comprehensive summary and critical appraisal of existing evidence. They relate to answering a research question, and are conducted using methods which seek to minimize risk of bias in results and conclusions. A systematic review should include a meta-analysis, which uses statistical techniques to pool the results of multiple individual studies into a combined summary result.

Short Communication: Short Communications may be submitted that express opinions and concerns, suggest research priorities and question conventional methodologies and conclusions. Manuscripts should include an Abstract, Introduction, Presentation of the Concerns or Analysis and Conclusions. References, Tables and Illustrations should be used sparingly. The manuscript should not exceed 12 double-spaced pages. The Editors will evaluate all manuscripts for suitability of publication, including peer review.

Correspondence: Readers are encouraged to write to any of the Editors (Letter to Editor) and raise issues and concerns about papers published in the journal. Editors or authors will be invited to submit a Reply to respond to points raised. The Editor will decide on the publication of Correspondence and Replies based on scientific merit, importance to the raised issues, and interest to the general audience. Correspondence and Replies of an unprofessional or unscientific nature, or containing personal invective, will not be considered.

Corrigendum: Authors may submit Corrigenda to alert the readership to errors discovered in their articles post-publication.

Special Issues: Proposals for Special Issues of Full Research Papers that focus on a specific topic or theme will also be considered. Special Issues will be published on emerging thematic issues and innovative conferences. An Editor or Associate Editor should be contacted early in the conference planning process to get approval and for guidelines on special issues of the journal. Furthermore, the Editors or Associate Editors will invite leading experts as Guest Editors for Special Issues. Editorials for Special issues are submitted by invitation only through the Editorial Manager portal.

Peer review

This journal follows a single anonymized review process. Your submission will initially be assessed by our editors to determine suitability for publication in this journal. If your submission is deemed suitable, it will typically be sent to a minimum of two reviewers to assess the scientific quality. The decision as to whether your article is accepted or rejected will be taken by our editors. This decision is final.

Our editors are not involved in making decisions about papers which:

- they have written themselves.
- have been written by family members or colleagues.
- relate to products or services in which they have an interest.

Any such submissions will be subject to the journal's usual procedures and peer review will be handled independently of the editor involved and their research group.

Read more about peer review.

Special issues and article collections

The peer review process for special issues and article collections follows the same process as outlined above for regular submissions, except, a guest editor will send the submissions out to the reviewers and recommend a decision to the journal editor. The journal editor oversees the peer review process of all special issues and article collections to ensure the high standards of publishing ethics and responsiveness are respected and is responsible for the final decision regarding acceptance or rejection of articles.

Open access

We refer you to our open access information page to learn about open access options for this journal.

Ethics and Policies

Ethics in publishing

Authors must follow ethical guidelines stated in Elsevier's Publishing Ethics Policy.

Submission declaration

When authors submit an article to an Elsevier journal it is implied that:

- the work described has not been published previously except in the form of a preprint, an abstract, a published lecture, academic thesis or registered report. See our policy on multiple, redundant or concurrent publication.
- the article is not under consideration for publication elsewhere.
- the article's publication is approved by all authors and tacitly or explicitly by the responsible authorities where the work was carried out.
- if accepted, the article will not be published elsewhere in the same form, in English or in any other language, including electronically without the written consent of the copyright-holder.

To verify compliance of the above, we may check your article using the plagiarism detection tool Crossref Similarity Check and other originality or duplicate checking software.

Authorship

All authors should have made substantial contributions to all of the following:

1. The conception and design of the study, or acquisition of data, or analysis and interpretation of data.
2. Drafting the article or revising it critically for important intellectual content.
3. Final approval of the version to be submitted.

All authors should agree to be accountable for all aspects of the work to ensure that the questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Changes to authorship

The editors of this journal generally will not consider changes to authorship once a manuscript has been submitted. It is important that authors carefully consider the authorship list and order of authors and provide a definitive author list at original submission.

The policy of this journal around authorship changes:

- All authors must be listed in the manuscript and their details entered into the submission system.
- Any addition, deletion or rearrangement of author names in the authorship list should only be made prior to acceptance, and only if approved by the journal editor.
- Requests to change authorship should be made by the corresponding author, who must provide the reason for the request to the journal editor with written confirmation from all authors, including any authors being added or removed, that they agree with the addition, removal or rearrangement.
- Only in exceptional circumstances will the journal editor consider the addition, deletion or rearrangement of authors post acceptance.
- Publication of the manuscript may be paused while a change in authorship request is being considered.
- Any authorship change requests approved by the journal editor will result in a corrigendum if the manuscript has already been published.
- Any unauthorised authorship changes may result in the rejection of the article, or retraction, if the article has already been published.

Declaration of interests

All authors must disclose any financial and personal relationships with other people or organizations that could inappropriately influence or bias their work. Examples of potential competing interests include:

- Employment
- Consultancies
- Stock ownership
- Honoraria

- Paid expert testimony
- Patent applications or registrations
- Grants or any other funding

The Declaration of Interests tool should always be completed.

Authors with no competing interests to declare should select the option, “I have nothing to declare”.

The resulting Word document containing your declaration should be uploaded at the "attach/upload files" step in the submission process. It is important that the Word document is saved in the .doc/.docx file format. Author signatures are not required.

We advise you to read our policy on conflict of interest statements, funding source declarations, author agreements/declarations and permission notes.

Funding sources

Authors must disclose any funding sources who provided financial support for the conduct of the research and/or preparation of the article.The role of sponsors, if any, should be declared in relation to the study design, collection, analysis and interpretation of data, writing of the report and decision to submit the article for publication.If funding sources had no such involvement this should be stated in your submission.

List funding sources in this standard way to facilitate compliance to funder's requirements:

Funding: This work was supported by the National Institutes of Health [grant numbers xxxx, yyyy]; the Bill & Melinda Gates Foundation, Seattle, WA [grant number zzzz]; and the United States Institutes of Peace [grant number aaaa].

It is not necessary to include detailed descriptions on the program or type of grants, scholarships and awards. When funding is from a block grant or other resources available to a university, college, or other research institution, submit the name of the institute or organization that provided the funding.

If no funding has been provided for the research, it is recommended to include the following sentence:

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Declaration of generative AI in scientific writing

Authors must declare the use of generative AI in scientific writing upon submission of the paper. The following guidance refers only to the writing process, and not to the use of AI tools to analyse and draw insights from data as part of the research process:

- Generative AI and AI-assisted technologies should only be used in the writing process to improve the readability and language of the manuscript.
- The technology must be applied with human oversight and control and authors should carefully review and edit the result, as AI can generate authoritative-sounding output that can be incorrect, incomplete or biased. Authors are ultimately responsible and accountable for the contents of the work.

- Authors must not list or cite AI and AI-assisted technologies as an author or co-author on the manuscript since authorship implies responsibilities and tasks that can only be attributed to and performed by humans.

The use of generative AI and AI-assisted technologies in scientific writing must be declared by adding a statement at the end of the manuscript when the paper is first submitted. The statement will appear in the published work and should be placed in a new section before the references list. An example:

- Title of new section: Declaration of generative AI and AI-assisted technologies in the writing process.
- Statement: During the preparation of this work the author(s) used [NAME TOOL / SERVICE] in order to [REASON]. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the published article.

The declaration does not apply to the use of basic tools, such as tools used to check grammar, spelling and references. If you have nothing to disclose, you do not need to add a statement.

We advise you to read our policy for authors on the use of generative AI and AI-assisted technologies for Elsevier.

Please note: to protect authors' rights and the confidentiality of their research, this journal does not currently allow the use of Generative AI or AI-assisted technologies such as ChatGPT or similar services by reviewers or editors in the peer review and manuscript evaluation process. We are actively evaluating compliant AI tools and may revise this policy in the future.

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Authors may share preprints, anywhere and at any time, in line with Elsevier's article sharing policy. Sharing preprints, such as on a preprint server, will not count as prior publication.

We advise you to read our policy on multiple, redundant or concurrent publication.

For journals following a double anonymized peer review process, submissions should not be published as a preprint until a final decision has been made.

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In support of open science this journal offers authors a free preprint posting service on SSRN to ensure early registration and dissemination of research and facilitate early citations and collaboration.Posting to SSRN is subject to SSRN's standard checks.

You will be provided with the option to release your manuscript on SSRN during the submission process. Agreeing to this option will have no effect on the editorial process or outcome, and your manuscript will remain publicly available and free to read on SSRN whether our editors accept or reject your manuscript.

You will receive an email when your preprint is posted online on SSRN and a Digital Object Identifier (DOI) is assigned.

Corresponding authors must seek approval from all co-authors before agreeing to release a manuscript publicly on SSRN.

We advise you to read about SSRN, including the SSRN Terms of Use and SSRN FAQs before selecting this option.

Use of inclusive language

Inclusive language acknowledges diversity, conveys respect to all people, is sensitive to differences, and promotes equal opportunities. Authors should ensure their work uses inclusive language throughout and contains nothing which might imply one individual is superior to another on the grounds of:

- age
- gender
- race
- ethnicity
- culture
- sexual orientation
- disability or health condition

We recommend avoiding the use of descriptors about personal attributes unless they are relevant and valid. Write for gender neutrality with the use of plural nouns ("clinicians, patients/clients") as default. Wherever possible, avoid using "he, she," or "he/she."

No assumptions should be made about the beliefs of readers and writing should be free from bias, stereotypes, slang, reference to dominant culture and/or cultural assumptions.

These guidelines are meant as a point of reference to help you identify appropriate language but are by no means exhaustive or definitive.

Reporting sex- and gender-based analyses

There is no single, universally agreed-upon set of guidelines for defining sex and gender. We offer the following guidance:

- Sex and gender-based analyses (SGBA) should be integrated into research design when research involves or pertains to humans, animals or eukaryotic cells. This should be done in accordance with any requirements set by funders or sponsors and best practices within a field.
- Sex and/or gender dimensions of the research should be addressed within the article or declared as a limitation to the generalizability of the research.
- Definitions of sex and/or gender applied should be explicitly stated to enhance the precision, rigor and reproducibility of the research and to avoid ambiguity or conflation of terms and the constructs to which they refer.

We advise you to read the Sex and Gender Equity in Research (SAGER) guidelines and the SAGER checklist (PDF) on the EASE website, which offer systematic approaches to the use of sex and gender information in study design, data analysis, outcome reporting and research interpretation.

For further information we suggest reading the rationale behind and recommended use of the SAGER guidelines.

Definitions of sex and/or gender

We ask authors to define how sex and gender have been used in their research and publication. Some guidance:

- Sex generally refers to a set of biological attributes that are associated with physical and physiological features such as chromosomal genotype, hormonal levels, internal and external anatomy. A binary sex categorization (male/female) is usually designated at birth ("sex assigned at birth") and is in most cases based solely on the visible external anatomy of a newborn. In reality, sex categorizations include people who are intersex/have differences of sex development (DSD).
- Gender generally refers to socially constructed roles, behaviors and identities of women, men and gender-diverse people that occur in a historical and cultural context and may vary across societies and over time. Gender influences how people view themselves and each other, how they behave and interact and how power is distributed in society.

Image manipulation

We accept that authors sometimes need to manipulate images for clarity but any manipulation of images for the purpose of deception or fraud will be seen as scientific ethical abuse and will be dealt with accordingly.

Authors must adhere to this journal's policy for graphical images:

- No specific feature within an image may be enhanced, obscured, moved, removed or introduced.
- Adjustments of brightness, contrast, or color balance are acceptable if, and only as long as, they do not obscure or eliminate any information present in the original image.
- Nonlinear adjustments such as changes to gamma settings must be disclosed in the figure legend.
- We do not permit the use of generative AI or AI-assisted tools to create or alter images in submitted manuscripts. Please read our policy on the use of generative AI and AI-assisted tools in figures, images and artwork.

To verify compliance with the above, this journal may send your images to a third-party service who screen for image irregularities. Our editors may ask you to provide original data or images if any questions arise as a result of the screening. The final decision as to whether images are acceptable will be taken by our editors.

Authors are encouraged to carefully check all images before submission and to connect all the data in any figures to the original, unprocessed data.

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We respect the decisions taken by our authors as to how they choose to designate territories and identify their affiliations in their published content. Our policy is to take a neutral position with respect to territorial disputes or jurisdictional claims, including, but not limited to, maps and institutional affiliations.

- Maps: Readers should be able to locate any study areas shown within maps using common mapping platforms. Maps should only show the area actually studied and authors should not include a location map which displays a larger area than the bounding box of the study area.During the review process, our editors may request authors to change maps if such cases occur.
- Institutional affiliations: Authors should use either the full, standard title of their institution or the standard abbreviation of the institutional name so that the institutional name can be independently verified for research integrity purposes.

Studies in humans and animals

Authors must follow ethical guidelines for studies carried out in humans and animals.

Studies in humans

Work which involves the use of human subjects should be carried out in accordance with the World Medical Association Declaration of Helsinki: Ethical principles for medical research involving human subjects.

Manuscripts should follow the International Committee of Medical Journal Editors (ICMJE) recommendations for the conduct, reporting, editing and publication of scholarly work in medical journals and aim to be representative of human populations in terms of sex, age and ethnicity. Sex and gender terms should be used correctly, as outlined by WHO (World Health Organization).

Manuscripts must include a statement that all procedures were performed in compliance with relevant laws and institutional guidelines and have been approved by the appropriate institutional committee(s). The statement should contain the date and reference number of the ethical approval(s) obtained.

Manuscripts must also include a statement that the privacy rights of human subjects have been observed and that informed consent was obtained for experimentation with human subjects.

This journal will not accept manuscripts that contain data derived from unethically sourced organs or tissue, including from executed prisoners or prisoners of conscience, consistent with recommendations by Global Rights Compliance on Mitigating Human Rights Risks in Transplantation Medicine. For all studies that use human organs or tissues, sufficient evidence must be provided that these were procured in line with WHO Guiding Principles on Human Cell, Tissue and Organ Transplantation. The source of the organs or tissues used in clinical research must be transparent and traceable. If your manuscript describes organ transplantation you must additionally declare within the manuscript that:

- autonomous consent free from coercion was obtained from the donor(s) or their next of kin.
- organs and/or tissues were not sourced from executed prisoners or prisoners of conscience.

Studies in animals

All animal experiments should comply with ARRIVE (Animal Research: Reporting of In Vivo Experiments) guidelines.

Studies should be carried out in accordance with Guidance on the operation of the Animals (Scientific Procedures) Act 1986 and associated guidelines, EU Directive 2010/63 for the protection of animals

used for scientific purposes or the NIH (National Research Council) Guide for the Care and Use of Laboratory Animals (PDF).

The sex of animals, and where appropriate, the influence (or association) of sex on the results of the study must be indicated and a statement included in your manuscript that such guidelines as listed above have been followed.

Writing and Formatting

File format

We ask you to provide editable source files for your entire submission (including figures, tables and text graphics). Some guidelines:

- Save files in an editable format, using the extension .doc/.docx for Word files and .tex for LaTeX files. A PDF is not an acceptable source file.
- Lay out text in a single-column format.
- Use spell-check and grammar-check functions to avoid errors.

We advise you to read our Step-by-step guide to publishing with Elsevier.

LaTeX

We encourage you use our LaTeX template when preparing a LaTeX submission. You will be asked to provide all relevant editable source files upon submission or revision.

Support for your LaTeX submission:

- LaTeX submission instructions and templates
- Journal Article Publishing Support Center LaTeX FAQs and support
- Researcher Academy's Beginners' guide to writing a manuscript in LaTeX

Title page

You are required to include the following details in the title page information:

- Article title. Article titles should be concise and informative. Please avoid abbreviations and formulae, where possible, unless they are established and widely understood, e.g., DNA).
- Author names. Provide the given name(s) and family name(s) of each author. The order of authors should match the order in the submission system. Carefully check that all names are accurately spelled. If needed, you can add your name between parentheses in your own script after the English transliteration.
- Affiliations. Add affiliation addresses, referring to where the work was carried out, below the author names. Indicate affiliations using a lower-case superscript letter immediately after the author's name and in front of the corresponding address. Ensure that you provide the full postal address of each affiliation, including the country name and, if available, the email address of each author.
- Corresponding author. Clearly indicate who will handle correspondence for your article at all stages of the refereeing and publication process and also post-publication. This responsibility includes answering any future queries about your results, data, methodology and materials. It

is important that the email address and contact details of your corresponding author are kept up to date during the submission and publication process.
- Present/permanent address. If an author has moved since the work described in your article was carried out, or the author was visiting during that time, a "present address" (or "permanent address") can be indicated by a footnote to the author's name. The address where the author carried out the work must be retained as their main affiliation address. Use superscript Arabic numerals for such footnotes.

Abstract

You are required to provide a concise and factual abstract. The abstract should briefly state the purpose of your research, principal results and major conclusions. Some guidelines:

- Abstracts must be able to stand alone as abstracts are often presented separately from the article.
- Avoid references. If any are essential to include, ensure that you cite the author(s) and year(s).
- Avoid non-standard or uncommon abbreviations. If any are essential to include, ensure they are defined within your abstract at first mention.

Keywords

You are required to provide 1 to 7 keywords for indexing purposes. Keywords should be written in English. Please try to avoid keywords consisting of multiple words (using “and” or “of”).

We recommend that you only use abbreviations in keywords if they are firmly established in the field.

Highlights

You are required to provide article highlights at submission.

Highlights are a short collection of bullet points that should capture the novel results of your research as well as any new methods used during your study. Highlights will help increase the discoverability of your article via search engines. Some guidelines:

- Submit highlights as a separate editable file in the online submission system with the word “highlights” included in the file name.
- Highlights should consist of 3 to 5 bullet points, each a maximum of 85 characters, including spaces.

We encourage you to view example article highlights and read about the benefits of their inclusion.

Graphical abstract

You are required to provide a graphical abstract at submission.

The graphical abstract should summarize the contents of your article in a concise, pictorial form which is designed to capture the attention of a wide readership. A graphical abstract will help draw more attention to your online article and support readers in digesting your research. Some guidelines:

- Submit your graphical abstract as a separate file in the online submission system.

- Ensure the image is a minimum of 531 x 1328 pixels (h x w) or proportionally more and is readable at a size of 5 x 13 cm using a regular screen resolution of 96 dpi.
- Our preferred file types for graphical abstracts are TIFF, EPS, PDF or MS Office files.

We encourage you to view example graphical abstracts and read about the benefits of including them.

Units

This journal requires you to use the international system of units (SI) which follows internationally accepted rules and conventions. If other units are mentioned within your article, you should provide the equivalent unit in SI.

Math formulae

- Submit math equations as editable text, not as images.
- Present simple formulae in line with normal text, where possible.
- Use the solidus (/) instead of a horizontal line for small fractional terms such as X/Y.
- Present variables in italics.
- Denote powers of e by exp.
- Display equations separately from your text, numbering them consecutively in the order they are referred to within your text.

Tables

Tables must be submitted as editable text, not as images. Some guidelines:

- Place tables next to the relevant text or on a separate page(s) at the end of your article.
- Cite all tables in the manuscript text.
- Number tables consecutively according to their appearance in the text.
- Please provide captions along with the tables.
- Place any table notes below the table body.
- Avoid vertical rules and shading within table cells.

We recommend that you use tables sparingly, ensuring that any data presented in tables is not duplicating results described elsewhere in the article.

Figures, images and artwork

Figures, images, artwork, diagrams and other graphical media must be supplied as separate files along with the manuscript. We recommend that you read our detailed artwork and media instructions. Some excerpts:

When submitting artwork:

- Cite all images in the manuscript text.
- Number images according to the sequence they appear within your article.
- Submit each image as a separate file using a logical naming convention for your files (for example, Figure_1, Figure_2 etc).
- Please provide captions along with the artwork.

- Text graphics may be embedded in the text at the appropriate position. If you are working with LaTeX, text graphics may also be embedded in the file.

Artwork formats

When your artwork is finalized, "save as" or convert your electronic artwork to the formats listed below taking into account the given resolution requirements for line drawings, halftones, and line/halftone combinations:

- Vector drawings: Save as EPS or PDF files embedding the font or saving the text as "graphics."
- Color or grayscale photographs (halftones): Save as TIFF, JPG or PNG files using a minimum of 300 dpi (for single column: min. 1063 pixels, full page width: 2244 pixels).
- Bitmapped line drawings: Save as TIFF, JPG or PNG files using a minimum of 1000 dpi (for single column: min. 3543 pixels, full page width: 7480 pixels).
- Combinations bitmapped line/halftones (color or grayscale): Save as TIFF, JPG or PNG files using a minimum of 500 dpi (for single column: min. 1772 pixels, full page width: 3740 pixels).

Please do not submit:

- files that are too low in resolution (for example, files optimized for screen use such as GIF, BMP, PICT or WPG files).
- disproportionally large images compared to font size, as text may become unreadable.

Figure captions

All images must have a caption. A caption should consist of a brief title (not displayed on the figure itself) and a description of the image. We advise you to keep the amount of text in any image to a minimum, though any symbols and abbreviations used should be explained.

Provide captions in a separate file.

Color artwork

If you submit usable color figures with your accepted article, we will ensure that they appear in color online.

Please ensure that color images are accessible to all, including those with impaired color vision. Learn more about color and web accessibility.

For articles appearing in print, you will be sent information on costs to reproduce color in the printed version, after your accepted article has been sent to production. At this stage, please indicate if your preference is to have color only in the online version of your article or also in the printed version.

Generative AI and Figures, images and artwork

Please read our policy on the use of generative AI and AI-assisted tools in figures, images and artwork, which states:

- We do not permit the use of Generative AI or AI-assisted tools to create or alter images in submitted manuscripts.

- The only exception is if the use of AI or AI-assisted tools is part of the research design or methods (for example, in the field of biomedical imaging). If this is the case, such use must be described in a reproducible manner in the methods section, including the name of the model or tool, version and extension numbers, and manufacturer.
- The use of generative AI or AI-assisted tools in the production of artwork such as for graphical abstracts is not permitted. The use of generative AI in the production of cover art may in some cases be allowed, if the author obtains prior permission from the journal editor and publisher, can demonstrate that all necessary rights have been cleared for the use of the relevant material, and ensures that there is correct content attribution.

Supplementary material

We encourage the use of supplementary materials such as applications, images and sound clips to enhance research. Some guidelines:

- Cite all supplementary files in the manuscript text.
- Submit supplementary materials at the same time as your article. Be aware that all supplementary materials provided will appear online in the exact same file type as received. These files will not be formatted or typeset by the production team.
- Include a concise, descriptive caption for each supplementary file describing its content.
- Provide updated files if at any stage of the publication process you wish to make changes to submitted supplementary materials.
- Do not make annotations or corrections to a previous version of a supplementary file.
- Switch off the option to track changes in Microsoft Office files. If tracked changes are left on, they will appear in your published version.

We recommend you upload research data to a suitable specialist or generalist repository. Please read our guidelines on sharing research data for more information on depositing, sharing and using research data and other relevant research materials.

Video

This journal accepts video material and animation sequences to support and enhance your scientific research. We encourage you to include links to video or animation files within articles. Some guidelines:

- When including video or animation file links within your article, refer to the video or animation content by adding a note in your text where the file should be placed.
- Clearly label files ensuring the given file name is directly related to the file content.
- Provide files in one of our recommended file formats. Files should be within our preferred maximum file size of 150 MB per file, 1 GB in total.
- Provide "stills" for each of your files. These will be used as standard icons to personalize the link to your video data. You can choose any frame from your video or animation or make a separate image.
- Provide text (for both the electronic and the print version) to be placed in the portions of your article that refer to the video content. This is essential text, as video and animation files cannot be embedded in the print version of the journal.

We publish all video and animation files supplied in the electronic version of your article.

For more detailed instructions, we recommend that you read our guidelines on submitting video content to be included in the body of an article.

Research data

We are committed to supporting the storage of, access to and discovery of research data, and our research data policy sets out the principles guiding how we work with the research community to support a more efficient and transparent research process.

Research data refers to the results of observations or experimentation that validate research findings, which may also include software, code, models, algorithms, protocols, methods and other useful materials related to the project.

Please read our guidelines on sharing research data for more information on depositing, sharing and using research data and other relevant research materials.

Option C: Research data deposit, citation and linking

For this journal, the following instructions from our research data guidelines apply.

You are **required** to:

- Deposit your research data in a relevant data repository.
- Cite and link to this dataset in your article.
- If this is not possible, make a statement explaining why research data cannot be shared.

Data statement

To foster transparency, you are required to state the availability of any data at submission.

Ensuring data is available may be a requirement of your funding body or institution. If your data is unavailable to access or unsuitable to post, you can state the reason why (e.g., your research data includes sensitive or confidential information such as patient data) during the submission process. This statement will appear with your published article on ScienceDirect.

Read more about the importance and benefits of providing a data statement.

Data linking

Linking to the data underlying your work increases your exposure and may lead to new collaborations. It also provides readers with a better understanding of the described research.

If your research data has been made available in a data repository there are a number of ways your article can be linked directly to the dataset:

- Provide a link to your dataset when prompted during the online submission process.
- For some data repositories, a repository banner will automatically appear next to your published article on ScienceDirect.

- You can also link relevant data or entities within the text of your article through the use of identifiers. Use the following format: Database: 12345 (e.g. TAIR: AT1G01020; CCDC: 734053; PDB: 1XFN).

Learn more about linking research data and research articles in ScienceDirect.

Research Elements co-submission

You are encouraged to publish research **Data**, **Methods** or **Protocols** related to your manuscript as an additional article in Data in Brief or MethodsX. Please follow the ***co-submission process*** active for this journal:

- Describe the research data, methods or protocols in a separate paper to be considered for publication in Data in Brief or in MethodsX after peer review.
- Adhere to one of the following submission templates:
    - Data article template (*Data in Brief*)
    - Methods article template (*MethodsX*)
    - Protocol article template (*MethodsX*)
- Use the option to upload your co-submission in the submission process, when submitting your original manuscript to this journal.

In case both your original research article and your co-submission article get accepted for publication, they will be linked together on ScienceDirect.

Article structure

Introduction

The introduction should clearly state the objectives of your work. We recommend that you provide an adequate background to your work but avoid writing a detailed literature overview or summary of your results.

Material and methods

The materials and methods section should provide sufficient details about your materials and methods to allow your work to be reproduced by an independent researcher. Some guidelines:

- If the method you used has already been published, provide a summary and reference the originally published method.
- If you are quoting directly from a previously published method, use quotation marks and cite the source.
- Describe any modifications that you have made to existing methods.

Results

Results should be clear and concise.We advise you to read the sections in this guide on supplying tables, artwork, supplementary material and sharing research data.

Discussion

The discussion section should explore the significance of your results but not repeat them. You may combine your results and discussion sections into one section, if appropriate. We recommend that you avoid the use of extensive citations and discussion of published literature in the discussion section.

Conclusion

The conclusion section should present the main conclusions of your study. You may have a stand-alone conclusions section or include your conclusions in a subsection of your discussion or results and discussion section.

Glossary

Please provide definitions of field-specific terms used in your article, in a separate list.

Abbreviations

Abbreviations which are not standard in the field should be defined in a footnote on the first page of your article.

Abbreviations which are essential to include in your abstract should be defined at first mention in your abstract, as well as in a footnote on the first page of your article.

Before submission we recommend that you review your use of abbreviations throughout your article to ensure that it is consistent.

Acknowledgements

Include any individuals who provided you with help during your research, such as help with language, writing or proof reading, in the acknowledgements section. Acknowledgements should be placed in a separate section which appears directly before the reference list. Do not include acknowledgements on your title page, as a footnote to your title, or anywhere else in your article other than in the separate acknowledgements section.

Author contributions: CRediT

Corresponding authors are required to acknowledge co-author contributions using CRediT (Contributor Roles Taxonomy) roles:

- Conceptualization
- Data curation
- Formal analysis
- Funding acquisition
- Investigation
- Methodology
- Project administration
- Resources
- Software

- Supervision
- Validation
- Visualization
- Writing – original draft
- Writing – review and editing

Not all CRediT roles will apply to every manuscript and some authors may contribute through multiple roles.

We advise you to read more about CRediT and view an example of a CRediT author statement.

Funding sources

Authors must disclose any funding sources who provided financial support for the conduct of the research and/or preparation of the article.The role of sponsors, if any, should be declared in relation to the study design, collection, analysis and interpretation of data, writing of the report and decision to submit the article for publication.If funding sources had no such involvement this should be stated in your submission.

List funding sources in this standard way to facilitate compliance to funder's requirements:

Funding: This work was supported by the National Institutes of Health [grant numbers xxxx, yyyy]; the Bill & Melinda Gates Foundation, Seattle, WA [grant number zzzz]; and the United States Institutes of Peace [grant number aaaa].

It is not necessary to include detailed descriptions on the program or type of grants, scholarships and awards. When funding is from a block grant or other resources available to a university, college, or other research institution, submit the name of the institute or organization that provided the funding.

If no funding has been provided for the research, it is recommended to include the following sentence:

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Appendices

We ask you to use the following format for appendices:

- Identify individual appendices within your article using the format: A, B, etc.
- Give separate numbering to formulae and equations within appendices using formats such as Eq. (A.1), Eq. (A.2), etc. and in subsequent appendices, Eq. (B.1), Eq. (B. 2) etc. In a similar way, give separate numbering to tables and figures using formats such as Table A.1; Fig. A.1, etc.

References

References within text

Any references cited within your article should also be present in your reference list and vice versa. Some guidelines:

- References cited in your abstract must be given in full.
- We recommend that you do not include unpublished results and personal communications in your reference list, though you may mention them in the text of your article.
- Any unpublished results and personal communications included in your reference list must follow the standard reference style of the journal. In substitution of the publication date add “unpublished results” or “personal communication.”
- References cited as “in press” imply that the item has been accepted for publication.

Linking to cited sources will increase the discoverability of your research.

Before submission, check that all data provided in your reference list are correct, including any references which have been copied. Providing correct reference data allows us to link to abstracting and indexing services such as Scopus, Crossref and PubMed. Any incorrect surnames, journal or book titles, publication years or pagination within your references may prevent link creation.

We encourage the use of Digital Object Identifiers (DOIs) as reference links as they provide a permanent link to the electronic article referenced. See the example below, though be aware that the format of such citations should be adapted to follow the style of other references in your paper.

DOI link example (for an article not yet in an issue):
VanDecar J.C., Russo R.M., James D.E., Ambeh W.B., Franke M. (2003). Aseismic continuation of the Lesser Antilles slab beneath northeastern Venezuela. Journal of Geophysical Research, https://doi.org/10.1029/2001JB000884.

Reference format

This journal does not set strict requirements on reference formatting at submission. Some guidelines:

- References can be in any style or format as long as the style is consistent.
- Author names, journal or book titles, chapter or article titles, year of publication, volume numbers, article numbers or pagination must be included, where applicable.
- Use of DOIs is recommended.

Our journal reference style will be applied to your article after acceptance, at proof stage. If required, at this stage we will ask you to correct or supply any missing reference data.

Reference style

Indicate references by adding a number within square brackets in the text. You can refer to author names within your text, but you must always give the reference number, e.g., "as demonstrated [3,6]. Barnaby and Jones [8] obtained a different result ....".

Number references in the order they appear in your article.

Examples:

Reference to a journal publication:

[1] J. van der Geer, T. Handgraaf, R.A. Lupton, The art of writing a scientific article, J. Sci. Commun. 163 (2020) 51 – 59. https://doi.org/10.1016/j.sc.2020.00372.

Reference to a journal publication with an article number:

[2] J. van der Geer, T. Handgraaf, R.A. Lupton, 2022. The art of writing a scientific article. Heliyon. 19, e00205. https://doi.org/10.1016/j.heliyon.2022.e00205.

Reference to a book:

[3] W. Strunk Jr., E.B. White, The Elements of Style, fourth ed., Longman, New York, 2000.

Reference to a chapter in a book:

[4] G.R. Mettam, L.B. Adams, How to prepare an electronic version of your article, in: B.S. Jones, R.Z. Smith (Eds.), Introduction to the Electronic Age, E-Publishing Inc., New York, 2020, pp. 281 - 304.

Reference to a website:

[5] Cancer Research UK, Cancer statistics reports for the UK. http://www.cancerresearchuk.org/aboutcancer/statistics/cancerstatsreport/, 2023 (accessed 13 March 2023).

Reference to a dataset:

[6] M. Oguro, S. Imahiro, S. Saito, T. Nakashizuka, Mortality data for Japanese oak wilt disease and surrounding forest compositions [dataset], Mendeley Data, v1, 2015. https://doi.org/10.1234/abc12nb39r.1.

Reference to software:

[7] E. Coon, M. Berndt, A. Jan, D. Svyatsky, A. Atchley, E. Kikinzon, D. Harp, G. Manzini, E. Shelef, K. Lipnikov, R. Garimella, C. Xu, D. Moulton, S. Karra, S. Painter, E. Jafarov, S. Molins, Advanced Terrestrial Simulator (ATS) v0.88 (Version 0.88) [software], Zenodo, March 25, 2020. https://doi.org/10.1234/zenodo.3727209.

Reference style

All citations in the text should refer to:

- Single author: the author's name (without initials, unless there is ambiguity) and the year of publication.
- Two authors: both authors' names and the year of publication.
- Three or more authors: first author's name followed by 'et al.' and the year of publication.

Citations can be made directly (or parenthetically). Groups of references can be listed either first alphabetically, then chronologically, or vice versa. Examples: “as demonstrated (Allan, 2020a, 2020b; Allan and Jones, 2019)” or “as demonstrated (Jones, 2019; Allan, 2020). Kramer et al. (2023) have recently shown”.

The list of references should be arranged alphabetically and then chronologically if necessary. More than one reference from the same author(s) in the same year must be identified by the letters 'a', 'b', 'c', etc., placed after the year of publication.

Examples:

Reference to a journal publication:

Van der Geer, J., Handgraaf, T., Lupton, R.A., 2020. The art of writing a scientific article. J. Sci. Commun. 163, 51–59. https://doi.org/10.1016/j.sc.2020.00372.

Reference to a journal publication with an article number:

Van der Geer, J., Handgraaf, T., Lupton, R.A., 2022. The art of writing a scientific article. Heliyon. 19, e00205. https://doi.org/10.1016/j.heliyon.2022.e00205.

Reference to a book:

Strunk Jr., W., White, E.B., 2000. The Elements of Style, fourth ed. Longman, New York.

Reference to a chapter in a book:

Mettam, G.R., Adams, L.B., 2023. How to prepare an electronic version of your article, in: Jones, B.S., Smith, R.Z. (Eds.), Introduction to the Electronic Age. E-Publishing Inc., New York, pp. 281–304.

Reference to a website:

Cancer Research UK, 2023. Cancer statistics reports for the UK. http://www.cancerresearchuk.org/aboutcancer/statistics/cancerstatsreport/ (accessed 13 March 2023).

Reference to a dataset:

Oguro, M., Imahiro, S., Saito, S., Nakashizuka, T., 2015. Mortality data for Japanese oak wilt disease and surrounding forest compositions [dataset]. Mendeley Data, v1. https://doi.org/10.17632/xwj98nb39r.1.

Reference to software:

Coon, E., Berndt, M., Jan, A., Svyatsky, D., Atchley, A., Kikinzon, E., Harp, D., Manzini, G., Shelef, E., Lipnikov, K., Garimella, R., Xu, C., Moulton, D., Karra, S., Painter, S., Jafarov, E., & Molins, S., 2020. Advanced Terrestrial Simulator (ATS) v0.88 (Version 0.88) [software]. Zenodo. https://doi.org/10.5281/zenodo.3727209.

Journal abbreviations

We ask you to abbreviate journal names according to the List of Title Word Abbreviations (LTWA).

Web references

When listing web references, as a minimum you should provide the full URL and the date when the reference was last accessed. Additional information (e.g. DOI, author names, dates or reference to a source publication) should also be provided, if known.

You can list web references separately under a new heading directly after your reference list or include them in your reference list.

Data references

We encourage you to cite underlying or relevant datasets within article text and to list data references in the reference list.

When citing data references, you should include:

- author name(s)
- dataset title
- data repository
- version (where available)
- year
- global persistent identifier

Add [dataset] immediately before your reference. This will help us to properly identify the dataset. The [dataset] identifier will not appear in your published article.

Preprint references

We ask you to mark preprints clearly. You should include the word “preprint” or the name of the preprint server as part of your reference and provide the preprint DOI.

Where a preprint has subsequently become available as a peer-reviewed publication, use the formal publication as your reference.

If there are preprints that are central to your work or that cover crucial developments in the topic, but they are not yet formally published, you may reference the preprint.

Reference management software

Most Elsevier journals have their reference template available in popular reference management software products. These include products that support Citation Style Language (CSL) such as Mendeley Reference Manager.

If you use a citation plug-in from these products, select the relevant journal template and all your citations and bibliographies will automatically be formatted in the journal style. We advise you to remove all field codes before submitting your manuscript to any reference management software product.

If a template is not available for this journal, follow the format given in examples in the reference style section of this Guide for Authors.

Submitting your Manuscript

Submission checklist

Before completing the submission of your manuscript, we advise you to read our submission checklist:

- One author has been designated as the corresponding author and their full contact details (email address, full postal address and phone numbers) have been provided.
- All files have been uploaded, including keywords, figure captions and tables (including a title, description and footnotes) included.
- Spelling and grammar checks have been carried out.
- All references in the article text are cited in the reference list and vice versa.
- Permission has been obtained for the use of any copyrighted material from other sources, including the Web.
- For gold open access articles, all authors understand that they are responsible for payment of the article publishing charge (APC) if the manuscript is accepted. Payment of the APC may be covered by the corresponding author's institution, or the research funder.

Suggest reviewers

To support the peer review process, we ask you to provide names and institutional email addresses of several potential reviewers for their manuscript. Some guidelines:

- Reviewers should not be colleagues or have co-authored or collaborated with you during the last three years.
- Do not suggest reviewers with whom you have competing interests.
- Suggest reviewers who are located in different countries or regions from yourself. This helps to provide a broad and balanced assessment of your work and to ensure scientific rigor.
- Consider diversity in your reviewer suggestions, such as gender, race and ethnicity and career stage.
- Do not suggest members of our Editorial Board.

The journal editors will take the final decision on whether to invite your suggested reviewers.

Submit online

Our online submission system guides you through the process steps of entering your manuscript details and uploading your files. The system converts your article files to a single PDF file used in the peer-review process.

Editable files (e.g., Word, LaTeX) are required to typeset your article for final publication. All correspondence, including notification of the editor's decision and requests for revision, is sent by email.

Please follow this link to submit your paper.

After Receiving a Final Decision

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If your manuscript is more suitable for an alternative Elsevier journal, you may receive an email asking you to consider transferring your manuscript via the Elsevier Article Transfer Service.

The recommendation could come from the journal editor, a dedicated in-house scientific managing editor, a tool-assisted recommendation or a combination.

If you agree with the recommendation, your manuscript will be transferred and independently reviewed by the editors of the new journal. You will have the opportunity to make revisions, if necessary, before the submission is complete at the destination journal.

Publishing agreement

Authors will be asked to complete a publishing agreement after acceptance. The corresponding author will receive a link to the online agreement by email. We advise you to read Elsevier's policies related to copyright to learn more about our copyright policies and your, and your employer's/institution's, additional rights for subscription and gold open access articles.

License options

Authors will be offered open access user license options which will determine how you, and third parties, can reuse your gold open access article. We advise that you review these options and any funding body license requirements before selecting a license option.

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If excerpts from other copyrighted works are included in your article, you must obtain written permission from the copyright owners and credit the source(s) within your article using Elsevier's permission request and license form (Word).

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To ensure a fast publication process we will ask you to provide proof corrections within two days.

Corresponding authors will be sent an email which includes a link to our online proofing system, allowing annotation and correction of proofs online. The environment is similar to Word. You can edit text, comment on figures and tables and answer questions raised by our copy editor. Our web-based proofing service ensures a faster and less error-prone process.

You can choose to annotate and upload your edits on the PDF version of your article, if preferred. We will provide you with proofing instructions and available alternative proofing methods in our email.

The purpose of the proof is to check the typesetting, editing, completeness and correctness of your article text, tables and figures. Significant changes to your article at the proofing stage will only be considered with approval of the journal editor.

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