Association of tuberculosis infection with asthma and respiratory symptoms in a Nordic-Baltic multicentre population study

Master Thesis

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Master in Global Health

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This thesis is submitted in partial fulfilment of the requirements for the degree of Master of Philosophy in Global Health at the Centre for International Health, University of Bergen.

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Abbreviations

BCG	Bacillus Calmette-Guérin
BMI	Body Mass Index
CI	Confidence Interval
COPD	Chronic Obstructive Pulmonary Disease
DAG	Directed acyclic graph
ECRHS	European Community Respiratory Health Survey
GINA	Global Initiative for Asthma
HIV	Human Immunodeficiency Virus
Mtb	Mycobacterium tuberculosis
OR	Odds Ratio
RHINE	Respiratory Health in Northern Europe
RHINESSA	Respiratory Health in Northern Europe, Spain, and Australia
SD	Standard deviation
TB	Tuberculosis
Th	T helper cells
WHO	World Health Organization

FOREWORDS

When I joined my master's degree in August 2020, the world was not the same as it was before. The COVID-19 pandemic ruled us; everything seemed dark at the time of restriction. As a student like others, I, too, faced many challenges. Fortunately, we were able to adapt quickly to the digital ways of teaching. Today, COVID is much less overwhelming, and it feels like the war is won. Although the pandemic made life worse, there are lots of positive things we can learn from this pandemic about preparedness, adaptation to new challenges, and care for mother earth. Also, this pandemic gave a message about the critical role of global public health.

I was passionate about respiratory diseases before joining this master's degree. My journey to learn about respiratory diseases began in 2010 while working at the National Tuberculosis Centre in Nepal. I am so lucky to meet such wonderful people working with respiratory conditions while pursuing this degree, who helped me fulfil my passion in every step.

I wrote this master thesis as "a scientific paper with a mantel." The article follows from page 26 and is prepared for submission to the *BMC: Respiratory Research*, following the journal's guidelines which are attached in the appendix.

I take this opportunity to express my humble and sincere thanks to my supervisor Dr Rajesh Shigdel, Department of Clinical Science, University of Bergen, for his meticulous guidance, support, and inspiration. I also want to extend my heartfelt thanks and gratitude to my co-supervisor, Professor Cecilie Svanes, Department of Global Public Health and Primary Care, the University of Bergen, for her academic support, encouragement, motivation, and constructive suggestions. Further, I am deeply grateful to my co-supervisor, Professor Tehmina Mustafa, Department of Global Public Health and Primary Care, the University of Bergen, for her kind support and suggestions during my study period.

Likewise, I express my heartiest gratitude to co-supervisor Dr Juan Pablo López Cervantes, PhD candidate, Department of Global Public Health and Primary Care, the University of Bergen, for his tremendous support, suggestions, and encouragement. I also thank Prof Ane Johannessen, RHINE IV coordinator, Bergen, for her continuous cooperation. In addition, I like to extend heartfelt acknowledgement to all the members of the RHINESSA research group for their kind encouragement and precious suggestions. I feel proud to be between such wonderful and creative people.

The enthusiasm and support of my respected parents and my brothers contributed significantly to completing this work. Moral support from my wife, Ranjana Gautam, played a vital role in completing my assignment. No successful event in my life has been possible without their blessings.

Finally, I would like to dedicate this thesis to my lovely daughter Aadya.

Sanjay Gyawali

1 Introduction

1.1 Tuberculosis

Tuberculosis (TB) is an infectious bacterial disease caused by the bacillus *Mycobacterium tuberculosis* (Mtb). It is a severe public health problem worldwide (1), which has affected humans for ages (2). TB is transmitted when sick people with TB expel bacteria into the air, which are inhaled by others and reach the lungs and become internalized to finally invade the respiratory epithelium. Pulmonary TB is the most common type of TB. However, TB can affect other body sites causing extrapulmonary TB. The common locations of extrapulmonary TB infection are lymph nodes, serous membranes, meninges, bones, and liver, among others (3).

The incidence of TB in a community may be affected by many factors, including the density of the population, the extent of overcrowding, the general standard of living, and the health care (4). Certain groups have a high risk of developing TB, mainly nursing home residents, people who have HIV, people with physical and psychological stress, and socially marginalised people (5). Age is also an essential determinant of the risk of developing TB after infection, and the incidence of TB is highest during late adolescence and early adulthood (6).

1.1.1 Global burden of TB

TB is the leading cause of death as caused by any infectious disease (7). It is the most common cause of death due to a single organism among persons over five years of age in low-income countries (8). The World Health Organization (WHO) estimates that in 2020, almost 10 million people were infected with TB. As per the report by WHO, TB caused 1.3 million deaths among patients who are HIV-negative and 214 000 deaths among those who are HIV-positive in 2020. Among the total infected with TB worldwide

in 2020, 56% were men and 33% were women above 15 years of age, and 11% were children below 15 years of age (9). The incidence of TB has slowly decreased in the WHO European Region, nearly reaching the 2020 milestone set by The End TB Strategy with a reduction of 19% in the TB incidence rate between 2015 and 2019 (3). **Figure 1** illustrates the estimated TB incidence rate (per 100 000 population) worldwide in 2020.

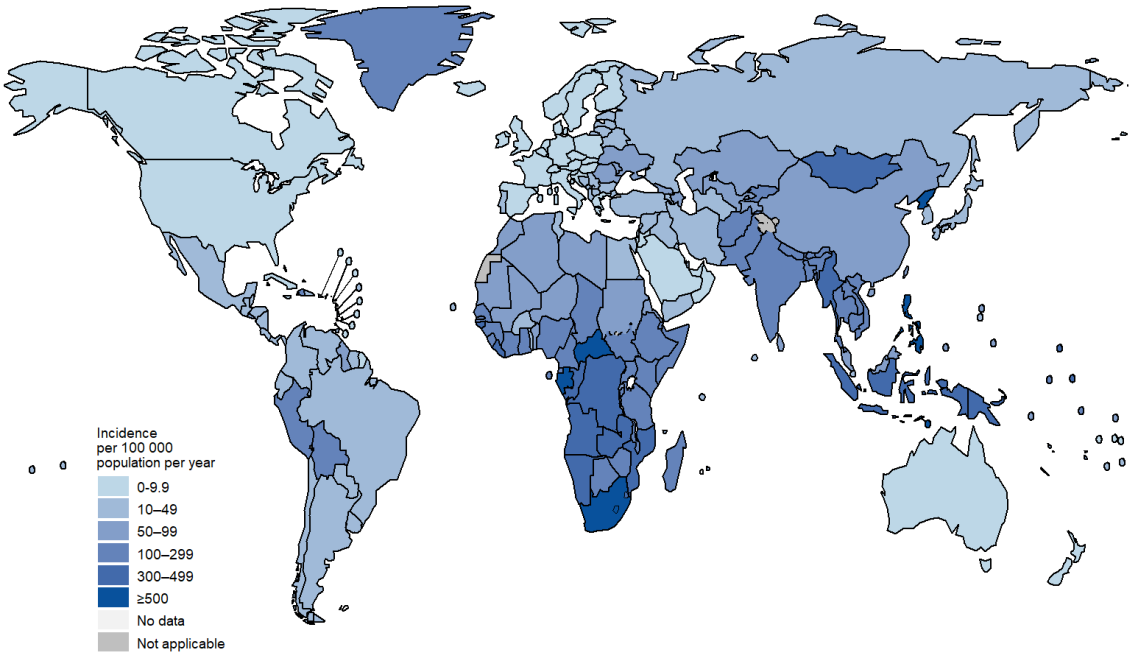


Figure 1: Estimated TB incidence rate in 2020 (per 100 000 population), by region (9)

1.1.2 TB in the Nordic region

TB was endemic in the Nordic region until the 1950s. In Norway, in the 1900s, every fifth death was caused by TB (10). The situation was similar in Sweden, where 25% of all deaths were linked to TB (11). The mortality due to TB was also high in Finland and Denmark, accounting for about 170 and 50 deaths per 100 000 population, respectively (12) (13). In Iceland, TB was responsible for 217 deaths per 100 000 people in 1911, which later decreased to 20 deaths per 100 000 in 1950 (14). In the early 1990s, the

incidence of TB was 4.1 in Denmark, 3.2 in Iceland and Sweden, 3.8 in Norway, and 10.0 in Finland per 100 000 persons, for persons born in the corresponding country. However, the incidence was 6-14 folds higher for people born outside the respective countries (15).

Due to advancements in the medical sector and improved lifestyles, cases declined in all Nordic regions after the 1950s (12) (16). According to the Norwegian Institute of Public Health, 100 to 300 cases of TB are diagnosed every year in Norway. Almost two-thirds of these cases are pulmonary TB. As shown in **Figure 2,** the incidence has decreased in recent years, with most of the actual cases occurring among immigrants, accounting for approximately 85% of new TB cases. Norwegian-born persons diagnosed with TB are predominantly older adults, which suggests they possibly were infected when tuberculosis was more common in Norway (16).

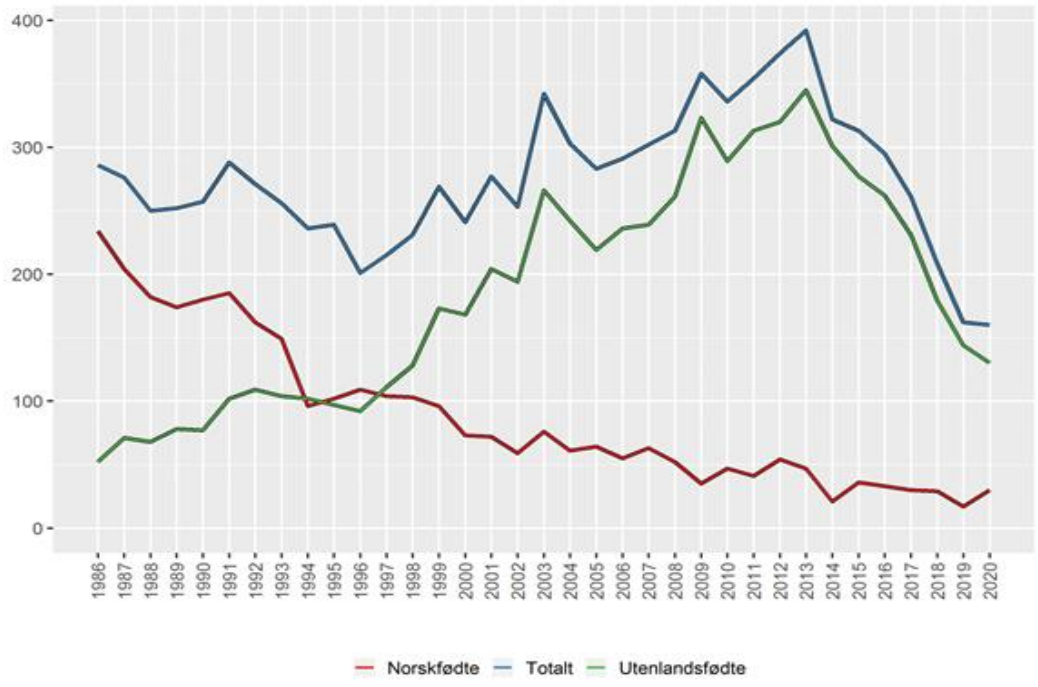


Figure 2: Tuberculosis cases in Norway 1984-2020 by place of birth; Norskfødte: born in Norway, Totalt: total, and Utenlandsfødte: born in foreign countries (17)

1.1.3 Pathogenesis of TB

Inhalation of a single tubercle bacillus can lead to infection, however, only about 15% to 20% of infected people go on to develop the disease. (18). The first infection with Mtb is known as primary TB infection which is usually subpleural. The bacilli arrive at the draining lymph nodes at the hilum of the lung within an hour of reaching the lungs, and few bacilli make it into the bloodstream. The bacteria are ingested by alveolar macrophages in the initial response, where bacteria proliferated. As a response macrophage releases chemokines and cytokines, that draw neutrophils, granulocytes, monocytes, and other inflammatory cells. To further build cellular immunity, macrophage presents the antigen to the T lymphocytes. Subsequently, the tissue necrosis occurs as a result of a delayed hypersensitivity reaction which is the classical pathology of TB (19).

The development of a mass of granulomas **(Figure 3)** close to an area of caseation leads to the appearance of the primary lesion in the lung, which is known as 'Ghon focus'. If the bacilli spread before immunity is established, secondary foci may be found in other organs. The organisms gradually lose viability, and these foci resolve after an immune response is fixed. Nevertheless, infection of Mtb may persist for many years (20).

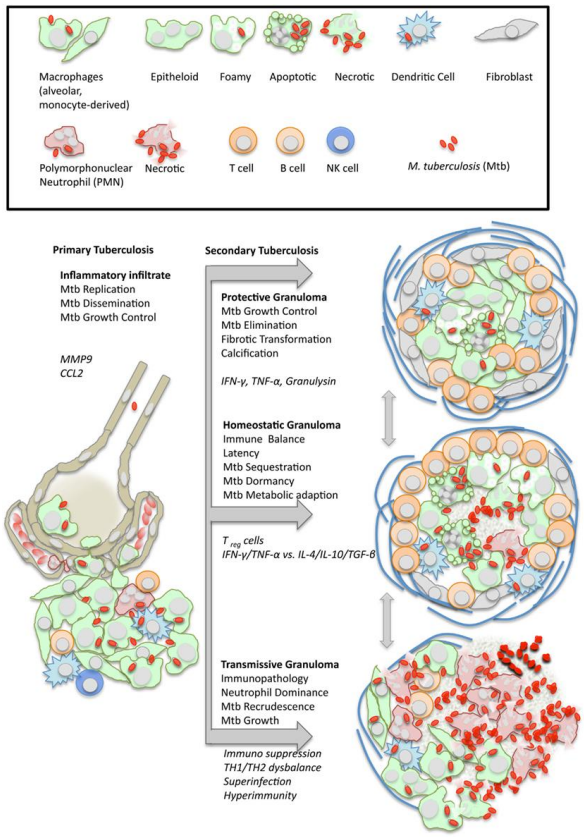


Figure 3: Granuloma formation and pathology in tuberculosis infection (21)

Post-primary TB occurs after a latent period which could be years following primary infection. Reactivation may be in response to different triggers, such as weakening of the immune system secondary to HIV infection. Though it can affect any part of the body, post-primary infection typically affects the lungs (4) (22).

1.2 Asthma

Asthma is a common chronic disease that affects people of all ages, including children and young adults. The Global Initiative for Asthma (GINA) defines asthma as *"a chronic inflammatory disorder of the airways in which many cells and cellular elements play a role. Chronic inflammation causes an associated*

increase in airway hyperresponsiveness that leads to recurrent episodes of wheezing, breathlessness, chest tightness, and coughing, particularly at night or in the early morning. These episodes are usually associated with widespread but variable airflow obstruction that is often reversible spontaneously or with treatment" (23).

The global burden of asthma has increased in the past 20 years as asthma cases have sharply increased in recent years (24). It is estimated that currently, approximately 300 million people worldwide have asthma. Furthermore, its prevalence has increased by 50% in the last decade (25).

People with asthma suffer from narrowing airways and airflow limitation, with difficulty breathing. The most common asthma symptoms are shortness of breath, wheezing, or cough. These symptoms are mainly secondary to the inflammation of bronchial tubules (26). **Figure 4** illustrates the inflammation and thickening of bronchial tubules in the asthmatic lung compared to the normal lung.

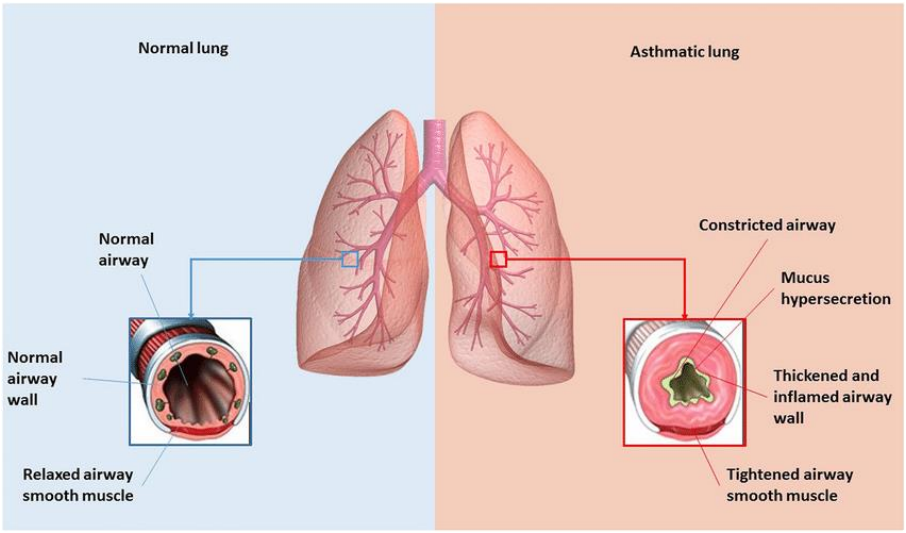
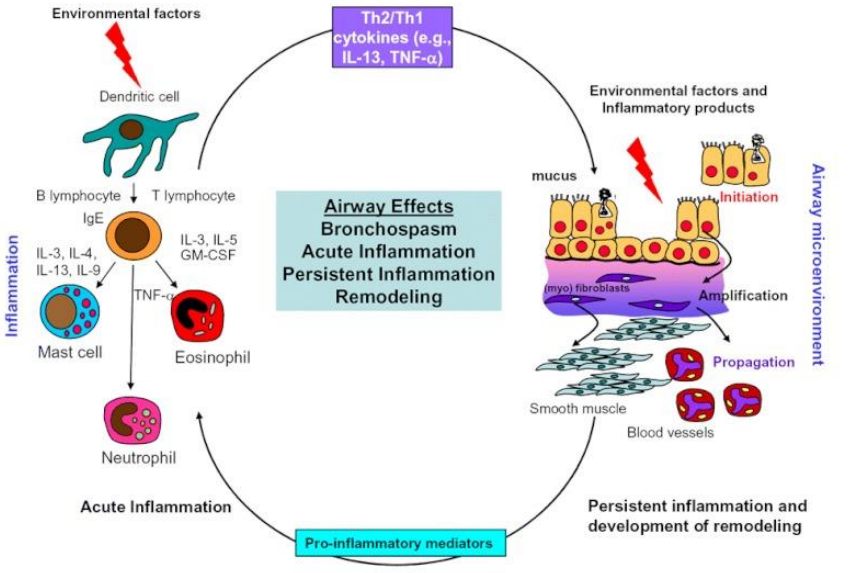


Figure 4. Airways in the normal lung as compared to the asthmatic lung (27)

Asthma can be caused and triggered by a myriad of factors, which are not completely understood. Generally, when asthma is triggered by allergens, it is classified as allergic asthma (28). If the asthma is

not related to allergens, it is referred to as non-allergic asthma. Today, non-allergic asthma and allergic asthma appear to be approximately equally common, also among children (29).

A complex multicellular process is involved in airway inflammation in people with asthma, yet many aspects are not fully understood. As seen in **Figure 5**, central cells involved in the pathology of asthma are eosinophils, CD4 T-lymphocytes, and mast cells (30). To trigger the asthma attacks associated with allergen exposure, the airways may recognize environmental allergens and generate specific Th2 cytokines in response to them. These allergens are captured and processed by dendritic cells and later presented as antigens to T cell receptors, which causes sensitization and the subsequent immune (31). Earlier, it was believed that asthma was a typical Th2-mediated disease, however, results from recent research indicate that Th2, Th17, and Th9 cells subset play an essential role in the pathophysiology of asthma (32).



Key: GM-CSF, granulocyte-macrophage colony-stimulating factor; IgE, immunoglobulin E; IL-3, interleukin 3 (and similar); TNF-α, tumor necrosis factor-alpha

Figure 5. Factors limiting airflow in acute and persistent asthma (30)

1.3 Literature review

TB may cause damage to the airways and lung tissue as described above. On the other hand, it has been suggested that the immunological changes from TB might also have a beneficial impact. The hygiene hypothesis proposed in 1989 by Strachan, states that the decrease in infectious diseases in developed countries is associated with the increase in allergic and autoimmune diseases (33). The causes behind this assumption are not entirely understood, which has led to several studies trying to prove the hypothesis in different settings (34).

Several studies have been carried out to investigate the relationship between tuberculosis and the further presence of respiratory diseases, including asthma and Chronic Obstructive Pulmonary Disease (COPD) (35-48). Included in **Table 1**, the most important evidence has been gathered to summarize the diverse efforts to shed light on the above-mentioned relationship.

Some studies show evidence that partially supports the “hygiene hypothesis”. A study conducted in Sudan compared the prevalence of asthma in patients with and without TB and concluded that infection with TB might possibly decrease the prevalence of asthma (35). Similarly, a study by Erika von Mutius et al. among children aged 13-14 years suggested that previous TB infection might decrease the lifetime prevalence of wheezing and asthma (36). In a study from India, Karahyla J. reported that among participants with asthma and TB, 69.6% developed asthma after having had TB, while 30.4% of the patients developed TB after the onset of asthma. The study emphasised that only 2.9% of participants who developed asthma after TB infection had a positive family history of asthma (37).

On the other hand, a multinational population-based cross-sectional study conducted in 18 different countries found a positive association between TB infection and obstructive pulmonary diseases (38).

Likewise, another cross-sectional study conducted in India also has similar findings and concluded that pulmonary TB infection could be one of the risk factors for COPD (39).

A review by Malay Sarkar et al. stated that TB infection could be an essential risk factor for COPD. The authors emphasised that to reduce the burden of COPD, early diagnosis and proper treatment of TB are crucial. Chronic airflow obstruction is reported in people with past TB infections and may develop during TB infection or after the treatment of TB (40). Further, infection with TB may cause scarring and bronchiectasis, which might contribute to the development of airflow obstruction (41).

Table 1: List of relevant literatures that have discussed/studied the association between asthma and TB

S.no	Author(s)/Journal	Title	Sample	Result/Conclusion in the study
1	Erika von Mutius, et al. *Thorax, 2000, Vol.55 (6), p.449-453*	International patterns of tuberculosis and the prevalence of symptoms of asthma, rhinitis, and eczema	Answer to questionnaire on respiratory symptoms from 235 477 children aged 13–14 years from 85 centres in 23 countries; analysed with TB notification rate from the WHO	TB notification rate was associated with a decrease in lifetime prevalence of wheeze and asthma (36)
2	Shirtcliffe P, et al., *Respirology (Carlton, Vic.), 2002, Vol.7 (2), p.153-155*	An inverse correlation between estimated tuberculosis notification rates and asthma symptoms	Estimated TB incidence rate and prevalence data of asthma and allergies for 6–7 and 13–14 years age groups in 38 and 55 countries respectively	An inverse relation between estimated TB incidence and prevalence of key asthma symptoms in the age 6-7 years old whereas in the age group 13-14, an inverse relationship was only found for “asthma ever” (42)
3	Pasipanodya JG, et al., *Chest. 2007;131(6):1817-24.*	Pulmonary impairment after tuberculosis	Case-control study; 107 patients with pulmonary TB and 210 latent tuberculosis infection	Pulmonary TB patients have 5.4 times more likely to have abnormal pulmonary function compared to those with latent TB infection (43)

4	Flohr C, et al., *Pediatric Allergy and Immunology. 2012 Volume 23, Issue 4 p. 324-331*	Tuberculosis, bacillus Calmette-Guérin vaccination, and allergic disease: findings from the International Study of Asthma and Allergies in Childhood Phase Two	Cross sectional study; 23 901 school children aged 8–12 years from 20 centres	Allergic diseases were linked with a history of TB (44)
5	Simone de Sousa Elias Nihues, et al. *The Brazilian journal of infectious diseases, 2015-09, Vol.19 (5), p.492-497*	Chronic symptoms and pulmonary dysfunction in post-tuberculosis Brazilian patients	Cross sectional study; 120 participants aged 18-65 years diagnosed with TB	High prevalence of chronic respiratory symptoms and pulmonary dysfunction in post-TB patients (45)
6	Amaral AFS, et al., *European Respiratory Journal. 2015;46(4):1104-12.*	Tuberculosis associates with both airflow obstruction and low lung function: BOLD results	Cross sectional study; 14 050 adult participants aged 40 years and older from 19 sites	History of TB was found to be associated with airflow obstruction (38)
7	Bashir A, Abdallah IE, Musa O. *European Respiratory Journal. 2016;48(suppl 60):PA2706.*	TB infection decreases asthma prevalence and severity of symptoms	322 adult participants with TB	The prevalence of asthma in TB patients was found to be lower as compared to the control group (35)

8	Kranti Garg, Jai Kishan Karahyla *Int J Res Med Sci. 2017 Aug;5(8):3566-3569*	Association between tuberculosis and bronchial asthma	69 patients (21-60 years old) of TB at medical college complaining about respiratory symptoms were screened	Asthma and TB can occur together, asthma found in treated cases of TB (37)
9	Mattila T, et al., *COPD: Journal of Chronic Obstructive Pulmonary Disease. 2017;14(2):143-9.*	Tuberculosis, Airway Obstruction and Mortality in a Finnish Population	Population-based study including 3 125 men and 3 576 women	Past TB is risk factor for COPD (46)
10	Tariq Mahmood, et al. *Lung India, 2017, 34 (2)*	Prevalence and etiological profile of COPD in non-smokers	Cross sectional study; 200 COPD patients older than 18 years of age	Pulmonary TB infection is one of the risk factors for COPD(39)
11	Basham CA, et al., *European Respiratory Journal. 2020:2000384.*	Post-tuberculosis airway disease: A population-based cohort study of people immigrating to British Columbia, Canada, 1985-2015	Retrospective cohort including 1 005 328 people immigrating to Canada	Increased risk of respiratory disease after TB was observed (47)

12	Mancuzo EV, et al., *J Bras Pneumol. 2020;46(2):e20180198-*	Spirometry results after treatment for pulmonary tuberculosis: comparison between patients with and without previous lung disease: a multicenter study	Cross-sectional study; 378 aged 18-50 years	Patients had spirometric changes after treated for pulmonary TB and chronic lung disease is observed after TB (48)

1.4 The rationale of the study

The exact relation between TB and respiratory diseases cannot be concluded based on available investigations. Some studies conclude that TB may increase the risk of asthma, while others find that it has an apparent protective effect. Thus, it is essential to carry out novel research in a different setting to help disentangle this complex relationship.

1.5 Research question

Does prior TB infection influence the risk of developing respiratory symptoms and/or asthma later in life?

1.6 Hypothesis

Given that TB induces profound systemic immunological and inflammatory changes, we hypothesized that having TB may influence the risk of developing respiratory symptoms and asthma.

2 Study objectives

Main objective

To investigate the association between TB and the development of asthma and respiratory symptoms in a Nordic-Baltic population-based study.

Specific objectives

1. To study the association of TB infection with subsequent asthma and respiratory symptoms.
2. To further explore the association of TB infection more specifically with allergic and non-allergic asthma.

3 Methodology

We used the data from the Respiratory Health in Northern Europe study (RHINE) to study the association of TB with asthma and respiratory symptoms. The RHINE study is a follow-up study of the participants that participated in stage 1 of the European Community Respiratory Health Survey (ECRHS, www.ecrhs.org) in 1990. In ECHRS I, young adults between 20 to 44 years were randomly selected from the population registry in respective centres. They were later followed for 10, 20, and 30 years in 2000, 2010, and 2020/2021 respectively.

3.1 Study design and data sources

The RHINE study (www.rhine.nu) is based on a standardized questionnaire (Appendix I and II) from seven study centres in the Nordic-Baltic countries: Bergen (Norway), Aarhus (Denmark), Reykjavik (Iceland), Umeå, Uppsala, and Gothenburg (Sweden), and Tartu (Estonia) (**Figure 6**). The information about participants' general characteristics, disease conditions, habits, and respiratory symptoms were collected using the questionnaire.

The analyses presented in this thesis are based on the questionnaires from RHINE III and RHINE IV. The question about our exposure was included in RHINE IV, completed recently and data from all the participants from all the centres were not ready by the time of analyses. Thus, the data in this study are preliminary, with a few participants yet to be included in each centre.

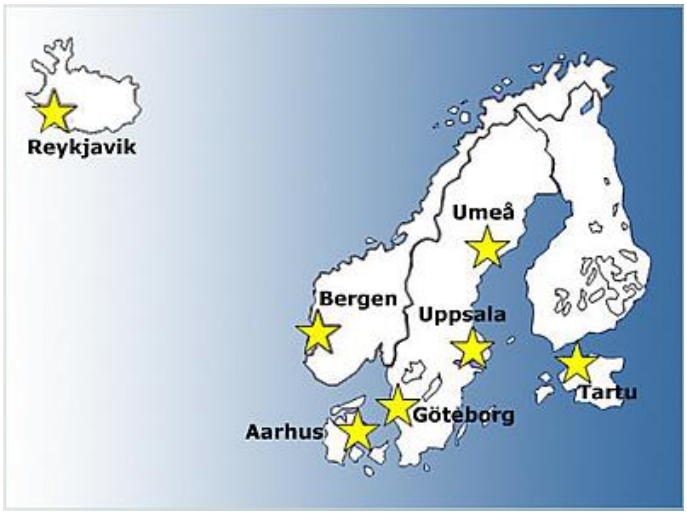


Figure 6: Study centres of the Respiratory Health in Northern Europe (RHINE) study (Source: RHINE www.rhine.nu)

3.2 Operational definitions

Exposure

Definition of Tuberculosis

A question on TB, "Have you ever had tuberculosis? (yes/no)," was included in the RHINE IV questionnaire. Those who answered "yes" to the question were considered to have TB. Those who answered yes to the above question were asked "When were you treated (for the first time) for tuberculosis?" Based on this the age of the participants when first treated for TB was calculated.

Outcomes

Asthmatic symptoms score:

During the RHINE IV study, participants were asked about asthma-like symptoms in the previous 12 months. A new variable, "asthma symptoms score," was created based on the following five questions.

1. Have you had wheezing and whistling in your chest at any time in the last 12 months? (yes/no)
2. Have you woken up with a feeling of tightness in your chest at any time in the last 12 months? (yes/no)?
3. Have you been woken by an attack of shortness of breath at any time in the last 12 months? (yes/no).
4. Have you been woken by an attack of coughing at any time in the last 12 months? (yes/no)?
5. Do you walk slower than people of the same age on the level because of breathlessness, or do you have to stop for breath when walking on your own pace on the level? (yes/no).

For each of the questions answered "yes," a score of 1 was given. If the asthma symptoms score was ≥ 3, then the symptoms score was considered high, and if the symptoms score was < 3, the symptoms score was considered low.

Definition of Asthma

In this research, asthma is defined in three different ways:

- Definition 1: Participants who answered yes to the questions "Have you had an attack of asthma in the last 12 months?" or "Are you currently taking any medicine (including inhalers, aerosols, or tablets) for asthma?" were considered as having asthma (current asthma medication/ asthma attack).
- Definition 2: Asthmatic symptoms score calculated earlier was used to define asthma. Definition 2, thus, consisted of having asthma symptoms score of ≥ 3 points (≥ 3 asthma symptoms).
- Definition 3: participants were considered to have asthma if they met definitions 1 and/or 2 (current asthma and/or ≥ 3 asthma symptoms).

Definition of Allergy

Participants who answered “yes” to the question “Do you have any nasal allergies, including hay fever?” were considered to have allergies.

Definition of allergic asthma

Participants who had asthma and nasal allergies were defined as allergic asthma.

Definition of non-allergic asthma

Non-allergic asthma was defined as asthma without nasal allergies.

Definition of respiratory symptoms

The respiratory symptoms were defined based on an affirmative answer to the respective questions. The questions used to describe these symptoms are presented in **Table 2**.

Table 2: Respiratory symptoms and their definitions

Respiratory symptoms	Questions from questionnaire
Wheezing*	Have you had wheezing or whistling in your chest at any time in the last 12 months?
Wheezing with shortness of breath	Have you been at all breathless when the wheezing noise was present?
Wheezing without cold	Have you had this wheezing or whistling when you did not have a cold?
Awoken with tightness in chest*	Have you woken up with a feeling of tightness in your chest at any time in the last 12 months?
Awoken with shortness of breath*	Have you been woken by an attack of shortness of breath at any time in the last 12 months?
Awoken with attack of cough*	Have you been woken by an attack of coughing at any time in the last 12 months?
Shortness of breath when active	Do you get breathless with strenuous exercise?
Difficult breathing when walking on ground level	Do you get short of breath when hurrying on the level or walking up a slight hill?
Breathlessness*	Do you walk slower than people of the same age on the level because of breathlessness, or do you have to stop for breath when walking on your own pace on the level?
Shortness of breath while walking	Do you stop for breath after walking about 100 metres or after a few minutes on the level?
Phlegm	Do you usually bring up phlegm or do you have phlegm in your lungs which you have difficulty bringing up?
Chronic phlegm	Do you bring up phlegm in this way almost every day for at least three months every year?
Voice tire, strain or get hoarse	Does your voice tire, strain or get hoarse when you talk? Disregard symptoms that depend on current cold or upper-airway infection. The voice symptoms may vary but try to estimate an average.
Nasal allergies	Do you have any nasal allergies including hay fever?
Attack of asthma	Have you had an attack of asthma in the last 12 months?
Currently taking asthma medication	Are you currently taking any medicine (including inhalers, aerosols or tablets) for asthma?

* Questions used to create a new variable; “asthma symptoms score.”

3.3 Inclusion and exclusion criteria

We included the participants from the RHINE IV follow-up carried out in 2020/2022 from five centres (Bergen, Reykjavik, Umeå, Uppsala, and Gothenburg). We excluded the participants who did not answer the questions for TB, asthma-like symptoms, and allergies. From the five centres, a total of 4 530 participants were included after excluding 419 participants based on exclusion criteria (**Figure 7**).

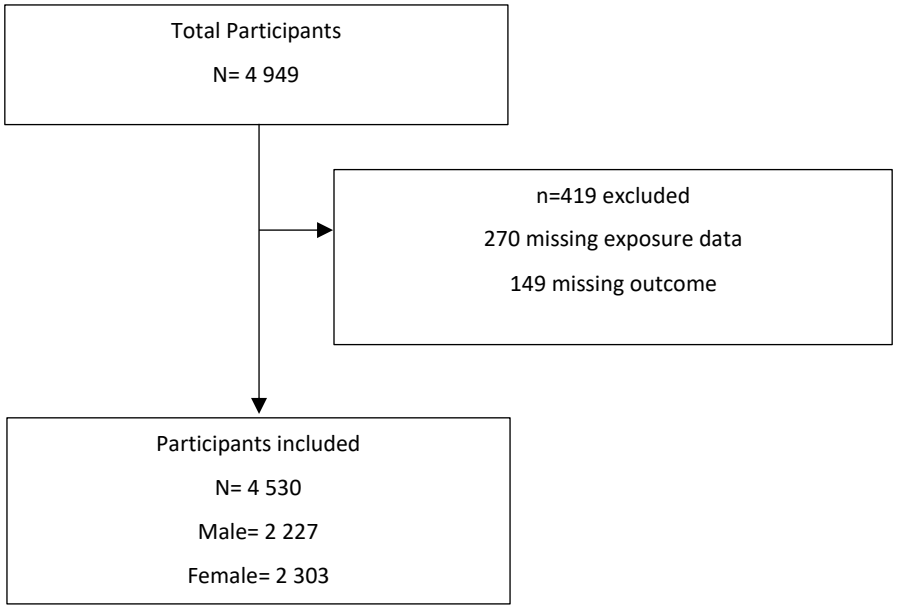


Figure 7: Flowchart of participants in the study

3.4 Covariates

We selected potential confounders based on previous knowledge and available literature. In addition to that, a directed acyclic graph (DAG) was created with DAGitty software to identify the potential confounders that needed to be adjusted (**Figure 8**). The minimal adjustment set included age, sex, smoking, BMI, and parental education (a proxy for socioeconomic status); these variables were adjusted in the analyses of the associations of TB with respiratory outcomes.

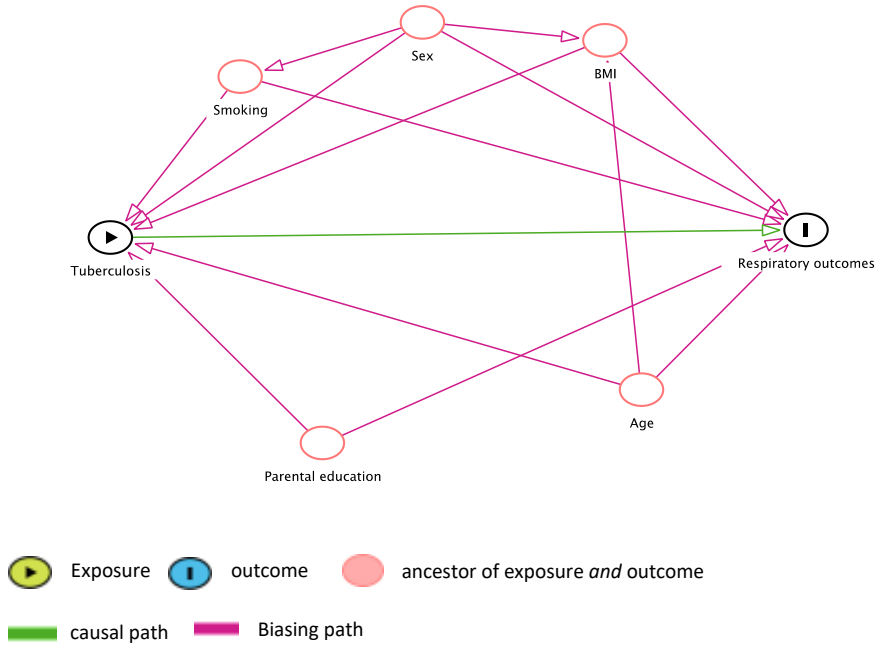


Figure 8: Direct acyclic graph linking the tuberculosis infection (exposure) to asthma (outcome)

3.5 Statistical analyses

The general characteristics of the participants were described including disease condition, airway symptoms, and asthmatic condition. Additionally, we calculated the prevalence of respiratory outcomes among participants with and without TB infection. Furthermore, the year of the first treatment for TB was also tabulated for participants who reported TB. The associations between the exposure (TB) and the outcomes (asthma/respiratory symptoms) were analysed using univariate and multivariate logistic regression, adjusting for potential confounders (age, sex, smoking, BMI, and parental education). Further stratified analysis was performed based on sex.

Stata/SE 17.0 (Stata Corp, College Station, TX, USA) software was used for the statistical analyses.

4 Ethical permissions

Written consent was obtained from all the participants at each stage of the RHINE study, and the study was approved by all the ethical committees in each country.

The names of the regional ethics committees in each study centre that approved the study are mentioned below:

- Bergen: The Regional Ethics Committee in Western Norway (Ref. nr. 2010/759)
- Gothenburg, Umeå and Uppsala: The Regional Ethical Review Board of Uppsala University (Ref. nr. 1999/313 and 2010/068)
- Reykjavik: The National Bioethics Committee, Iceland (Ref. nr. VSNb2011090016/03.11)

Results

The findings of the study are presented in the attached manuscript.

Discussion

A discussion of the study is presented in the attached manuscript.

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Association of tuberculosis infection with asthma and respiratory symptoms in a Nordic-Baltic multicentre population study

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"take-home" message of the paper

Tuberculosis might possibly contribute to increase the risk of non-allergic asthma. Tuberculosis patients need to be followed up after completion of treatment for timely detection and management of potential obstructive lung diseases.

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ABSTRACT

Background: Tuberculosis (TB) infection induces profound systemic immunological and inflammatory changes that could influence the risk of occurrence of other respiratory diseases for instance asthma. The association of TB with asthma is only partly understood.

Objective: To study the association of TB with asthma and respiratory symptoms in a Nordic-Baltic population-based study.

Methods: In this study, we included the data from five study centres (Bergen, Reykjavik, Umeå, Uppsala, and Gothenburg) of the fourth follow-up of the RHINE study. Information on general characteristics, TB infection, asthma, and asthma-like symptoms were collected using standardized questionnaires. Asthma was defined as a positive answer to questions on asthma medication and/or attack of asthma in the last twelve months ("current asthma"), and by asthma symptoms score of ≥ 3 symptoms out of five self-reported symptoms ("≥ 3 asthma symptoms"). In addition, allergic/ non-allergic asthma was defined as asthma with/ without nasal allergy. The associations of TB with asthma outcomes were analysed using logistic regressions with adjustment for age, sex, smoking, BMI, and parental education.

Results: A total of 4 530 participants aged 50 to 75 years were included, of which 35 participants reported having had TB (32 out of these before age 25 years). Approximately 40% of participants with TB reported current asthma and/or ≥ 3 asthma symptoms compared to 20% among those without TB. In adjusted analyses, participants with a history of TB had a higher risk of current asthma and/or asthma symptoms (OR: 2.47, 95%CI 1.22-4.99); particularly non-allergic asthma (OR: 3.85, 95%CI 1.85-8.01).

Conclusion: This study suggests an association of previous TB infection with asthma and respiratory symptoms. This was found for non-allergic asthma while no effect or a protective effect was indicated

for allergic asthma, supporting both a hypothesis of lung damage due to the TB infection and a potential beneficial role of TB infection regarding immune maturation and type 2 responses.

Keywords: asthma, post-tuberculosis, sequelae, allergy, RHINE

INTRODUCTION

Tuberculosis (TB) is a severe infectious disease caused mainly by *Mycobacterium tuberculosis* (Mtb). TB still represents a major global health threat, especially in low and middle-income countries, causing 1.5 million deaths worldwide in 2020 (1). The tubercle bacilli enter the human body mainly via the respiratory tract following inhalation of droplets (2). Bacilli may spread from the lungs to other body parts through the bloodstream, the lymphatic system, or direct extension (3). Therefore, TB can affect any part of the body, causing pulmonary TB where the lungs are infected and extrapulmonary TB, with peripheral lymph nodes as the most typical affected site (4).

Asthma is a chronic inflammatory disease of the airways with the common symptoms of wheezing, breathlessness, chest tightness, and coughing (5). When asthma is triggered by allergens, it is referred to as allergic asthma, otherwise, it is termed as non-allergic asthma (6). Asthma currently affects estimated 300 million people worldwide, with around 250 000 deaths annually (7).

Pulmonary TB triggers lung damage and airflow obstruction by modifying parenchyma and scaring the lungs (8). Various research has suggested that the block of airflow following TB infection is not only due to the scaring of parenchyma but also because of bronchiectasis and bronchial stenosis, both of which can be a sequela of TB disease (9). This damage to the lungs probably may increase the risk of asthma and other respiratory diseases following TB. On the other hand, the immunological response towards TB is suspected to confer protection towards Th2 skewed reactions like in allergies. The exact relation between TB and respiratory diseases is not well understood from previous investigations, as some studies conclude that TB may increase the risk of asthma, while others find that it has an apparent protective effect (10-15). Due to limited and contradictory knowledge, we aimed to investigate the association of TB with asthma and respiratory symptoms in a large community-based study in the Northern European region.

METHODOLOGY

Study design, settings, and participants

Data were obtained from standardized questionnaires where participants provided information on general characteristics, habits, and respiratory symptoms. These questionnaires were administered in the Respiratory Health in Northern Europe study (RHINE, www.rhine.nu), which is a follow-up of a population-based cohort that participated in stage 1 of the European Community Respiratory Health Survey (ECRHS, www.ecrhs.org) in 1990 from seven study centres in the Nordic-Baltic countries: Bergen, Aarhus, Reykjavik, Umeå, Uppsala, Gothenburg, and Tartu. In ECHRS I, young adults aged 20 to 44 years were randomly selected and followed up in 2000 (RHINE II), 2010 (RHINE III), and 2020/2021 (RHINE IV).

The analyses presented in this study are from a preliminary dataset based on the RHINE III and RHINE IV from five centres (Bergen, Reykjavik, Umeå, Uppsala, and Gothenburg). Of the 4 949 participants from the five centres, a total of 4 530 participants were included in the study. In total, 419 participants who did not answer the questions on exposures or outcome variables relevant for the study were excluded from the analyses.

Written consent was obtained from all the participants at each stage of the RHINE study, and each stage of the study was approved by the local ethical committee in each study centre.

Definition of exposure and outcomes

Exposure

Participants with TB were defined as people with a positive answer to the question, "Have you ever had tuberculosis?" Given that the question did not differentiate between active or latent TB, “TB”

was used indistinctively for both. Based on the question "When were you treated (for the first time) for tuberculosis?" the age of the participants when first treated for TB was calculated.

Outcomes

The respiratory outcomes were defined based on an affirmative answer to the questions presented in the supplementary data in **Table S1**.

Asthma was defined in three different ways: i) Participants were considered to have asthma if they had given a positive answer to either of the questions *"Have you had an attack of asthma in the last 12 months?*" or "*Are you currently taking any medicine (including inhalers, aerosols, or tablets) for asthma?"* (*Definition 1*, current asthma medication/ asthma attack); ii) The five self-reported asthma-like symptoms: wheezing, awoken with tightness in the chest, awoken with shortness of breath, awoken with an attack of cough, and breathlessness **(Table S2),** were used to define an asthma symptom score (assigning one point to each question). *Definition 2*, thus, consisted of having asthma symptoms score of ≥ 3 points (≥ 3 asthma symptoms); iii) As per *definition 3*, participants were considered to have asthma if they met definitions 1 and/or 2 (current asthma and/or ≥ 3 asthma symptoms).

Furthermore, allergic outcomes were described based on the aforementioned definitions of asthma/ symptoms and a positive answer to the question: "*Do you have any nasal allergies, including hay fever?*". Similarly, non-allergic asthma was defined as having asthma without nasal allergy.

Statistical analyses

Potential confounders were assessed using a directed acyclic graph (DAG) based on currently available evidence. The minimal adjustment set included age, sex, smoking, BMI, and parental

education (a proxy for socioeconomic status); these variables were adjusted in the analyses of the associations of TB with respiratory outcomes.

First, we calculated the prevalence of respiratory outcomes among participants with and without TB infection. Further, we performed logistic regression analyses to study the crude and adjusted association of TB with respiratory outcomes. The odds ratio (OR) with a 95% confidence interval (CI) were calculated for both unadjusted and adjusted models. The statistical analyses were performed using Stata/SE 17.0 (Stata Corp, College Station, TX, USA).

RESULTS

The general characteristics of the study population related to TB status are presented in **Table 1.** The analyses included 4 530 participants, of which 35 participants reported TB. Among those who reported having had TB, 48.6% were males, and 51.4% were ex-smoker. The participants were born between 1945 and 1971.

Table 1: Basic characteristics of participants of a Nordic-Baltic general population study according to the TB status (preliminary data)

	All participants (N=4 530)	With TB (n=35)	Without TB (n=4 495)
Sex, n (%)			
Male	2 227 (49.2)	17 (48.6)	2 210 (49.1)
Female	2 303 (50.8)	18 (51.4)	2 285 (50.8)
Birthyear (Range)	1945-1971	1946-1970	1945-1971
Mean age (SD)	63.1 (7.0)	67.3 (5.8)	62.8 (6.9)
Smoking, n (%)			
Never	2 395 (52.9)	14 (40.0)	2 381 (53.0)
Current	304 (6.7)	3 (8.6)	301 (6.7)
Ex-smoker	1 831 (40.4)	18 (51.4)	1 813 (40.3)
Parental education (any of the parents), n (%)			
Primary school	1 498 (33.1)	14 (40.0)	1 484 (33.0)
Secondary school	1 202 (26.5)	11 (31.4)	1 191 (26.5)
College or university	815 (18.0)	4 (11.4)	811 (18.0)
Do not know of both	1 015 (22.4)	6 (17.1)	1 009 (22.5)
Mean height (SD)	173.3 (9.4)	171.5 (9.7)	173.3 (9.4)
Mean weight (SD)	80.6 (16.1)	81.6 (16.2)	80.6 (16.5)
Mean BMI (SD)	26.7 (4.4)	27.6 (4.7)	26.7 (4.5)

TB: Tuberculosis, BMI: Body-mass index, SD: Standard deviation

Only 2 (0.5%) participants from Gothenburg reported having had TB, whereas 18 (1.3%) participants from Reykjavik reported TB. Most of the participants reporting TB had been treated before the year 1980. Also, most of the study participants had undergone TB treatment before 15 years of age, and 32 out of 35 were younger than 25 years old when they were first treated for TB **(Table 2).**

Table 2: Distribution of study participants with TB by study centre, year, and age when they were treated for the first time (N=35/ 4 530)

Number (%) of TB cases by study centre	
Bergen	6/ 806 (0.7)
Reykjavik	18/ 1 393 (1.3)
Gothenburg	2/ 438 (0.5)
Umeå	3/ 523 (0.6)
Uppsala	6/ 1 335 (0.4)
Year first treated for TB	
1950-1959	16
1960-1969	9
1970-1979	6
1980-1989	1
1990-1999	0
2000-2010	3
Age at the first treatment (years)	
0-5	14
6-10	8
11-15	6
16-20	2
21-25	2
26-39	0
Above 40	3

The distribution of respiratory and nasal outcomes among participants is shown in **Table 3.** Overall, participants who reported TB had a higher prevalence of all respiratory outcomes. Approximately 37.1% of participants who reported TB had wheezing compared to 18.6% among those without TB. Also, approximately 40% of participants with TB reported current asthma medication/ asthma attack and/or ≥ 3 asthma symptoms compared to 20% among those without TB.

Table 3: Respiratory outcomes according to TB status in the study population

	TB (N=35) n (%)	Without TB (N=4 495) n (%)
Wheezing[1]	13 (37.1)	835 (18.6)
Wheezing with shortness of breath[1]	9 (25.7)	531 (11.8)
Wheezing without cold[1]	11 (31.4)	631 (14.0)
Awoken with tightness in chest[1]	7 (20.0)	476 (10.6)
Awoken with shortness of breath[1]	1 (2.9)	166 (3.7)
Awoken with an attack of cough[1]	14 (41.7)	960 (21.3)
Shortness of breath when active[1]	25 (71.4)	2 577 (57.3)
Difficult breathing when walking on ground level[1]	14 (40.0)	911 (20.2)
Breathlessness[1]	4 (11.4)	343 (7.6)
Shortness of breath while walking[1]	4 (11.5)	130 (2.9)
Phlegm[1]	7 (20)	661 (14.9)
Nasal allergies	11 (31.4)	1 247 (27.7)
Asthma definition 1 (current asthma medication/ asthma attack)	8 (22.8)	592 (13.1)
Asthma definition 2 (≥ 3 asthma symptoms)	11 (31.4)	608 (13.5)
Asthma definition 3 (current asthma and/or ≥ 3 asthma symptoms)	14 (40.0)	901 (20.0)

[1]: In the last twelve months; TB: Tuberculosis

The association of TB infection with the risk of respiratory symptoms and asthmatic conditions is presented in **Table 4**. The risk of self-reported wheezing was 2.5 times higher among those who reported TB compared to those who did not report TB, and this association remained significant even after adjustment for confounders (OR: 2.36; 95% CI 1.14-4.88). TB was associated with a higher risk for asthma as per all three definitions of asthma (OR: 1.8; 95% CI 0.8-4.04, OR: 2.73; 95% CI 1.28-5.81, and OR: 2.47; 95% CI 1.22-4.99 as per definitions 1, 2, and 3, respectively).

Table 4: Association of respiratory symptoms and asthmatic condition with TB infection

	Crude		Adjusted*	
	OR (95% CI)	P value	OR (95%CI)	P value
Wheezing	2.59 (1.29-5.16)	0.007	2.36 (1.14-4.88)	0.020
Wheezing with shortness of breath	2.80 (1.36-5.76)	0.005	2.54 (1.20-5.39)	0.010
Wheezing without cold	2.58 (1.20-5.54)	0.015	2.31 (1.05-5.1)	0.030
Awoken with tightness in chest	2.11 (0.91-4.85)	0.070	1.92 (0.82-4.48)	0.130
Awoken with attack of cough	2.45 (1.25-4.84)	0.010	2.35 (1.17-4.71)	0.016
Difficult breathing when walking on ground level	2.6 (1.31-5.1)	0.006	2.26 (1.06-4.8)	0.03
Shortness of breath when active	1.86 (0.89-3.88)	0.098	1.71 (0.80-3.64)	0.164
Phlegm	1.42 (0.61-3.27)	0.400	1.22 (0.52-2.87)	0.640
Asthma definition 1 (current asthma medication/ asthma attack)	1.95 (0.89-4.3)	0.090	1.8 (0.8-4.04)	0.150
Asthma definition 2 (≥ 3 asthma symptoms)	2.93 (1.42-6.01)	0.003	2.73 (1.28-5.81)	0.009
Asthma definition 3 (current asthma and/or ≥ 3 asthma symptoms)	2.65 (1.34-5.25)	0.005	2.47 (1.22-4.99)	0.010

*Adjusted for age, sex, smoking, education, BMI
Abbreviations: CI: confidence interval; OR: odds ratio

Finally, the association of TB infection with allergic and non-allergic asthma is presented in **Table 5**. Our results indicate a positive association between TB and non-allergic asthma with OR: 3.27; 95% CI 1.38-7.73 as per definition 1, OR: 3.66; 95% CI 1.63-8.17 as per definition 2 and OR: 3.85; 95% CI 1.85-8.01 as per definition 3.

Table 5: Association of allergic and non-allergic asthma as related to TB infection

	Crude		Adjusted*	
	OR (95% CI)	P value	OR (95%CI)	P value
Allergic asthma definition 1 (hay fever + current asthma medication/ asthma attack)	0.40 (0.54-2.94)	0.360	0.38 (0.05-2.85)	0.353
Allergic asthma definition 2 (hay fever + ≥ 3 asthma symptoms)	0.9 (0.23-4.15)	0.990	0.89 (0.22-4.09)	0.964
Allergic asthma definition 3 (hay fever + current asthma and/or ≥ 3 asthma symptoms)	0.60 (0.14- 2.52)	0.454	0.58 (0.13- 2.45)	0.462
Non-allergic asthma definition 1 (current asthma medication/ asthma attack without hay fever)	3.69 (1.59-8.52)	0.002	3.27 (1.38-7.73)	0.007
Non-allergic asthma definition 2 (≥ 3 asthma symptoms without hay fever)	4.11 (1.91-8.84)	0.001	3.66 (1.63-8.17)	0.007
Non-allergic asthma definition 3 (current asthma and/or ≥ 3 asthma symptoms without hay fever)	4.25 (2.1-8.6)	0.001	3.85 (1.85-8.01)	0.000

*Adjusted for age, sex, smoking, education, BMI
Abbreviations: CI: confidence interval; OR: odds ratio

We performed stratified analyses according to sex and study centre, as presented in supplementary **Tables S3** and **S4**. While keeping in mind the limited number of persons with TB, we did not observe any convincing differences between men and women, or between Reykjavik and the other study centres.

DISCUSSION

This is one of the first studies addressing the association of TB with asthma and respiratory symptoms in a large Nordic-Baltic population-based study. Our study found that a history of TB infection, mostly before young adulthood, was associated with a higher risk for asthma and respiratory symptoms at age 50-75 years. This association was strong and consistent for non-allergic asthma, while our study indicated no association or a possible protective effect of TB on allergic asthma.

Several studies support our findings regarding more respiratory symptoms and/or asthma after TB. A study conducted in India found that out of 69 patients with TB and asthma, 69.6% developed asthma after TB infection (10). A multinational population-based cross-sectional study conducted in 19 sites, as well as a cross-sectional study conducted in India, found a positive association between TB and obstructive pulmonary diseases (11) (12). One study conducted in Brazil reported reduced pulmonary function after the treatment of pulmonary TB (13). In contrast, a study carried out to estimate the prevalence of asthma among TB patients concluded that TB infection was associated with a lower prevalence of asthma (14). Nonetheless, this analysis did not differentiate between allergic and non-allergic asthma.

Our study is one of the few available that analysed the association between TB infection and respiratory outcomes in a low TB incidence setting. Relatedly, a case-control study conducted in the United States found that the group with active TB had a higher risk of pulmonary impairment compared to the group with latent TB infection (15). Similarly, a cross-sectional study in Finland showed a positive association between previous TB infections and airway obstruction (16). Likewise, a recent cohort study conducted among immigrants in Canada reported a two-fold higher risk of airway diseases in patients with a history of pulmonary TB compared to controls (17).

The exact mechanisms that could cause obstructive pulmonary disease following the TB infection are still under investigation, and several hypotheses have been presented (18). Our study does not shed light on the mechanisms behind the associations between TB and respiratory outcomes. Most literature agrees that the host immune response and host-pathogen interaction may damage the lungs by distortion of the airway, reduced elasticity, damage to bronchial walls, and impairment to the lung parenchyma and vasculature which may cause chronic airflow obstruction (19). In agreement with this, we found that non-allergic asthma was substantially more common among participants with previous TB infections.

On the other hand, our study might indicate a protective or no effect of TB on allergic asthma. This is consistent with earlier findings on less allergies after childhood TB infection: A large case-control study conducted in Finland concluded that TB infection might prevent the future development of allergic asthma (20). Likewise, a cross-sectional study from South Africa reported that exposure to infectious bacteria such as Mtb might decrease the occurrence of allergic diseases (21). Less allergic asthma after TB infection might be explained on the ground of the hygiene hypothesis, which states that the decrease in infectious diseases in developed countries could be important for the increase in allergic and autoimmune diseases. The causes behind this assumption are not entirely understood (22), but it has been suggested that contact with pathogens in early childhood may help our immune system develop and reduce the likelihood of future allergic conditions (23).

Some researchers suggest that children receiving BCG vaccination may develop less allergic diseases later in life (24). In the Nordic countries, the vaccination policy of routine BCG vaccination was replaced by selective vaccination after 1975 (25). As all participants of the present study were born between 1945 and 1971, it is most likely that most study participants have received BCG vaccination. Unfortunately, we do not have data on BCG vaccination and cannot contribute to this topic.

The present study has several strengths. To the best of our knowledge, this is the first study to address the association of respiratory symptoms and asthmatic conditions with TB in participants from the Nordic-Baltic region in a single study. Importantly, the assessment of the participants was done by using a standardized questionnaire and included population-based study samples from different study centres, which reduced the risk of sampling bias.

One of the strengths of our study is that we investigated the association of TB with several definitions of asthma, reflecting different presentations. Definition 1 represents doctor-diagnosed asthma; this may be relatively specific, but there is a chance of doctor bias. Definition 2 is based on respiratory symptoms, which could be more sensitive than definition 1 and be less influenced by doctor bias.

According to Sunyer et al. the use of asthma symptoms score based on a combination of questions on asthma is more valid when analysing asthma in epidemiological studies (26). Definition 3 gives us asthma from patients' and doctors' views. It is more sensitive than either of the two because some patients are so well treated that they have very few symptoms. Consistency of the results across the different definitions strengthens the inference that the results are likely due to biological mechanisms rather than misclassification bias.

TB and respiratory outcomes have some common risk factors and these shared risk factors may influence the findings in the research (27). Possible confounders were selected using DAG, based on previous knowledge and published articles. We used parental education as a proxy for socioeconomic status. As most participants with TB had the infection before 15 years of age, we used parental education as a proxy rather than the education of the participants themselves.

Undeniably, there are certain limitations in this study. As the participants reporting TB were few, we observed relatively broad confidence intervals around the estimates (28). Moreover, because of the few participants, we had to limit our sub-analysis. The follow-up response rate after 20 years-follow up was about 53% in the RHINE study. Regarding loss to follow-up, Johannessen et al. in an analysis of the RHINE study found that disease prevalence was slightly affected by the loss to follow-up, whereas exposure-outcome associations were not affected (29). Further, it is not expected that selection bias would have influenced the associations as it is unlikely that the non-responders would have the opposite association than that observed in the responders. The use of self-reported information may lead to information bias. As TB, asthma, and allergies are chronic conditions, they are difficult to ignore, however, recall bias cannot be ruled out. In addition, social desirability bias may have occurred as it has been described that persons with a history of TB may answer negatively to questions about having had the disease (30). The social desirability bias was minimized by hiding the identity of participants in the questionnaire. It is likely that misclassification bias in our study

would be non-differential, as it is not likely that misclassification of TB would be systematic with regard to asthmatic conditions (31). Non-differential misclassification would imply that the actual association of TB with asthma and respiratory symptoms might be stronger than observed.

In the present study, we observed a relatively large number of the participants reporting TB were from Reykjavik. The incidence of TB is reported to be not very different between the Nordic countries in the 1990s (32), and most TB cases in our study underwent the disease before 1980. A stratified analysis of participants from Reykjavik and from the other study centres showed no indication of differences in the association of TB with asthma outcomes between Reykjavik and the other study centres, strengthening the interpretation of a biological rather than a socio-cultural explanation for the results.

The findings from the current study can be useful for global public health as our research highlights the need to follow up with TB patients after completion of treatment for timely detection and management of potential obstructive lung diseases. It is alarming that 40% of TB patients reported an asthma-related outcome at the age of 50-75 years. In addition, the study also suggests that unifying efforts to mitigate the burden of TB worldwide may help reduce the prevalence of asthma and respiratory symptoms.

CONCLUSION

We conclude from our analyses that TB has a positive association with asthma and respiratory symptoms. Further, our study suggests that TB might possibly increase the occurrence of non-allergic asthma, while allergic asthma was not higher among persons with a history of TB. Further register-based and mechanistic studies with extensive data would be important to strengthen the knowledge basis for targeted intervention efforts.

CONFLICT OF INTEREST

The research team declares no conflict of interest.

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Table S1: Respiratory symptoms and their definitions

Respiratory symptoms	Questions from questionnaire
Wheezing	Have you had wheezing or whistling in your chest at any time in the last 12 months?
Wheezing with shortness of breath	Have you been at all breathless when the wheezing noise was present?
Wheezing without cold	Have you had this wheezing or whistling when you did not have a cold?
Awoken with tightness in chest	Have you woken up with a feeling of tightness in your chest at any time in the last 12 months?
Awoken with shortness of breath	Have you been woken by an attack of shortness of breath at any time in the last 12 months?
Awoken with an attack of cough	Have you been woken by an attack of coughing at any time in the last 12 months?
Shortness of breath when active	Do you get breathless with strenuous exercise?
Difficult breathing when walking on ground level	Do you get short of breath when hurrying on the level or walking up a slight hill?
Breathlessness	Do you walk slower than people of the same age on the level because of breathlessness, or do you have to stop for breath when walking on your own pace on the level?
Shortness of breath while walking	Do you stop for breath after walking about 100 metres or after a few minutes on the level?
Phlegm	Do you usually bring up phlegm or do you have phlegm in your lungs which you have difficulty bringing up?
Nasal allergies	Do you have any nasal allergies including hay fever?
Attack of asthma	Have you had an attack of asthma in the last 12 months?
Currently taking asthma medication	Are you currently taking any medicine (including inhalers, aerosols, or tablets) for asthma?

Table S2: Questions used to create a new variable, "asthma symptoms score."

Asthma-like symptoms	Definition
Wheezing	Have you had wheezing and whistling in your chest at any time in the last 12 months?
Awoken with Tightness in the chest	Have you woken up with a feeling of tightness in your chest at any time in the last 12 months?
Awoken with Shortness of breath	Have you been woken by an attack of shortness of breath at any time in the last 12 months?
Awoken with an Attack of cough	Have you been woken by an attack of coughing at any time in the last 12 months?
Breathlessness	Do you walk slower than people of the same age on the level because of breathlessness, or do you have to stop for breath when walking on your own pace on the level?

Table S3: Association of tuberculosis infection with the odds of respiratory symptoms and asthmatic condition after stratification by sex

	Male		Female	
	OR (95% CI)	P value	OR (95% CI)	P value
Asthma definition 1 (current asthma medication/ asthma attack)	3.28 (1.14-9.39)	0.02	1.13 (0.32-3.93)	0.84
Asthma definition 2 (≥ 3 asthma symptoms)	4.51(1.65-12.33)	0.003	1.99 (0.70-5.61)	0.19
Asthma definition 3 (current asthma and/or ≥ 3 asthma symptoms)	3.31 (1.25-8.77)	0.016	2.18 (0.84-5.65)	0.109
Allergic asthma definition 2 (hay fever + ≥ 3 asthma symptoms)	1.42 (0 .18- 10.8)	0.73	0.75(0.09-5.67)	0.78
Allergic asthma definition 3 (hay fever + current asthma and/or ≥ 3 asthma symptoms)	0.75 (0.10-5.76)	0.79	0.49 (0.06-3.74)	0.49
Non-allergic asthma definition 1 (current asthma medication/ asthma attack without hay fever)	5.08 (1.63-15.83)	0.005	2.67 (0.76- 9.33)	0.12
Non-allergic asthma definition 2 (≥ 3 asthma symptoms without hay fever)	5.93 (2.06- 17.07)	0.001	2.91 (0.95- 8.93)	0.61
Non-allergic asthma definition 3 (current asthma and/or ≥ 3 asthma symptoms without hay fever)	5.01 (1.83-13.67)	0.002	3.67 (1.36-9.85)	0.01

Table S4: Association of tuberculosis infection with the asthmatic condition after stratification based on study centre

	Reykjavik		Other	
	OR (95% CI)	P value	OR (95% CI)	P value
Asthma definition 2 (≥ 3 asthma symptoms)	2.94 (1.09- 7.94)	0.033	2.76(0.97-7.89)	0.05
Non-allergic asthma definition 2 (≥ 3 asthma symptoms without hay fever)	4.65 (1.72 -12.61)	0.002	2.89(0.82-10.14)	0.09

*All sub-analyses were not possible due to low sample size.

Appendix I

RHINE IV Questionnaire

RESPIRATORY HEALTH IN NORTHERN EUROPE

1. What is your birth date? ..... /..... /.....
   day month year

2. What is today's date? ..... /..... /.....
   day month year

3. Are you male or female? ☐ Male ☐ Female

RESPIRATORY SYMPTOMS

4. Have you had wheezing or whistling in your chest at any time in the last 12 months? ☐ No ☐ Yes

   If NO go to question 5, if YES::

   4.1 Have you been at all breathless when the wheezing noise was present? ☐ No ☐ Yes

   4.2 Have you had this wheezing or whistling when you did not have a cold? ☐ No ☐ Yes

5. Have you woken up with a feeling of tightness in your chest at any time in the last 12 months? ☐ No ☐ Yes

6. Have you been woken by an attack of shortness of breath at any time in the last 12 months? ☐ No ☐ Yes

7. Have you been woken by an attack of coughing at any time in the last 12 months? ☐ No ☐ Yes

8. Have you had an attack of asthma in the last 12 months? ☐ No ☐ Yes

9. Are you currently taking any medicine (including inhalers, aerosols or tablets) for asthma? ☐ No ☐ Yes

10. Do you have any nasal allergies including hay fever? ☐ No ☐ Yes

11. Do you get breathless with strenuous exercise? ☐ No ☐ Yes

12. Do you get short of breath when hurrying on the level or walking up a slight hill? ☐ No ☐ Yes

13. Do you walk slower than people of the same age on the level because of breathlessness, or do you have to stop for breath when walking on your own pace on the level? ☐ No ☐ Yes

14. Do you stop for breath after walking about 100 metres or after a few minutes on the level? ☐ No ☐ Yes

15. Are you too breathless to leave the house or are you breathless when dressing or undressing? ☐ No ☐ Yes

16. How tall are you? ..... cm

17. How much do you weigh? ..... kg

18. What is your waist circumference? ..... cm

19. In recent years, have you been troubled by a protracted cough? ☐ No ☐ Yes

20. Do you usually bring up phlegm or do you have phlegm in your lungs which you have difficulty bringing up? ☐ No ☐ Yes

If NO go to question 21, if YES:

20.1 Do you bring up phlegm in this way almost every day for at least three months every year? ☐ No ☐ Yes

20.2 Have you had periods of this kind for at least two years in a row? ☐ No ☐ Yes

20.3 How old were you when these problems began? ..... years

21. Does your voice tire, strain or get hoarse when you talk? Disregard symptoms that depend on current cold or upper-airway infection. The voice symptoms may vary but try to estimate an average.

☐ No ☐ Yes, to a small extent ☐ Yes, to a great extent

SMOKING HABITS

22. Are you a smoker (this applies even if you only smoke the odd cigarette/cigar or pipe every week)? ☐ No ☐ Yes

23. Are you an ex-smoker? ☐ No ☐ Yes

24. Have you ever used any other nicotine containing products on a regular basis? ☐ No ☐ Yes

If NO to questions 22, 23 and 24 go to question 25, if YES to any of the above:

24.1 How much do or did you smoke, or how many portions of snus do/did you use? Please provide an average.

..... cigarettes/day ..... e-cigarettes/day ..... cigars/week

..... pkts pipe tobacco/week ..... portions snus/day

..... waterpipe sessions/week

24.2 Have you ever used quitting aids such as nicotine replacement products? ☐ No ☐ Yes

24.3 For how many years have you been smoking? (applies to both smokers and ex-smokers) ..... years

24.4 How old were you when you started smoking? ..... years

24.5 If you are an ex-smoker, when did you stop smoking? ..... (year)

24.6 Used snus for ..... years (applies both if you currently use snus or if you previously used snus)

24.7 How old were you when you started using snus? ..... years

24.8 When did you stop using snus? ..... (year)

UPPER AND LOWER AIRWAYS

25. Do you have or have you ever had asthma? ☐ No ☐ Yes

If NO go to question 26, if YES:

25.1 Have you ever had asthma diagnosed by a doctor? ☐ No ☐ Yes

25.2 How old were you when you first experienced asthma symptoms? ..... years

25.3 In which year did you last experience asthma symptoms? ..... (year)

26. Has a doctor ever told that you have COPD? ☐ No ☐ Yes

27. Have you ever experienced nasal symptoms such as nasal congestion, rhinorrvoea (runny nose) and/or sneezing attacks without having a cold? ☐ No ☐ Yes

If NO go to question 28, if YES:

27.1 How old were you when you experienced them for the first time? ..... years

27.2 Have you had these kinds of nasal symptoms in the last 12 months? ☐ No ☐ Yes

27.3 At which time of the year are your nasal symptoms worst?
☐ Spring ☐ Summer ☐ Autumn ☐ Winter ☐ Always ☐ Don´t know

28. Has your nose been blocked for more than 12 consecutive weeks during the last 12 months? ☐ No ☐ Yes

29. Have you had pain or pressure around the forehead, nose or eyes for more than 12 consecutive weeks during the last 12 months? ☐ No ☐ Yes

30. Have you had discoloured nasal discharge (snot) or discoloured mucus in the throat for more than 12 weeks during the last 12 consecutive months? ☐ No ☐ Yes

31. Has your sense of smell been reduced or absent for more than 12 consecutive weeks during the last 12 months? ☐ No ☐ Yes

32. Are you currently using nasal steroid spray? ☐ No ☐ Yes

INDOOR AND OUTDOOR ENVIRONMENT

33. When was your present home built or properly renovated? ..... (year)

34. In which type of accommodation do you live?
☐ Detached house ☐ Semidetached or terraced house
☐ Apartment ☐ Other

34.1 If you live in an apartment, which floor do you live on?
☐ Ground floor ☐ 1st floor ☐ 2nd floor
☐ 3rd floor ☐ 4th floor or higher

35. When did you move to your current home? ..... (year)

36. How many days per year do you normally stay at another address? ..... days

37. Does tobacco smoking take place in your present home?
☐ Yes, every day ☐ Yes, frequently *(1–4 times/week)*
☐ Yes, sometimes *(1–3 times/month)* ☐ No, never

38. Have any of the following been identified in your home during the past 12 months:

38.1 Water leakage or water damage indoors in walls, floor or ceilings ☐ No ☐ Yes

38.2 Bubbles or yellow discoloration on plastic floor covering, or black discoloration of parquet floor? ☐ No ☐ Yes

38.3 Visible mould growth indoors on walls, floor or ceilings? ☐ No ☐ Yes

39. Have you seen any signs of damp, water leakage or mould in your home at any time during the past 10 years? ☐ No ☐ Yes

40. During the Covid-19 pandemic, how much time do you spend in a car each day? Approx ..... minutes/day

41. How much time would you normally have spent in a car each day in that period? Approx ..... minutes/day

42. During the Covid-19 pandemic, how much time do you spend in green areas (e.g. parks, forests, gardens) each day? Approx ..... minutes/day

43. How much time would you normally have spent in green areas (e.g. parks, forests, gardens) each day in that period? Approx ..... minutes/day

44. If you work, how do you usually travel to and from work during each season?
(please tick only ONE main transport alternative pr season)

	Spring	Summer	Autumn	Winter	N/A
Car					
Bus/tram					
Train					
Walk					
Bicycle					

45. How many km do you normally travel from home to work (single travel)? ..... km

DEMOGRAPHY

46. What is your marital status?

☐ Single ☐ Currently married ☐ Cohabitating ☐ Separated or divorced
☐ Widowed ☐ Do not wish to answer

47.1 What term best describes the place where you live now?

☐ Farm ☐ Village in rural area ☐ Small town ☐ Suburb of city
☐ Inner city

47.2 What term best describes the place where you lived in 2010?

☐ Farm ☐ Village in rural area ☐ Small town ☐ Suburb of city
☐ Inner city

OCCUPATION AND WORK

48. What term best describes your current work situation?

☐ Employed ☐ Self-employed ☐ Unemployed, looking for work
☐ Not working because of poor health ☐ Full-time homemaker ☐ Retired ☐ Other

49. If currently working, what is your work address?

.....

.....

50. If you are retired: at what age did you retire? ..... years

51. Has the Covid-19 pandemic affected your work situation? ☐ No ☐ Yes

51.1. If yes, how has it affected your work situation?
(select all answers that apply to you)

- ☐ loss of job
- ☐ reduced working hours
- ☐ increased working hours
- ☐ increased job insecurity
- ☐ loss of partner's job
- ☐ reduced working hours for partner
- ☐ other, please specify: .....

.....

52. Please list all jobs that you have ever had for six months or more since year 2000. These jobs may be outside the house or at home, **excluding homemaking or housework**, full time or part time, paid or unpaid, including self-employment, for example in a family business. Please include part time jobs only if you had been doing them for 20 or more hours per week. Please start with your current or last held job.

Job	Occupation - Job Title: Please provide a short description of the job	Industry / Branch: What does (did) your firm or employer make or what services does (did) it provide?	Start month	Start year	End month	End year (If current job please enter CURRENT)
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						
16						
17						
18						
19						
20						

53. Have you seen any signs of damp, water leakage or mould in your workplace at any time during the past 10 years? ☐ No ☐ Yes

54. Have you ever worked shift work? ☐ No ☐ Yes

54.1 If "Yes", between which years? Start year: ..... End year: .....

55. Have you ever worked night shift? ☐ No ☐ Yes

55.1 If "Yes", between which years? Start year: ..... End year: .....

56. Have you ever worked as a hairdresser? ☐ No ☐ Yes

56.1 If yes, when did you start? If yes, when did you start? ..... (year)

56.2 If yes, for how many years? ..... years

57. Have you ever changed job because the job affected your breathing? ☐ No ☐ Yes

57.1 If "Yes", in which years? ..... (year)

57.2 If "Yes", from which occupation/job did you change? *(could be several)*

.....

.....

58. Have you ever changed job because of other health problems/diseases? ☐ No ☐ Yes

58.1 If Yes, in which years? ..... (year)

58.2 If "Yes", which occupation/job did you change from? *(could be several)*

.....

.....

59. How many days have you been on sick leave during the last year?
☐ 0 days ☐ 1–7 days ☐ 8–30 days ☐ 31–90 days
☐ More than three months ☐ N/A

FAMILY

60. If you have biological children, please state the years of birth and gender for each of them in the table below

	Birth year	Gender
Child #1		☐ Male ☐ Female
Child #2		☐ Male ☐ Female
Child #3		☐ Male ☐ Female
Child #4		☐ Male ☐ Female
Child #5		☐ Male ☐ Female
Child #6		☐ Male ☐ Female
Child #7		☐ Male ☐ Female

61. We would like to ask about your parents and grandparents, whether they were ever treated for tuberculosis and when they were born. If you do not know the year of birth, please suggest crudely *(nearest 10 years)*:

	Ever treated for tuberculosis	Year of birth
Mother	☐ No ☐ Yes ☐ Don't know	
Father	☐ No ☐ Yes ☐ Don't know	
Maternal grandmother	☐ No ☐ Yes ☐ Don't know	
Mathernal grandfather	☐ No ☐ Yes ☐ Don't know	
Paternal grandmother	☐ No ☐ Yes ☐ Don't know	
Paternal grandfather	☐ No ☐ Yes ☐ Don't know	

SLEEP-RELATED SYMPTOMS AND DISORDERS

62. How likely are you to doze off or fall asleep in the following situations in contrast to feeling just tired? This refers to your usual way of life in recent times. Even if you have not done some of these things recently try to work out how they would have affected you.

Use the following scale to choose the most appropriate number for each situation:

0 = would **never** doze, 1 = **slight** chance of dozing,
2 = **moderate** chance of dozing, 3 = **high** chance of dozing.

62.1	Sitting and reading	☐ 0	☐ 1	☐ 2	☐ 3
62.2	Watching TV	☐ 0	☐ 1	☐ 2	☐ 3
62.3	Sitting inactive in a public place *(e.g. a theatre or a meeting)*	☐ 0	☐ 1	☐ 2	☐ 3
62.4	As a passenger in a car for an hour without a break	☐ 0	☐ 1	☐ 2	☐ 3
62.5	Lying down to rest in the afternoon when circumstances permit	☐ 0	☐ 1	☐ 2	☐ 3
62.6	Sitting and talking to someone	☐ 0	☐ 1	☐ 2	☐ 3
62.7	Sitting quietly after a lunch without alcohol	☐ 0	☐ 1	☐ 2	☐ 3
62.8	In a car, while stopped for a few minutes in the traffic	☐ 0	☐ 1	☐ 2	☐ 3

The numbers mean
1: Never or almost never
2: Less than once a week
3: once or twice a week
4: 3–5 nights/days a week
5: Almost every day or night

63. How often has it occurred in the last months that:

63.1 ...You snore loudly and disturbingly? ☐1 ☐2 ☐3 ☐4 ☐5

63.2 ...You have heartburn or belching when you have gone to bed? ☐1 ☐2 ☐3 ☐4 ☐5

63.3 ...You have difficulty in getting to sleep at night? ☐1 ☐2 ☐3 ☐4 ☐5

63.4 ...You wake up repeatedly during the night? ☐1 ☐2 ☐3 ☐4 ☐5

63.5 ...You perspire heavily during the night? ☐1 ☐2 ☐3 ☐4 ☐5

63.6 ...You feel sleepy during the day? ☐1 ☐2 ☐3 ☐4 ☐5

63.7 ...Your sleep is unrefreshing? ☐1 ☐2 ☐3 ☐4 ☐5

63.8 ...You wake up too early and have difficulty in getting to sleep again? ☐1 ☐2 ☐3 ☐4 ☐5

63.9 ...You use sleep medicine? ☐1 ☐2 ☐3 ☐4 ☐5

64. Have you ever been told that you snore when you sleep? ☐ No ☐ Yes

If "No" go to question 65, if "Yes":

During the last month have you had or been told about the following symptoms *(please select only one answer per question)*:

		Never	Seldom	Sometimes	Frequently	Always
64.1	Loud snoring?	☐	☐	☐	☐	☐
64.2	Snorting or gasping?	☐	☐	☐	☐	☐
64.3	Your breathing stops, choke or struggle for breath?	☐	☐	☐	☐	☐

65. Have you ever had sleep apnoea diagnosed by a doctor? ☐ No ☐ Yes

If "No" go to question 66 if "Yes":

65.1 What year did you get the diagnosis of sleep apnoea? ..... (year)

65.2 If you are currently treated for sleep apnoea, what treatment do you have?

- ☐ CPAP
- ☐ Oral appliance (bite splint)
- ☐ Previous surgery in the throat or nose
- ☐ Others

66. How much do you usually sleep per night?

66.1 On weekdays ..... hours and ..... minutes

66.2 On weekends ..... hours and ..... minutes

66.3 At what time do you usually fall asleep? At ..... o´clock

66.4 At what time do you usually wake up? At ..... o´clock

I have irregular sleep schedule ☐

OTHER DISEASES

67. Have you ever had hypertension (high blood pressure) diagnosed by a doctor? ☐ No ☐ Yes

If NO go to question 68, if YES:

67.1 When did you get the diagnosis hypertension *(high blood pressure)*? ..... (year)

67.2 Are you currently taking any medication for hypertension (high blood pressure)? ☐ No ☐ Yes

68. Have you ever had stroke? ☐ No ☐ Yes

If NO go to question 69 if YES:

68.1 If you have had stroke, in which year was it? ..... (year)

69. Have you ever been treated in hospital because of heart infarction or angina pectoris? ☐ No ☐ Yes

If NO go to question 70 if YES:

69.1 When were you treated (for the first time) at a hospital because of heart infarction or angina pectoris? ..... (year)

70. Have you ever had atrial fibrillation diagnosed by a doctor? ☐ No ☐ Yes

71. Have you ever had leg oedema? ☐ No ☐ Yes

72. Have you ever had diabetes diagnosed by a doctor? ☐ No ☐ Yes

If NO go to question 73 if YES:

72.1 What year did you get the diagnosis diabetes? ..... (year)

72.2 What treatment are you currently using for diabetes?
☐ Insulin ☐ Tablets ☐ Both insulin and tablets ☐ Only diet

73. Have you ever had tuberculosis? ☐ No ☐ Yes

73.1 If yes: When were you treated (for the first time) for tuberculosis? ..... (year)

74. Do you have or have you ever had inflammatory bowel disease (ulcerative colitis or Crohn's disease)? ☐ No ☐ Yes

74.1 If yes: how old were you when the disease started? ..... years

75. Have you ever been treated for depression? ☐ No ☐ Yes

75.1 If yes: Do you currently receive treatment for depression? ☐ No ☐ Yes

76. Have you ever been treated for anxiety? ☐ No ☐ Yes

76.1 If yes: Do you currently receive treatment for anxiety? ☐ No ☐ Yes

77. Have you ever had eczema or any kind of skin allergy diagnosed by a doctor? ☐ No ☐ Yes

If NO go to question 78, if YES:

77.1 Have you had eczema or any kind of skin allergy diagnosed by a doctor during the last 12 months? ☐ No ☐ Yes

77.2 How old were you when you first had eczema or skin allergy? _____ years

77.3 Did/does your eczema or skin allergy affect your hands? ☐ No ☐ Yes

77.4 Have you noticed that contact with certain materials, chemicals or anything else in your work makes your eczema worse? ☐ No ☐ Yes ☐ Don´t know

78. Have you ever had an itchy rash that was coming and going for at least 6 months? ☐ No ☐ Yes

If NO go to question 79, if YES:

78.1 Have you had this itchy rash in the last 12 months? ☐ No ☐ Yes

78.2 Has this itchy rash at any time affected any of the following places: the folds of the elbows, behind the knees, in front of the ankles, under the buttocks or around the neck, ears or eyes? ☐ No ☐ Yes

78.3 Has this itchy rash affected your hands at any time in the last 12 months? ☐ No ☐ Yes

79. Have you ever had an intestinal worm infection? ☐ No ☐ Yes

80. How many operations have you had the past 10 years? .....

81. In how many of them were you anesthetized? .....

82. What kind of surgery have you had during the past 10 years? (if you have had more than one surgery in any of the categories, list the year for the first surgery of that category)

Abdominal surgery	☐ No	☐ Yes	..... (year)
Gynecological surgery	☐ No	☐ Yes	..... (year)
Breast surgery	☐ No	☐ Yes	..... (year)
Urological surgery	☐ No	☐ Yes	..... (year)
Open heart or lung surgery	☐ No	☐ Yes	..... (year)
Orthopedic surgery	☐ No	☐ Yes	..... (year)
Ear, nose or throat surgery	☐ No	☐ Yes	..... (year)
Other surgery	☐ No	☐ Yes	..... (year)

83. Have you had cataract surgery? ☐ No ☐ Yes

If NO go to question 84, if YES:

83.1 When did you have your first cataract surgery? ..... (year)

84. Have you received a hip or knee prothesis? ☐ No ☐ Yes

85. Have you received radiation therapy to the breast or chest? ☐ No ☐ Yes

86. Have you taken any antibiotics during the last 12 months? ☐ No ☐ Yes

If NO go to question 87, if YES:

86.1 Have you taken any antibiotics for respiratory infections during the last 12 months? ☐ No ☐ Yes

GENERAL HEALTH

87. In general, how would you rate your overall health?
☐ Excellent ☐ Very good ☐ Good ☐ Fair ☐ Poor

88. How frequently do you exercise? (Give an average)
☐ Never ☐ Less than once a week ☐ once a week
☐ 2–3 times a week ☐ Almost every day

If you do such exercise as frequently as once or more times a week:

88.1 How hard do you push yourself? (Give an average)
☐ I take it easy without breaking into a sweat or losing my breath
☐ I push myself so hard that I lose my breath and break into a sweat
☐ I push myself to near-exhaustion

88.2 How long does each session last? (Give an average)
☐ Less than 15 minutes ☐ 16–30 minutes
☐ 30 minutes to 1 hour ☐ More than 1 hour

89. How often do you engage in the following activities:

	Never	< Once a week	Once a week	2–3 times a week	Nearly every day
Exercise in a training centre/gym?					
Exercise indoors at home?					
Walk/run/cycle in the woods/mountains/ nature?					
Walk/run/cycle along roads with little to medium traffic?					
Walk/run/cycle along heavily trafficated roads?					

90. On a typical weekday, what is your average daily step count? ..... steps ☐ Don´t know

91. How tall were you when you were 20 years old? ..... cm

92. How much did you weigh when you were 20 years old? ..... kg

93. How do you assess your own dental health?
☐ Exellent ☐ Very good ☐ Good ☐ Fair ☐ Poor

94. How often do you receive dental treatment?
☐ Twice or more per year
☐ Once a year
☐ Less than once a year
☐ Less than every second year

95. Does your gum bleed when you brush your teeth?
☐ Always ☐ Often ☐ Sometimes ☐ Rarely ☐ Never

96. How often do you usually brush your teeth?
☐ 2 times/day or more
☐ Once a daily
☐ Less than daily

97. Do you regularly perform interdental cleaning with dental floss or interdental brushes?
☐ 2 times/day or more
☐ Once daily
☐ Less than daily
☐ Never

98. Has your dentist ever told you that you have gum disease (periodontal disease)? ☐ No ☐ Yes

99. Have you ever had treatment for gum disease? ☐ No ☐ Yes ☐ Don´t know

We now wish to ask two questions about cleaning of clothes, since this gives information about how much your skin is exposed to detergents.

100. How many times do you usually use trousers before washing them?
☐ Once ☐ 2-3 times ☐ 4-6 times ☐ 7 times or more

101. How many times do you usually use shirts/blouses/T-shirts or other top near the body before washing them?
☐ Once ☐ 2-3 times ☐ 4-6 times ☐ 7 times or more

COVID-19 (CORONAVIRUS) AND YOUR HEALTH

102. Do you think you have had Covid-19? ☐ No ☐ Yes ☐ Don´t know

IF NO, you have completed this questionnaire, If YES, please answer the following questions

102.1 On what date do you think your Covid-19 infection began?
(make your best guess if you fo not know precisely)

_____ day _____ month _____ (year)

102.2 What makes you think you have had Covid-19?
(select all answers that apply to you)

☐ I was admitted to hospital and a doctor told me I had Covid-19 infection

☐ I had a test that showed I was suffering from Covid-19 – but did not get admitted to hospital

☐ I had a test that showed I had developed antibodies to Covid-19

☐ One of the people living in my household had a positive Covid-19 test

☐ One of my close contacts who does not live in my household had a positive Covid-19 test

☐ I spoke with a doctor (or nurse) and they told me it was likely to be Covid-19 – but I did not have a test

☐ In my opinion my symptoms were typical of Covid-19

☐ Other, please specify: .....

.....

INFORMATION AND CONTACT CONSENT

In case we need to get in touch with you again please write your contact information below.

E-mail address .....

Mobile number .....

THANK YOU FOR YOUR HELP!

Appendix II

RHINE III Questionnaire

Kompetansesenter for klinisk forskning
HAUKELAND SYKEHUS

Institutt for indremedisin
Seksjon for lungemedisin

Ernst Omenaas

Airways symptoms

1. Have you had wheezing or whistling in your chest at any time **in the last 12 months?** ☐ No ☐ Yes

   If NO go to question 2, if YES:

   1.1 Have you been at all breathless when the wheezing noise was present? ☐ No ☐ Yes

   1.2 Have you had this wheezing or whistling when you did not have a cold? ☐ No ☐ Yes

2. Have you woken up with a feeling of tightness in your chest at any time **in the last 12 months?** ☐ No ☐ Yes

3. Have you been woken by an attack of shortness of breath at any time **in the last 12 months?** ☐ No ☐ Yes

4. Have you been woken by an attack of coughing at any time **in the last 12 months?** ☐ No ☐ Yes

5. Have you had an attack of asthma **in the last 12 months?** ☐ No ☐ Yes

6. Are you currently taking any medicine *(including inhalers, aerosols or tablets)* for asthma? ☐ No ☐ Yes

7. Do you have any nasal allergies including hay fever? ☐ No ☐ Yes

8. What is your date of birth? *(dd/mm/yy)* ...../...../.....

9. What is today's date? *(dd/mm/yy)* ...../...../.....

10. Are you male or female? ☐ Male ☐ Female

11. How tall are you? ..... cm

12. How much do you weigh? ..... kg

    12.1 What is your waist circumference? ..... cm
    (Please use the provided tape measure, and measure your waist at the level of the navel, while standing and under your clothes.)

13. In recent years, have you been troubled by a protracted cough? ☐ No ☐ Yes

14. Do you usually bring up phlegm or do you have phlegm in your lungs which you have difficulty bringing up? ☐ No ☐ Yes

    If NO to question 13 and 14 go to question 15, if YES:

    14.1 Do you cough or bring up phlegm in this way almost every day for **at least three months every year?** ☐ No ☐ Yes

    14.2 Have you had periods of this kind for at least two years in a row? ☐ No ☐ Yes

    14.3 How old were you when these problems began? ..... years

2

15. Do you have or have you ever had asthma? ☐ No ☐ Yes

If NO go to question 16, if YES:

15.1 Have you ever had asthma diagnosed by a doctor? ☐ No ☐ Yes

15.2 How old were you when you first experienced asthma symptoms? ..... years

15.3 In which year did you last experience asthma symptoms? Year .....

16. Has a doctor ever told you that you have chronic obstructive pulmonary disease (COPD)? ☐ No ☐ Yes

17. Have you ever had wheezing or whistling in your chest? ☐ No ☐ Yes

17.1 If YES, how old were you when you first noticed wheezing or whistling in your chest? ..... years

17.2. If YES, when was the last year you noticed wheezing or whistling in your chest? Year .....

18. Have you ever experienced nasal symptoms such as nasal congestion, rhinorrhoea (runny nose) and/or sneezing attacks without having a cold? ☐ No ☐ Yes

If NO go to question 19, if YES:

18.1 How old were you when you experienced such nasal symptoms for the first time? ..... years

18.2 Have you had such nasal symptoms in the last 12 months? ☐ No ☐ Yes

18.3 At which time of the year are your nasal symptoms worst? *(tick one box only)*

Spring	Summer	Autumn	Winter	Always	Don't know
☐	☐	☐	☐	☐	☐

19. Has your nose been blocked **for more than 12 weeks during the last 12 months?** ☐ No ☐ Yes

20. Have you had pain or pressure around the forehead, nose or eyes **for more than 12 weeks during the last 12 months?** ☐ No ☐ Yes

21. Have you had discoloured nasal discharge (snot) or discoloured mucus in the throat **for more than 12 weeks during the last 12 months?** ☐ No ☐ Yes

22. Has your sense of smell been reduced or absent **for more than 12 weeks during the last 12 months?** ☐ No ☐ Yes

Smoking habits

23. Do you smoke? *(this applies even if you only smoke the odd cigarette/cigar or pipe every week)* ☐ No ☐ Yes

24. Did you smoke previously? ☐ No ☐ Yes

If NO to question 23 and 24 go to question 25, if YES:

24.1 How much do you smoke / did you smoke? *(give an average)*

..... cigarettes/day

..... cigars/week

..... pkts pipe tobacco/week

24.2 How old were you when you started smoking? ..... years

24.3 For how long have you smoked? *(applies to both smokers and ex-smokers)* .....years

24.4 If you are an ex-smoker, when did you stop smoking? Year .....

Marital status

25. What is your current marital status? *(tick one box only)*

Single	Married	Cohabitating	Separated or divorced	Widowed	Do not wish to answer
☐	☐	☐	☐	☐	☐

Education

26. Please mark the educational level which best describes your level. *(tick one box only)*

Primary school	Lower or upper secondary school, or technical school	College or university
☐	☐	☐

4

Occupation and work

27. Are you currently working? ☐ No ☐ Yes

28. Which is your current or most recent work or occupation? *(please use capital letters)*

……

28.1 How many years have you worked or did you work in this occupation? ……years

29. We assume that your work ability, when it was as best, was 100 percent. How would you rate your current work ability, expressed in percent? ..... %

30. Have you ever changed job because the job affected your breathing? ☐ No ☐ Yes

31. Have you ever changed job because of hayfever or nasal symptom ☐ No ☐ Yes

32. Have you ever changed job because of other health problems/diseases? ☐ No ☐ Yes

33. Have you ever worked as a painter? ☐ No ☐ Yes

If YES, for how many years? ..... years

34. Have you ever worked as a cleaner? ☐ No ☐ Yes

If YES, for how many years? ..... years

35. Have you been reporting any days of sick leave in **the last 12 months?** ☐ No ☐ Yes

35.1 *If YES,* how many days have you been on sick leave? *(tick one box only)*

1–7 days	8-30 days	31 days–90 days	More than three months
☐	☐	☐	☐

36. Have you been reporting any days of sick leave because of breathing problems in **the last 12 months?** ☐ No ☐ Yes

36.1 *If YES,* how many days have you been on sick leave for breathing problems? *(tick one box only)*

1–7 days	8–30 days	31 days–90 days	More than three months
☐	☐	☐	☐

In-door and out-door environment

37. In which type of accommodation do you live? *(tick one box only)*

Detached house	Semidetached or terraced house	Apartment	Other
☐	☐	☐	☐

38. When did you move to your current home? Year .....

39. How many hours per day do you spend in your home most days? Approx. ..... hours/day

40. Does tobacco smoking take place in your present home? *(tick one box only)*

Yes, every day	Yes, frequently 1-4 times/week	Yes, sometimes 1-3 times/month	No, never
☐	☐	☐	☐

41. Have any of the following been identified in your home in **the last 12 months:**

41.1 Water leakage or water damage indoors in walls, floor or ceilings ☐ No ☐ Yes

41.2 Bubbles or yellow discoloration on plastic floor covering, or black discoloration of parquet floor ☐ No ☐ Yes

41.3 Visible mould growth indoors on walls, floor or ceilings. ☐ No ☐ Yes

42. Have you seen any signs of damp, water leakage or mould in your home at any time in **the past 10 years?** ☐ No ☐ Yes

43. Have you seen any signs of damp, water leakage or mould in your workplace at any time in **the past 10 years?** ☐ No ☐ Yes

44. Is your bedroom window towards a nearby street (less than 20 m)? *(tick one box only)*

No	Yes a street with little traffic	Yes a street with moderate traffic	Yes a street with much traffic
☐	☐	☐	☐

45. Can you in your bedroom hear traffic noise? *(tick one box only)*

Not at all	A little	Much	Very much
☐	☐	☐	☐

46. How much time do you usually spend walking or travelling along streets with busy traffic a typical weekday? Approx ..... minutes/day

Childhood and family

47. What term best describes the place you lived most of the time when you were under the age of five years? *(tick one box only)*

- ☐ Farm with livestock
- ☐ Farm without livestock
- ☐ Village in rural area
- ☐ Small town
- ☐ Suburb of city
- ☐ Inner city

48. When you were a child, which of the following were mainly used for heating? *(more than one box may apply)*

Open wood	Stove with coke, coal or wood	Paraffin	Electricity	Gas or oil fired boiler
☐	☐	☐	☐	☐

49. Did you have a serious respiratory infection before the age of five years? ☐ No ☐ Yes ☐ Don't know

50.1 Did your father ever smoke regularly during your childhood? ☐ No ☐ Yes ☐ Don't know

50.2 Did your mother ever smoke regularly during your childhood? ☐ No ☐ Yes ☐ Don't know

50.3 Did other people (other than parents) smoke regularly at home during your childhood? ☐ No ☐ Yes ☐ Don't know

51. When you were a child, how often did you eat fresh fruits and berries? *(more than one box may apply)*

Never	Rarely	Every week	Almost daily	Almost daily in the autumn season
☐	☐	☐	☐	☐

52. Did your biological parents ever suffer from any of the following: *(more than one box may apply)*

	Mother *(tick box if Yes)*	Father *(tick box if Yes)*
Asthma	☐	☐
Chronich bronchitis, emphysema and/or COPD	☐	☐
Heart disease	☐	☐
Hypertension	☐	☐
Stroke	☐	☐
Diabetes	☐	☐
Cancer	☐	☐

53. Do you have children (including grown-up children)? ☐ No ☐ Yes

If N0 go to question 54, *If YES:*

53.1 how many children do you have? ..... children

53.2 Please write the years when your children were born, and tick *"YES"* if they have had any of the following:

	Asthma before 10 year Yes	Asthma after 10 years Yes	Hayfever/ rhinitis Yes	Atopic eczema/Skin allergies Yes
Child 1 born year	☐	☐	☐	☐
Child 2 born year	☐	☐	☐	☐
Child 3 born year	☐	☐	☐	☐
Child 4 born year	☐	☐	☐	☐
Child 5 born year	☐	☐	☐	☐
Child 6 born year	☐	☐	☐	☐
Child 7 born year	☐	☐	☐	☐

General health

54. Does your gum bleed when you brush your teeth? *(tick one box only)*

Always ☐ Often ☐ Sometimes ☐ Rarely ☐ Never ☐

55. How often do you usually brush your teeth? *(tick one box only)*

2 times/day or more ☐ Once daily ☐ Less than daily ☐

56. How frequently do you exercise? *(give an average, tick one box only)*

Never ☐ Less than once a week ☐ Once a week ☐ 2-3 times a week ☐ Almost every day ☐

If you do such exercise as frequently as once or more times a week:

56.1 How hard do you push yourself? *(tick one box only)*

☐ I take it easy without breaking into a sweat or losing my breath /
☐ I push myself so hard that I lose my breath and break into a sweat /
☐ I push myself to near-exhaustion

56.2 How long does each session last? *(give an average, tick one box only)*

Less than 15 minutes ☐ 16-30 minutes ☐ 30 minutes to 1 hour ☐ More than 1 hour ☐

8

Sleep and daytime symptoms

The numbers mean:

1: Never or almost never
2: Less than once a week
3: Once or twice a week
4: 3- 5 nights/days a week
5: Almost every day or night

How often has it occurred in the last months *(circle one number for each question):*

57. ...that you snore loudly and disturbingly?	1	2	3	4	5
58. ...that you have heartburn or belching when you have gone to bed?	1	2	3	4	5
59. ...that you have difficulty in getting to sleep at night?	1	2	3	4	5
60. ...that you wake up repeatedly during the night?	1	2	3	4	5
61. ...that you perspire heavily during the night?	1	2	3	4	5
62. ...that you feel drowsy in the daytime?	1	2	3	4	5
63. ...that you wake up too early and have difficulty in getting to sleep again?	1	2	3	4	5

64. Have you ever had sleep apnoea diagnosed by a doctor? ☐ No ☐ Yes

If NO go to question 65, if YES:

64.1 When did you get the diagnosis of sleep apnoea? Year .....

64.2 If you are currently treated for sleep apnoea, what treatment do you have? *(more than one box may apply)*

- ☐ CPAP
- ☐ Oral appliance (bite splint)
- ☐ Previous surgery in the throat or nose
- ☐ Others

65. How long time do you usually sleep per night? .....hours and .....minutes

Other diseases

66. Have you ever had hypertension (high blood pressure) diagnosed by a doctor? ☐ No ☐ Yes

If YES:

66.1 When did you get the diagnosis hypertension (high blood pressure)? Year .....

66.2 Are you currently taking any medication for hypertension (high blood pressure)? ☐ No ☐ Yes

67. Have you ever had stroke? ☐ No ☐ Yes

67.1 *If YES,* when did you have stroke (for the first time)? Year .....

68. Have you ever been treated in hospital because of heart infarction or angina pectoris? ☐ No ☐ Yes

68.1 *If YES,* when were you treated (for the first time) at a hospital because of heart infarction or angina pectoris? Year .....

69. Have you ever had diabetes diagnosed by a doctor? ☐ No ☐ Yes

If YES:

69.1 When did you get the diagnosis diabetes? Year: .....

69.2 What treatment are you currently using for diabetes? *(tick one box only)*

Insulin	Tablets	Both insulin and tablets	Only diet
☐	☐	☐	☐

70. Have you ever had ulcerative collitis? ☐ No ☐ Yes

70.1 *If YES:* how old were you when the disease started? ..... years

71. Have you ever had Crohn's disease? ☐ No ☐ Yes

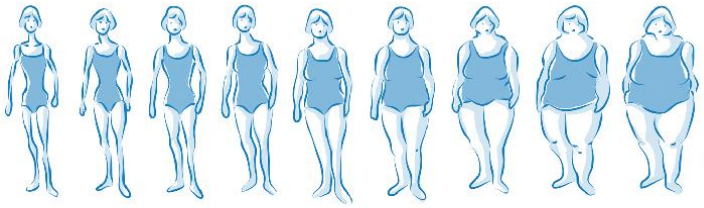
71.1 *If YES:* how old were you when the disease started? .....years

10

Body shape

72. What picture best describes your body shape at each age
(tick one box only for each age / period you have reached)

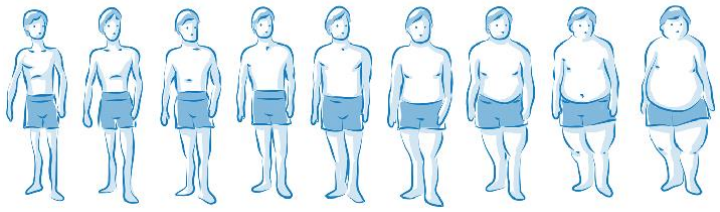
WOMEN



Current	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 8 years	☐	☐	☐	☐	☐	☐	☐	☐	☐
At first menstruation	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 30	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 45	☐	☐	☐	☐	☐	☐	☐	☐	☐
At menopause	☐	☐	☐	☐	☐	☐	☐	☐	☐

(periods stopped 12 months or more)

MEN



Current	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 8 years	☐	☐	☐	☐	☐	☐	☐	☐	☐
At age voice broke	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 30	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 45	☐	☐	☐	☐	☐	☐	☐	☐	☐
Age 55	☐	☐	☐	☐	☐	☐	☐	☐	☐

73. What picture best describes the body shape of each of your biological parents at age 50 years?

Mother	☐	☐	☐	☐	☐	☐	☐	☐	☐
Father	☐	☐	☐	☐	☐	☐	☐	☐	☐

Don't know ☐ Mother
☐ Father

Information and contact conscent

In case we need to get in touch with you again please write your telephone number below

Telephone number: Mobile phone .....

Daytime .....

Evening .....

THANK YOU FOR YOUR HELP

Appendix III

Invitation letter for European Respiratory Society, International Congress 2022

20/06/2022

Dear Mr. Sanjay Gyawali,

We are pleased to inform you that your abstract entitled:

"Association of tuberculosis infection with asthma in a european multinational cohort"

has been selected for presentation in Thematic poster entitled

"Miscellaneous: biomarkers for the management of chronic lung diseases"

at the ERS International Congress 2022. The session will be held on 04/09/2022 from 08:30 to 09:30 in TP-13.

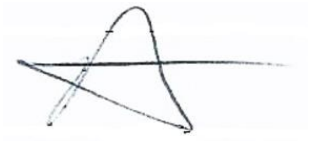
Carefully read the instructions and FAQ on how to prepare your abstract presentation on our website.

In order to increase the visibility of your accepted abstract at Congress outside of your abstract's session type you need to create an E-Poster in addition to your onsite .pptx presentation/ or poster presentation. All abstracts and E-Posters will be available on the ERS Congress Platform from 22 August 2022.

We would like to point out that:

1. At least one of the authors (the presenting author) must be a registered as participant at the ERS Congress. In order to register, please go to our registration webpage.
2. No changes can be made to the abstract content.
3. Real or perceived conflicts of interest that relate to your presentation must be disclosed when presenting your abstract (on poster or on slide presentation) and on your E-Poster.
4. If, for any reason, you have to cancel your presentation, it is important that you notify us immediately. Use the following contact details:
   - To cancel your abstract presentation both in the *Programme* and the *Publication*: ERS Scientific Dept. in Lausanne by e-mail: abstracts@ersnet.org
   - To cancel your registration to the Congress, please contact the ERS Congress Registration team at congress.registrations@ersnet.org. You may consult the General Terms & Conditions at any time.

Yours sincerely,
ERS Scientific Programme department on behalf of



Prof. Chris Brightling, ERS Science Council Chair

N.B. No-shows:
Every year, a number of authors with accepted abstracts fail to attend the Congress or to present their work.

We wish to remind you that if you do not show up, or do not have a co-author present in your place, you may not be invited to present your work at future ERS meetings.

https://www.ersnet.org/

Appendix IV

Guidelines from Journal

Respiratory Research

Research

Presubmission enquiries

If you wish to make a presubmission enquiry about the suitability of your manuscript, please email the editors who will respond to your enquiry as soon as possible.

Criteria

Research articles should report on original primary research.

Respiratory Research strongly encourages that all datasets on which the conclusions of the paper rely should be available to readers. We encourage authors to ensure that their datasets are either deposited in publicly available repositories (where available and appropriate) or presented in the main manuscript or additional supporting files whenever possible. Please see Springer Nature's information on recommended repositories.

Preparing your manuscript

The information below details the section headings that you should include in your manuscript and what information should be within each section.

Please note that your manuscript must include a 'Declarations' section including all of the subheadings (please see below for more information).

Title page

The title page should:

- present a title that includes, if appropriate, the study design e.g.:
  - "A versus B in the treatment of C: a randomized controlled trial", "X is a risk factor for Y: a case control study", "What is the impact of factor X on subject Y: A systematic review"
  - or for non-clinical or non-research studies a description of what the article reports
- list the full names and institutional addresses for all authors
  - if a collaboration group should be listed as an author, please list the Group name as an author. If you would like the names of the individual members of the Group to be searchable through their individual PubMed records, please include this information in the "Acknowledgements" section in accordance with the instructions below
- indicate the corresponding author

Abstract

The Abstract should not exceed 350 words. Please minimize the use of abbreviations and do not cite references in the abstract. Reports of randomized controlled trials should follow the CONSORT extension for abstracts. The abstract must include the following separate sections:

- **Background:** the context and purpose of the study
- **Methods:** how the study was performed and statistical tests used
- **Results:** the main findings
- **Conclusions:** brief summary and potential implications
- **Trial registration:** If your article reports the results of a health care intervention on human participants, it must be registered in an appropriate registry and the registration number and date of registration should be in stated in this section. If it was not registered prospectively (before enrollment of the first participant), you should include the words 'retrospectively registered'. See our editorial policies for more information on trial registration

Keywords

Three to ten keywords representing the main content of the article.

Background

The Background section should explain the background to the study, its aims, a summary of the existing literature and why this study was necessary or its contribution to the field.

Methods

The methods section should include:

- the aim, design and setting of the study
- the characteristics of participants or description of materials
- a clear description of all processes, interventions and comparisons. Generic drug names should generally be used. When proprietary brands are used in research, include the brand names in parentheses
- the type of statistical analysis used, including a power calculation if appropriate

Results

This should include the findings of the study including, if appropriate, results of statistical analysis which must be included either in the text or as tables and figures.

Discussion

This section should discuss the implications of the findings in context of existing research and highlight limitations of the study.

Conclusions

This should state clearly the main conclusions and provide an explanation of the importance and relevance of the study reported.

List of abbreviations

If abbreviations are used in the text they should be defined in the text at first use, and a list of abbreviations should be provided.

Declarations

All manuscripts must contain the following sections under the heading 'Declarations':

- Ethics approval and consent to participate
- Consent for publication
- Availability of data and materials
- Competing interests
- Funding
- Authors' contributions
- Acknowledgements
- Authors' information (optional)

Please see below for details on the information to be included in these sections.

If any of the sections are not relevant to your manuscript, please include the heading and write 'Not applicable' for that section.