Asthma risk and eosinophilic airway inflammation in allergen-exposed workers

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Abstract

Occupational asthma (OA) accounts for up to 15% of all adult-onset asthma. Though OA is preventable it is under-recognised in the UK. Fractional exhaled nitric oxide (FE_{NO}) is a non-invasive measure of eosinophilic airway inflammation used in asthma management, but its use in occupational settings is debated. In order to understand asthma risk and the role of airway inflammation in allergen-exposed workers, this thesis examined two hypotheses: firstly, that risk of respiratory symptoms and asthma increases with allergen exposure; and secondly, that airway inflammation relates to asthma risk in exposed populations.

A systematic review of asthma risk in woodworkers explores existing evidence for associations between allergen exposure and asthma. Three cross-sectional studies, including 773 workers, examine allergen exposure and asthma, airway inflammation, and lung function in the wood, foundry, and laboratory animal industries.

Symptoms were prevalent but did not clearly relate to allergen exposure. In woodworkers, atopy was the strongest modifier of asthma risk. Airway inflammation was common in foundry workers and related to increasing isocyanate exposure. Laboratory animal workers with more than three years' exposure had more airway inflammation and poorer spirometry. There was little overlap between airway inflammation and airflow obstruction in symptomatic workers. Airway inflammation significantly increased the risk of work-related symptoms and asthma among symptomatic workers. However, no clear relationship between allergen exposure and airway inflammation was found.

In OA, FE_{NO} could be useful in a number of ways: in health surveillance; in asthma diagnosis; and as a measure of allergen exposure. Longitudinal studies are needed to explore how airway inflammation relates to future asthma risk, and to understand how workplace allergen exposures modify airway inflammation in workers at risk of OA.

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Declaration

I, the author, confirm that the thesis is my own work. I am aware of the University's Guidance on the Use of Unfair Means (www.sheffield.ac.uk/ssid/unfair-means). This work has not been previously been presented for an award at this, or any other, university.

The majority of this data was collected as part of the HSE's Health Strategic Research Programme (SRP) into respiratory diseases, conducted whilst working at Centre for Workplace Health (CWH). Additional data on laboratory animal workers was collected as part of the Safe Practices in Reducing Allergy in Laboratories (SPIRAL) study, led by Dr Johanna Feary from the National Heart and Lung Institute, Imperial College London. All research was granted NHS REC ethics approval: references to each approval are included in the relevant chapters.

The concept for this study, design, data synthesis and analysis, and writing of this thesis is my own work. A version of Chapter two has been published previously (Wiggans RE, Evans G, Fishwick D, Barber CM. Asthma in furniture and wood processing workers: a systematic review. *Occupational Medicine*, 2016: 66(3): 193-201). I was the lead author on that publication, published under Open Access copyright. Clinical data on woodworkers, reported in Chapter two, was collected alongside my CWH colleagues Jade Sumner and Ed Robinson. The data presented in Chapter three on foundry workers was collected by CWH between 2011 and 2014. As part of the longitudinal project and by way of contribution to the data described in Chapter four, I: constructed a database; coded, cleaned and refined the data; co-developed a job-exposure matrix (JEM); designed and undertook an analysis plan; and conducted further recruitment visits to foundries. This represents the first analysis of this data, which has not yet been reported elsewhere. Data reported in Chapter five was collected during the SPIRAL study alongside Dr Johanna Feary and Bernadette Fitzgerald and is reproduced with Dr Feary's kind permission.

Introduction

1.1 The global burden of asthma

Asthma is a chronic allergic disease of the small and medium-sized airways. Its prevalence in northern European countries has been increasing over the past decades, and incidence in the UK is among the highest in the world with one in 12 adults and up to one in five children affected (1). Asthma is a treatable condition, yet deaths due to asthma attacks remain unacceptably common. In 2014 the National Review of Asthma Deaths (NRAD) issued a report criticising asthma care in the UK and highlighted the importance of recognising and modifying disease risk factors to avoid preventable deaths (2). As well as being life-threatening, asthma has a huge impact on patient morbidity. Globally, an estimated 15 million disability-adjusted life years (DALYs) are lost per year due to asthma (3).

Asthma is characterised by intermittent symptoms including wheeze, cough, chest tightness, or dyspnoea. These symptoms occur in the presence of variable airflow obstruction and demonstrate reversibility, with an improvement in airflow following the administration of bronchodilators (4). Variable airflow obstruction is a defining feature of asthma and symptoms may vary significantly from day to day. The clinical picture of asthma is heterogeneous and varies between individuals in its symptom predominance, associated biomarkers, treatment response, severity and prognosis. As a result, it is increasingly recognised as a spectrum of disease with distinct disease phenotypes and endotypes (5). This recognition has prompted the development of a number of targeted asthma therapies and specific asthma biomarkers that are increasingly used in the monitoring and treatment of the disease.

Asthma contributes £1 billion per year to NHS costs (6). However, both economic and health costs may impact elsewhere and often go unrecognised. A diagnosis of asthma presents a number of

potential economic issues to the individual. Many adults with asthma are of working age. Productivity when at work may be affected if ill-health or medication use impairs individual ability to perform duties at work. Poor health may lead to unemployment or early retirement, in turn leading to loss of earnings, an inability to contribute to general taxation, and dependence on state subsidies. Around 50% of the economic costs of asthma fall outside direct costs to health services, including loss of individual earnings, reduced national taxation, and loss of productivity at work (7). Efforts to reduce asthma incidence and severity in working-age adults have benefits not only in morbidity and mortality reduction, but also in significant improvements in productivity and unemployment (8). Thus, working age asthmatics may suffer disproportionately in social, financial and personal terms. Therefore, underscoring and understanding the intersection between asthma, work, and the individual is fundamental to improving outcomes for such people.

1.2 Occupational asthma: an opportunity to improve asthma outcomes

Occupational asthma (OA) is typified by the development of asthma symptoms following exposures to an allergen in the workplace. The British Occupational Health and Research Foundation (BOHRF) defines OA as 'asthma induced by exposure in the working environment to airborne dusts, vapours, or fumes, in workers with or without pre-existing asthma' (9). OA may be sensitiser or irritant-induced; the former characterised by symptoms following a latent period of exposure to a sensitising agent, the latter following an intense exposure to an irritant dust, vapour, gas or fume at work (also known as acute irritant induced asthma or AIIA) or from low-dose exposure to irritants such as cleaning agents (10). Population estimates indicate that 10-25% of all adult-onset asthma is occupational, and there are approximately 3000 new cases annually in the UK (11-13). The incidence of OA may be underestimated by a factor of up to 50%: many reporting schemes rely on physician-diagnosed cases but population-based studies suggest the incidence is much higher (9). A further 25% of workers with non-occupational asthma may have work-exacerbated disease (work-exacerbated asthma or WEA). Unique to OA is the prospect of cure, but only if the disease is

identified early and the patient moved away from the offending exposure (11). Therefore, the recognition and prevention of OA and WEA presents a huge opportunity to reduce the associated morbidity and mortality not only of these conditions, but also of adult asthma as a whole.

1.2.1 The burden of OA in the UK

A diagnosis of occupational asthma is particularly damaging as, by definition, it impacts employment as well as health. Ongoing exposure to respiratory sensitisers exacerbates OA and is associated with a poorer prognosis (14). Early detection and removal from exposure to the known cause represents the best health outcome for patients. However, many workers with OA are unable to be relocated to a non-exposed environment: their employer may be too small to accommodate relocation; there may be no alternative roles available in which to move; or individuals may be self-employed (9). Evidence from the UK suggests that employment outcomes after a diagnosis of OA are poor; one third of patients are unemployed 6 years after diagnosis, suffering an associated loss of income (9). The majority of people are dependent on their job for financial stability, and they may provide additional support for their spouse, children, or other dependents. A diagnosis of OA therefore introduces a direct conflict between health and wealth, and many individuals continue to be exposed in the workplace, through choice or necessity, sacrificing their health to a loss of earnings (15). The personal impact of an OA diagnosis can be devastating. In addition to developing potentially debilitating asthma, individuals report increased work-related stress, personal and professional relationship breakdown, job, income or property loss, and a loss of role in society (9, 16).

Furthermore, OA presents a significant economic burden to society through loss of earnings, tax revenue, sickness absence, and treatment costs to the National Health Service (NHS) (17). The economic burden of OA in Britain is unfairly distributed. The average worker with OA can expect to lose between 3.5 and 4.5 workdays per year, and in 2003 the cost of all cases of diagnosed OA was

estimated at £100 million. These costs are met mostly by the individual or the state, with employers incurring only 4% of the total (17). As a result, employers have little economic incentive to prevent OA among their workforce, despite health surveillance being a mandatory requirement where risk of OA cannot be reduced to levels as low as reasonably practicable and evidence that employers may lose out financially from OA due to associated productivity loss. This contrasts to practice in other countries such as Finland and Canada, where employers have a responsibility to contribute to insurance schemes that allow workers who develop occupational diseases to be retrained in alternative employment (18).

1.2.2 Occupational asthma – mechanisms for disease and trends in exposure

The pathophysiological mechanisms for OA are complex and most mechanistic understanding focuses on the development of airway inflammation. Sensitising agents may be high-molecular weight (HMW, greater than 10 kilodaltons) or low-molecular weight (LMW, less than 10 kilodaltons). Most HMW allergens act as whole antigens, directly binding to specific immunoglobulin E (IgE) on B cells and inducing a subsequent immune response. HMW allergens are also frequently associated with a T helper 2 (Th-2) lymphocyte response in which eosinophilic inflammation predominates (5, 10). In contrast, the mechanism by which LMW allergens cause disease is unclear. Some act as haptens, binding to larger proteins and inducing inflammation by binding to IgE, whereas other LMW allergens such as isocyanates are thought to up-regulate macrophage and monocyte function, causing chemokine release and a subsequent inflammatory response (10). LMW exposures most commonly cause Th-2 predominant airway inflammation, but LMW allergens have been associated with neutrophilic or mixed, rather than eosinophilic, airway inflammation (19). Specific IgE to LMW allergens is uncommon, and it lacks sensitivity for diagnosing OA in comparison with HMW allergen exposures (9).

Mechanisms for low-dose irritant-induced asthma, such as that caused by cleaning agents, are even less well established. Direct airway epithelial injury is likely to play a role, and irritant-induced asthma has largely been associated with neutrophilic inflammation and the absence of specific IgE (10). Mechanisms through which occupational exposures result in clinical exacerbations, airway remodelling, and neuronal responses remain poorly understood, but increasing efforts are being made to understand the heterogeneity of OA in association with its implicated causes (5, 10).

Nevertheless, allergen exposure is the key risk factor for sensitiser-induced OA with numerous studies reporting an increased risk of asthma in exposed versus non-exposed populations irrespective of their molecular weight (9, 12, 13, 20, 21). Many studies have also shown a positive exposure-response relationship between allergen exposure and asthma incidence, and the principle of exposure reduction has therefore become central in reducing the risk for OA (22). A principle of the Health and Safety at Work Act 1974 (HASAW) and subsequent legislation is that the risk of workplace exposure should be maintained at levels that are as low as reasonably practicable (ALARP) (23). In order to guide risk management in Britain, many common allergens have a defined workplace exposure limit (WEL) and these are enforceable by HSE (24). Through enforcing these legislative changes there has been a steady reduction in some workplace exposures over recent decades (25).

However, the most recent data from the Surveillance of Work-related and Occupational Respiratory Disease (SWORD) suggests incidence of OA has remained static over the last four years, and that rates are similar to those 10 years ago (26). The scheme identifies flour, cleaning agents, and isocyanates as the most common causes of OA in Britain (Figure 1). Metal treating and processing is identified as a high-risk occupation, (19.9 per 100,000), and a number of other manufacturing industries are associated with an increased risk of diagnosis (26). Whilst extremely useful, these data are likely to be a significant underestimate of the true burden of OA in Britain (27). There is a lack of

quantitative research on OA in industries where risk is high. As such, little is known about the risk of respiratory symptoms, asthma, and lung function in such industries, making efforts to reduce disease incidence and improve disease outcomes more challenging.

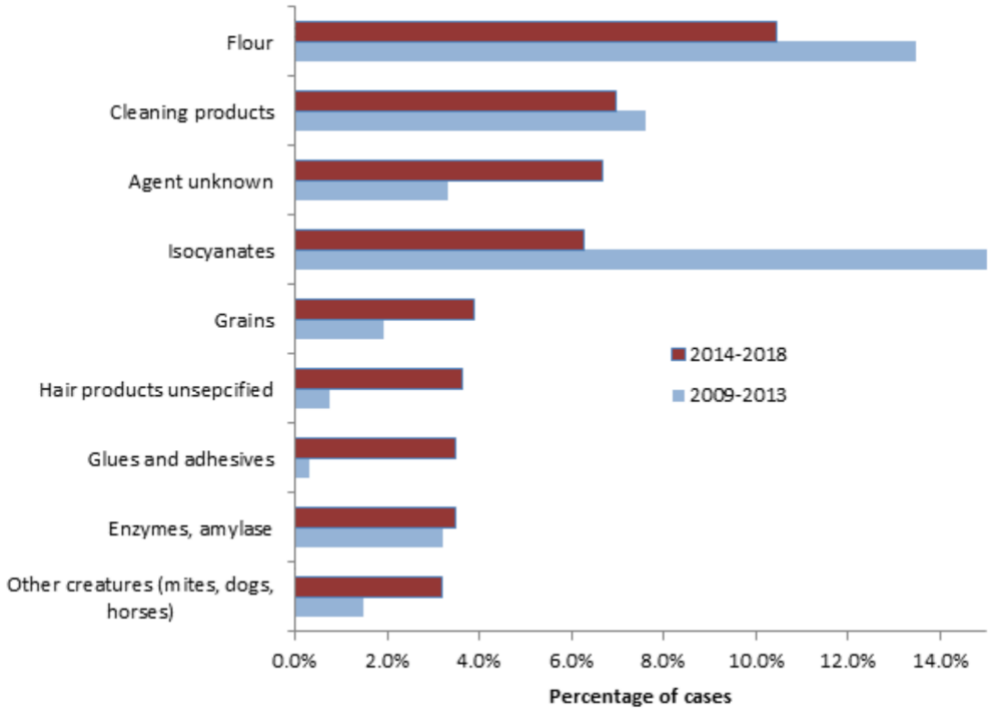

Figure 1: Most common agents for occupational asthma SWORD 2009-2013 and 2014-2018. From Work-related asthma statistics in Great Britain, 2019: Health and Safety Executive. Contains public sector information licensed under the Open Government Licence v3.0. (26) Accessed on 15/12/2019 from https://www.hse.gov.uk/statistics/causdis/asthma.pdf.

1.2.3 Health surveillance in occupational asthma

Evidence supports early recognition of the symptoms of OA to allow timely exposure modification and improve prognosis (28). Under the Control of Substances Hazardous to Health (COSHH) Regulations, health surveillance is mandatory for all workers where there is a risk of exposure to an occupational asthmagen (29). As well as reducing risk and improving outcomes for OA, health surveillance programmes may be additionally beneficial as they provide information about the global impact of workplace exposures on the workforce, present an opportunity to educate and

engage workers on the risks of their job, and may also identify non-work related illnesses that benefit from early diagnosis and management (9). In Britain, these programmes largely focus on identifying symptomatic workers with early disease through questionnaires that enquire about work-related ocular, nasal, and respiratory symptoms. Some workers employed in specific sectors or exposed to certain allergens undergo spirometry and, where relevant, sensitisation testing to occupational allergens through either skin prick testing or serum specific IgE.

In spite of this, health surveillance programmes have a number of potential flaws. Firstly, although easy to use and cheap to implement, the sensitivity and specificity of respiratory health questionnaires varies widely, with sensitivity reported at 58-100% and specificity at 45-100% to detect a diagnosis of OA (9). The true burden of OA may be underestimated where workers perceive risking their employment if they report symptoms, if symptoms are not recognised as work-related and therefore not reported as such, or if the questionnaires themselves are not sensitive for OA (30). Secondly, though spirometry remains an important part of health surveillance for OA, challenges in its use and interpretability have been reported. Commonly used spirometric measures in OA include forced expiratory volume in 1 second (FEV_1); forced vital capacity (FVC); FEV_1/FVC ratio; and peak expiratory flow (PEF). There is no universally accepted threshold of FEV_1 decline to detect a significant change that should prompt escalation of health surveillance or alteration of the working environment for those at risk of OA. In population-based studies of asthma a 15% age-adjusted loss in FEV_1 per year (31), absolute decline of 30 to 50mls per year (32) and lower limit of detection methods have all been proposed (33). Thirdly, single measures of spirometry cannot account for the natural variability of asthma, where readings may vary depending on the degree of airway inflammation present on a particular day: airway inflammation may additionally vary depending on workplace exposures. More importantly, workers with OA may have normal spirometry, so may not meet abnormality criteria until their disease has advanced significantly and may be irreversible (9). Problems with reproducibility and reliability of workplace spirometry have been reported and may

lead to underestimation of lung function and failure to identify workers with abnormal spirometry (34).

Finally, only a small proportion of at-risk workers undergo health surveillance in Britain. A recent survey suggested fewer than 20% of workplaces in sectors where the OA risk is high carried out health surveillance (35). Studies exploring asthma risk in industries where OA is highly prevalent are important in advising regulators and employers, in order to improve working conditions, provide evidence for enhanced surveillance schemes, and inform changes in legislation. These studies also represent an opportunity to explore novel physiological measurements in health surveillance, such as measures of airway inflammation, which may provide additional information to spirometry alone.

1.2.4 The role of fractional exhaled nitric oxide in diagnosing occupational asthma

Fractional exhaled nitric oxide (also called fraction of exhaled nitric oxide or FE_{NO}) is increasingly used in the diagnosis and management of asthma. Nitric oxide is a key signalling molecule in many physiological and pathological processes (36). In bronchial epithelium it is produced by upregulation of inducible nitric oxide synthase (iNOS) (37). Studies of new therapeutic drugs for asthma have shown FE_{NO} to be strongly related to interleukin (IL) 13 activity, with evidence for an association with other features of Th-2 inflammation including IL-4, sputum eosinophilia and periostin (37-39). As such, it has been used to identify individuals with eosinophilic airway disease and levels above 40 parts per billion (ppb) are associated with an increased risk of eosinophilic asthma (4). As well as being a diagnostic tool, FE_{NO} has also been utilised in asthma management. Along with other biomarkers, FE_{NO} is useful in phenotyping patients with Th2-high disease that may be more responsive to steroids or other immunomodulatory agents such as anti-IgE or anti-IL-4 or -13 therapies (5).

The use of FE_{NO} in OA is debated. Rises in FE_{NO} have been demonstrated following positive specific inhalation challenge (SIC) in workers with OA to HMW agents, with fewer studies reporting significant FE_{NO} rises following LMW challenge (40). In workplace studies, rises in FE_{NO} have been shown to predict incident bronchial hyperresponsiveness in workers at risk of OA, supporting its use as a potential health surveillance tool (41). Rises in FE_{NO} have been associated with increasing exposure to HMW agents including flour, chilli pepper, and laboratory animal allergens, in particular in those with sensitisation measurable by specific IgE (42-44). In contrast, the association between LMW exposure and FE_{NO} is less clear, with one study reporting an increase in FE_{NO} associated with increasing isocyanate exposure only in atopic non-smoking workers (45).

Guidance recommends using cut points for FE_{NO} that, correlated with clinical features, relate to an increased risk of asthma (46). Reference ranges incorporating modifying factors and cut-off points for interpretation have also been published (47). FE_{NO} has recently been recommended by the National Institute of Health and Care Excellence (NICE) in the diagnosis of asthma in primary care, and its use is supported by the recent British Thoracic Society/Scottish Intercollegiate Guideline Network (BTS/SIGN) Asthma Guideline (4, 48). FE_{NO} is simple to perform, reproducible, and equipment is increasingly available and inexpensive (46). Increasing availability and improved validation has led to FE_{NO} being considered as a potential health surveillance tool for OA. However, the implementation of FE_{NO} in screening allergen-exposed workers will require significant change to current practice, and any changes will require an evidence base.

1.2.5 Gaps in the evidence

Allergen exposure has been identified the major modifiable risk factor in the development of OA. The prevalence of OA remains high in certain sectors in Britain, including in wood, foundry, and laboratory animal workers. No recent epidemiological studies have been conducted in these industries, despite working populations being at high risk of exposure to occupational allergens and

other respiratory hazards and significant changes in practice over the last decade. There is a need to generate data to better understand the relationships between current allergen exposure and respiratory symptoms, self-reported asthma, and lung function, in order to predict and prevent future disease.

The evidence for using FE_{NO} in the assessment of OA in workplaces is conflicting. Few studies have considered the role of FE_{NO} in wood, foundry, or laboratory animal workers, and studies that have examined the role of FE_{NO} are now decades old and do not relate to current working conditions. Some studies have suggested FE_{NO} provides additional information to standard measurements used in health surveillance, and thus may help in the earlier detection or diagnosis of OA and a potential improvement in outcomes.

1.3 Asthma risk and eosinophilic airway inflammation in allergen-exposed workers

This thesis aims to address two key hypotheses:

1. It considers the relationship between exposure to respiratory allergens and asthma in three sectors – woodworking, foundry working and laboratory work – where the risk of OA is already established to be high. I hypothesise that increasing allergen exposure heightens the risk of developing respiratory symptoms and asthma in a dose-dependent fashion.
2. It addresses the utility of measuring eosinophilic airway inflammation (using FE_{NO}) in these industries, considering firstly whether FE_{NO} relates to other markers of asthma among allergen exposed workers, and secondly whether it relates to allergen exposure. By studying different exposure environments this thesis also aims to understand whether FE_{NO} relates better to markers of asthma in HMW or LMW exposed workers. I hypothesise that airway inflammation is associated with other markers of asthma, such as asthma symptoms or an

asthma diagnosis, and that risk of airway inflammation shows a dose-response effect with increasing allergen exposure.

1.4 Chapter overview

This thesis addresses the hypotheses over five chapters:

- Chapter two explores the key risk factors for OA in a systematic review, using the woodworking industry as a template.
- Chapter three evaluates the risk of asthma in a cross-sectional study of woodworkers exposed primarily to wood dust, examining associations between allergen exposure and symptoms, airway inflammation, lung function, and sensitisation.
- Chapter four examines asthma risk in foundry workers in a cross-sectional study of workers exposed to isocyanates and formaldehyde, evaluating associations between allergen exposure and respiratory symptoms, airway inflammation and lung function.
- Chapter five explores asthma risk in laboratory animal workers exposed primarily to mouse allergens and asks whether increasing allergen exposure is associated with an increased risk of respiratory symptoms, airway inflammation, abnormal spirometry, or sensitisation.
- Finally, chapter six examines FE_{NO} across the three study populations, exploring the key determinants of airway inflammation, how airway inflammation associates with other features of asthma, and any dose-response relationships with allergen exposure.

2 A systematic review of asthma risk in the furniture and wood processing industries

2.1.1 Introduction

2.1.2 Environmental wood dust exposure

Wood is a naturally occurring substance composed mainly of cellulose, hemicellulose and lignin. The remaining 1% comprises aliphatic compounds such as fatty acids and glycerides, terpenes and phenolic acids (49). Trees grow abundantly throughout the world and wood has many different industrial and domestic applications: millions of people are exposed to wood and wood by-products every day (50).

A variety of wood types are in common use. Hardwoods (deciduous angiosperms or tropical hardwoods) are generally denser and heavier in comparison to softwoods (coniferous gymnosperms). There are many timber derivatives in common use. Veneers are made from thinly sliced timber sheets. Plywood is manufactured from thin 'plies' of veneer adhered together with glue or resin. Medium-density fibreboard (MDF), oriented strand board (OSB) and chipboard are engineered from strands or chips of wood bonded together, often using urea-formaldehyde resins. Products can be used wet (unseasoned) or dry (seasoned). These forms of wood are widely available commercially and used throughout industry in the manufacture of furniture and fitted units, boat building, and in construction (51). Between 2000-2003 an estimated 3.5 million people were occupationally exposed to wood dust in the EU, with around 200,000 exposed in the UK alone (50, 52). Since that time there have been significant economic changes, and evidence points towards a downsizing of woodworking companies following the 2008 global recession (53). Exposure to wood dust in the new economic climate remains largely unquantified.

2.1.3 Wood dust and respiratory disease

Wood dust has been identified as a potentially hazardous substance for hundreds of years. Bernadino Ramazzini, the father of occupational medicine, reported respiratory symptoms in a sawer in his work De Morbis Artificium Diatriba, published in 1700 (54). A case report from 1941 describes asthma symptoms in a population of 50 men exposed to iroko wood dust (55). In her landmark 1969 study of furniture workers in High Wycombe, Hadfield described chronic respiratory symptoms and rhinitis in addition to increasing rates of sinonasal adenocarcinoma (56).

Exposure to wood dust has been most commonly associated with allergic airways disease and asthma. Prevalence of OA in some populations of wood workers has been estimated at 5%, and a significant number of workers who develop the condition continue to suffer symptoms long after their exposure has ceased (57, 58). Occupational exposure to wood dust in the UK may have been declining over time, but wood dust remains a common cause of OA in reports from SWORD (25, 26). Other respiratory diseases have been associated with wood dust exposure. Excess mortality from chronic obstructive pulmonary disease (COPD) has been reported in wood dust-exposed non-smoking Swedish construction (59). An increased risk of lung cancer has been reported in some woodworking populations (60). Epidemiological research has shown joiners to be at especially high risk of idiopathic pulmonary fibrosis (IPF) (61, 62). Although this research controlled for asbestos exposure, historically joiners have had high asbestos exposure which is thought to be a potential confounder in the risk of IPF in these people (63). Wood workers are also at increased risk of hypersensitivity pneumonitis through the handling of unseasoned mouldy wood (64, 65).

2.1.4 Molecular mechanisms of wood dust allergy

The mechanisms through which wood dust causes asthma remain poorly understood (66). Putative agents such as plicatic acid, monoterpenes, and endotoxin have been identified, but these agents are not universal to all species (67-69). Plicatic acid is a low-molecular weight agent shown to be

directly toxic to pulmonary epithelium, and causes histamine release from the basophils of workers with western red cedar (WRC) asthma (67, 70). The absence of a specific immunoglobulin E (SIgE) response to plicatic acid suggests immunological mechanisms other than type I hypersensitivity are important in the pathophysiology of WRC asthma, or at least not measurable through SIgE (71). Exposure to WRC dust has been associated with both early and late falls in FEV_1, raised sputum eosinophils, but no significant change in fractional exhaled nitric oxide (FE_{NO}) following specific inhalation challenge (SIC) (72).

In contrast to WRC asthma, SIgE has been measured in workers with OA` due to some tropical woods. Obeche (*triplochiton scleroxylon*) and iroko (*milicia excelsa*) exposure has been associated with a positive SIgE in workers with OA (73, 74). A number of high-molecular weight obeche proteins have been described with some showing cross-reactivity in individuals with latex allergy (75). Acute sustained falls in FEV_1 have been reported following SIC with obeche along with associated rises in sputum eosinophilia, suggesting that eosinophilic airway inflammation is an important feature in such asthmatic responses (76). IgE sensitisation to soft woods has been demonstrated less commonly, with prevalence of SIgE in populations exposed to pine less than 5% (77). This suggests that immunological mechanisms for wood dust exposure vary between species, as well as with exposure.

Terpenes are found in softwoods such as pine have been linked with both irritant and allergic respiratory disease. Exposures have been predominantly reported where wood is unseasoned in association with moulds, but high exposures in dry wood environments have also been reported (69, 78, 79). Formaldehyde-based stablisers used in wood composite manufacture have also been implicated as respiratory sensitisers (80).

Much of the data on the short and long-term asthmagenic effects of wood has come from studies of woodworkers exposed to unseasoned or wet wood, with an emphasis on WRC workers and the actions of plicatic acid (81, 82). WRC is not native to the UK and its use is uncommon; other hard and soft woods, along with wood composites, are used more frequently (52). Moreover, the incidence of wood dust OA has been increasing in Britain (83). Few studies have considered whether new measures of eosinophilic airway inflammation, such as FE_{NO}, are useful in evaluating risk of OA in wood-exposed populations. Consequently, the aim of this review is to explore how wood dust exposure relates to asthma and its key clinical in the furniture and wood manufacturing industries. Further, it aims to explore the evidence for a dose-response relationship between wood dust exposure and FE_{NO}.

2.2 Methods

2.2.1 Search criteria and methods

A systematic review was performed in accordance with PRISMA guidelines (84). Search terms were agreed by three team members (Table 1). The major (or, in PubMed, MeSH) heading from column one in Table 1 was combined consecutively, and in descending order, with terms from column two, followed by column three until no new references were generated. This process was repeated keeping the major term for columns two and three. A free text search using the same terms was then conducted to ensure no references were missed. Web of Science, PubMed, Embase, ProQuest, OSHUPDATE (including 'grey literature' from HSELINE, NIOSHTIC, RILOSH, and CISDOC), and the HSE e-library were searched from January 1970 to December 2014. OmniVis software (Instem Scientific v6.1.12) a reference management tool, was used to cluster references using the same terms from Table 1. This software groups references using Booleans and keywords. The clusters were then manually assessed for relevance using the following inclusion and exclusion criteria in order to generate final abstracts for review.

Table 1: Search terms employed to perform literature review. The * wild card was used to find plurals and word variants

	Column One	Column Two	Column Three
Major (MeSH) headings	Respiratory disease*	Occupation*	Wood*
Minor headings	Asthma	Work-related	Hardwood
	Lung function	Wood working	Softwood
	Airway inflammation	Exposure	Fibreboard
	Atopy	Toxicity	
	Sensitisation		
Subheadings	Symptoms	Joinery	MDF
	Dyspnoea	Furniture	Particleboard
	Wheeze	Manufacture	
	Cough	Flat pack	
	Lung function	Factory	
	Exhaled nitric oxide OR fraction* exhaled nitric oxide		

2.2.2 Abstract selection and review

Abstracts were reviewed independently by two assessors and then evaluated for agreement. Where the relevance of the abstract was not clear, or there was disagreement, the full paper was reviewed. Studies were considered eligible for inclusion where they met the following criteria: (i) adult workers from wood processing or furniture manufacturing industries; (ii) meta-analyses, controlled trials, longitudinal studies or cross-sectional analyses; (iii) respiratory or nasal symptoms, asthma, lung physiology or sensitisation identified as a study outcome; and (iv) English language papers. Studies containing data from both the wet and dry wood industries were included.

Papers were specifically excluded where they: (i) were case reports or letters; (ii) contained data only from the timber industry; (iii) contained hygiene data alone; or (iv) solely examined dermatitis, cancer, or immunological mechanisms.

2.2.3 Data grouping and analysis

As a high degree of heterogeneity was expected between the studies, articles were assessed for quality using both the Scottish Intercollegiate Guideline Network (SIGN) guidance and the Method

for Evaluating Research and Guideline Evidence (MERGE) (85, 86). MERGE was developed specifically for observational research and was employed as a second evaluation tool to ensure consistency, and that quality of papers was not under or overestimated (Table 2).

Table 2: Studies by SIGN or MERGE rating. All articles were systematically graded using both tools with a predefined proforma. Data for all but the methodology paper are presented here.

SIGN	Number of papers	MERGE	Number of papers
1-	1	**A**	14
2++	4	**B1**	14
2+	18	**B2**	15
2 -	31	**C**	11
Total:	54	**Total:**	54

Papers were grouped by common themes identified as specific study endpoints. These were: (i) ocular, nasal and respiratory symptoms; (ii) asthma; (iii) lung function; and (iv) sensitisation (including atopy, specific skin prick testing, and SIgE). Where wood exposures had been measured in the study population, evidence for a dose-response relationship was sought.

Data pertaining to each study endpoint were extracted and recorded on an agreed proforma, and then tabulated for comparison (Appendix A). Studies receiving a higher grading through MERGE or SIGN were given greater importance (Table 3). Prevalence or mean data were compared, with odds ratios or risk ratios where calculated. Exposure-response relationships were compared across studies. Confidence intervals (CI) are referred to in the text or in Table 3, where appropriate. Common confounders are included in Table 3. Due to the high degree of heterogeneity among the studies, further meta-analysis of the data was not conducted (87).

2.3 Results

2.3.1 Evidence quality and characteristics

Initial searching generated 1328 references, of which 446 abstracts were independently reviewed for relevance by the study team and 55 papers were included in the final review. Figure 2 shows the number of papers excluded at each stage.

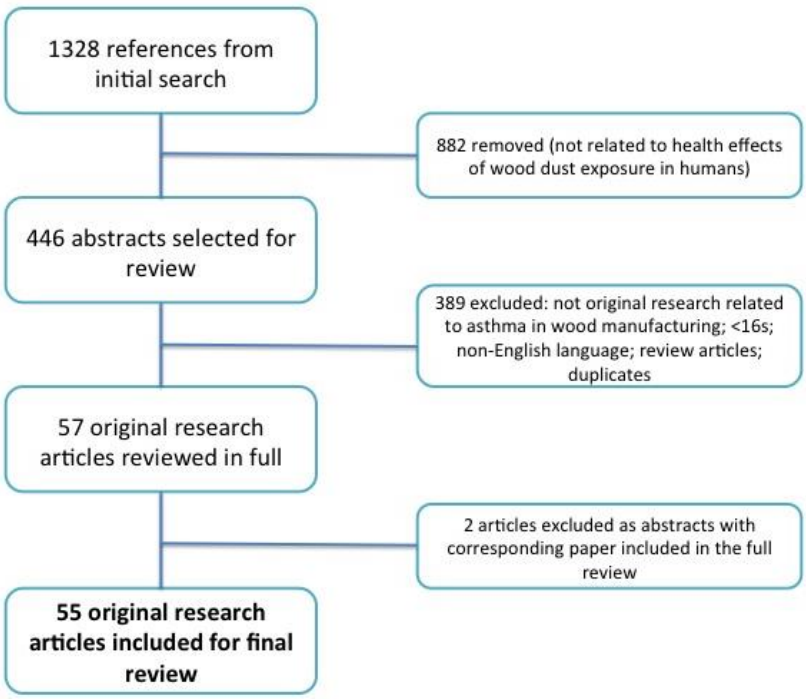

Figure 2: Flow diagram showing phases of literature review. Human adult studies in the wood processing and furniture manufacturing industry were included.

Using SIGN criteria, one of the 55 studies was rated 1-, and four were rated 2++ (Table 2). The remaining 50 were rated 2+ or below. Using MERGE, 14 of the 55 papers were graded A. Summary details for these 14 studies, grouped by study endpoint, are presented in Table 3. Details of the remaining studies are shown in Appendix A. The search identified one meta-analysis and eight longitudinal studies; the remaining studies were cross sectional or case control in design. The only

UK study was published in 1981, and a single methodology paper was also identified but contained no clinical data, and so was not included in the tabulated results (88).

Exposure measurements were made in 35 (64%) of the study populations. Seventeen studies measured inhalable dust concentrations; five of these recorded mean inhalable levels of wood dust less than 1mg/m^3. Other exposures of interest included: unspecified, total and respirable wood dust; formaldehyde; terpenes; endotoxin; β-D-glucan; and bacterial cell counts.

Thirty-three (60%) papers reported any data on prevalence or incidence of respiratory or nasal symptoms. Thirty-two (58%) measured lung physiology, including peak expiratory flow (PEFR); forced expiratory volume in one second (FEV_1); forced vital capacity (FVC); ratio of FEV_1 to FVC (FEV_1/FVC); forced expiratory flow rate (FEF); mid-expiratory flow rate (MEF); bronchodilator-induced reversibility (BDIR); bronchial hyperresponsiveness (BHR); transfer factor (TL_{CO}) and specific inhalation challenge (SIC). FE_{NO} measurements were not made in any studies. Asthma was recognised as a study endpoint in 8 (15%) of studies, with seven (13%) studies reporting sensitisation data.

Table 3: Fourteen studies rated 2++ or above with SIGN or A with MERGE, grouped by study endpoint.

Author, date	Study type; Country	Industry. Control population	No of workers exposed/controls	Wood used[a] Type(s)	Arithmetic mean exposures (mg/m³ unless otherwise stated)[b]	SIGN and MERGE rating	Common factors adjusted in analysis[c]	Summary findings (Symptom prevalence, OR, or RR with associated 95% CI)[d]
Study endpoint: Respiratory and nasal symptoms								
Rongo *et al.*, 2002.	Cross-sectional; Tanzania	Small scale wood industries. Non-exposed office workers	546/565	Variety of African hardwoods, podo, cypress, pine.	ID (gm) = 3.86	2+ A	Ag, Sm	OR in lower exposure group vs higher exposure group: wheeze = 4.6 (95% CI 1.9 – 11) vs 1.9 (0.5 – 7.3); dyspnoea = 3.2 (1.6 – 6.3) vs 2.0 (0.7 – 5.4); cough = 3.6 (2.5 – 5.1) vs 4.8 (3.0 – 7.8); sputum = 5.4 (3.6 – 8.0) vs 8.6 (5.2 – 14.0).
Schlünssen *et al.*, 2002a	Cross-sectional; Denmark	Furniture manufacture. Non-exposed factory workers from different industry within same geographical area	161/91	Softwood, wood composites, less hardwood	ID = 1.17	2+ A	Ag, Sm, Sx, He, Wt, At	VAS assessed nasal obstruction significantly difference in higher wood dust exposures: VAS difference after – before work in highest exposure group 0.63 (-10.0 - -7.7). vs 0.15 (-4.8 - -6.6) in lowest exposure group.
Study endpoint: Asthma (diagnosed by ICD 10 code, physiological testing, through national register or on physician diagnosis								
Pérez-Ríos *et al.*, 2010	Meta-analysis	Wood dust exposed. Varied between studies	19 studies		-	1- A	Included studies controlled for at least Ag and Sx.	Pooled RR 1.5 (1.25 – 1.87) for asthma in woodworkers vs general population
Heikkilä *et al.*, 2008.	Longitudinal; Finland	Wet and dry wood industries; boat building and repair; construction. All employed Finns aged 20 – 59.	158,000	Hard and softwoods, particleboard, MDF.	TD = 0.02 – 1.5	2++ A	Ag	RR 1.5 (1.2 – 1.8) for male and 1.5 (1.2 – 1.7) for female woodworkers vs general Finnish population. No relationship reported between increasing wood dust exposure and asthma risk.
Schlünssen *et al.*, 2004a	Cross-sectional; Denmark	Furniture manufacture. Non-exposed factory workers	373/71	Softwood, wood composites, less hardwood	ID (gm) = 0.96	2++ A	Ag, Sm, Sx, Ed	OR for clinical asthma diagnosed on symptoms plus BHR, BDIR or PEF variability 3.3 (1.09 – 5.53) in highest vs 2.1 (0.68 – 6.37) in lowest

		from different industry within same geographical area						exposure categories. Strong association between atopic workers and asthma.
				Study endpoint: Sensitisation				
Schlünssen *et al.*, 2011	Longitudinal; Denmark	Furniture manufacture. Non-exposed factory workers from different industry within same geographical area	1506/195	Softwood, wood composites, less hardwood	ID = 0.55	2++ A	Sm, Sx, At	OR 0.9 (0.3 – 3.5) for beech specific IgE and 0.2 (0.1 – 0.7) for pine specific IgE in those with asthma symptoms. Significantly more specific IgE positivity in high exposure group.
				Composite study endpoint, including measurements of lung function				
Jacobsen *et al.*, 2013.	Longitudinal; Demark	Furniture manufacture. Non-exposed factory workers from different industry within same geographical area	813/136	Softwood	Median ID = 0.96	2+ A	Ag, Sm, Sx, He, Wt, At, Ex	No significant difference in longitudinal lung function between workers and controls
Jacobsen *et al.*, 2009	Longitudinal; Denmark	Furniture manufacture. Non-exposed factory workers from different industry within same geographical area	1377/297	Softwood	Baseline ID = 0.94; follow up ID = 0.6	2+ A	Ag, Sm, Sx, At.	S: OR for cough (3.8; 1.5 – 9.7) and chronic bronchitis (6.0; 1.2 – 28.8) in female workers in highest vs lowest exposure categories. A: OR for ever asthma (3.4; 0.9 – 12.5) and current asthma (6.9; 0.9 – 55.8) in female workers vs controls. For female workers with no baseline symptoms OR for asthma symptoms = 11.3 (1.3 – 96.8)
Sripaiboonkij *et al.*, 2009	Cross-sectional; Thailand	Furniture manufacture. Non-exposed office workers	103/94	Rubber tree	ID = 0.02 - 2.93	2+ A	Ag, Sm, Sx, He, Wt, Ed	S: No significant difference in adjusted OR for different exposure levels A: OR 6.1 (0.7 – 53.7) for ever asthma in exposed workers. OR 8.4 (1.1 – 66.6) for ever asthma in low exposure vs control category. No

								effect across other exposure categories. LF: Incremental FEV_1 and FVC loss significantly more in factory workers vs controls and in highest vs lowest exposed groups.
Glindmeyer *et al.*, 2008.	Longitudinal; USA	Sawmill; plywood manufacture; cabinet and furniture. Between industry controls	1164	>70% hardwood in cabinet and furniture facilities. >90% softwood in sawmill and plywood	ID = 1.45; RD = 0.18	2++ A	Ag, Sm, Sx, Wt	S: Across industry groups, self-reported upper respiratory tract symptoms ranged from 45 – 53%, lower respiratory tract symptoms from 25-39%, ever asthma from 8.3 – 13%, pneumonia from 9.4 – 29%, and COPD from 2 – 4%. LF: Negative effect for respirable residual particulate matter in milling industry. Positive effect for respirable wood solids in sawmill/planing. No effect on LF in furniture/cabinetry and plywood.
Jacobsen *et al.*, 2008	Longitudinal; Denmark	Furniture manufacture. Non-exposed factory workers from different industry within same geographical area	1112/235	Softwood	Baseline ID = 0.94; follow up ID = 0.6	2+ A	Ag, Sm, Sx, He, Wt	Negative effect on LF for female woodworkers who smoked: annual change in % predicted FEV_1 per year = -1.28 ml/yr (SD 1.65). Dose response relationship observed among female workers.
Schlünssen *et al.*, 2002b	Cross-sectional; Denmark	Furniture manufacture. Non-exposed factory workers from different industry within same geographical area	2033/475	Softwood, wood composites, less hardwood	ID = 1.17	2+ A	Ag, Sm, Sx, He, Wt,	S: OR for morning cough (2.7; 1.2 – 6.0), wheeze (2.5; 1.1 – 5.7), and chronic bronchitis (6.9; 1.3 – 36.0) higher in women with 2-8 years industry service compared to controls. A: OR 5.4 (3.6 – 8.1) in female workers with atopy and self-reported asthma; 5.6 (0.9 – 34.0) for female workers with exposures >1.42mg/m^3 and self-reported asthma.

								LF: No significant difference observed when stratified by exposure.
Bohadana *et al.*, 2000	Cross-sectional l; France	Furniture manufacture. 13 unexposed workers in same industry and 200 historical controls from different industries.	114/213	Beech and oak	ID = 4.08 – 12.74	2+ A	Ag, Sm, Sx	S: No significant difference in symptoms observed between exposure groups. LF: Excess FEV_1 and FVC observed across exposure groups compared to controls. BHR increased significantly with exposures.
Mandryk *et al.*, 1999	Cross sectional; Australia	Sawmill and cabinet manufacture. Maintenance workers from same industry.	197/30	Mostly eucalyptus. Other Australian hardwoods, MDF and WRC also in use.	ID = 0.83 – 15.33; RD = 0.16 – 1.01; END ng/m^3 = 0.74 – 21.08; BDG = 0.33 – 4.63 ng/m^3	2+ A	Ag, Sm, Sx, He, Ex	S: Nasal and respiratory symptoms more common in exposed vs control populations. LF: Cross-shift loss in lung function significantly higher in joiners vs controls.

[a] Wood types where stated: MDF = medium density fibreboard; OSB = oriented strand board, WRC = western red cedar

[b] Exposures are expressed as single value or range: ID = inhalable dust, RD = respirable dust, TD = total dust, FORM = formaldehyde, END = endotoxin, TERP = terpenes, BGD = (1,3)-β-D-glucan, gm = geometric mean, ppm = parts per million.

[c] Common confounders controlled for in models: Ag = age; Sm = smoking; Sx = sex/gender; He = height; Wt = weight (including control for BMI); At = atopy; Ex = exposures (relevant exposures measured during the study); Ed = education level.

[d] Relevant themes where identified as study outcome measures: S = symptoms, A = asthma, LF = lung function, SZ = sensitisation. OR = odds ratio, RR = relative risk

2.3.2 Upper and lower airway symptoms

Most studies reported either mean symptom frequency, or odds ratios (OR), the latter measuring the increased risk of a particular health outcome as a function of exposure group; normally comparing exposed woodworkers with either lesser or non-exposed populations (Table 3 and Appendix A).

The most frequently reported respiratory symptom was cough, affecting between 6 and 80% of exposed workers (ORs ranged between 1.2 and 5.5). Wheeze and chest tightness were also commonly seen in exposed workers, with frequencies of between 9 and 40% (ORs between 1.3 and 5.9). Shortness of breath was excessively reported in woodworkers, with a range of frequencies of between 10 and 39% (ORs ranged between 1.7 and 10.6). Sputum production and bronchitis (or chronic bronchitis) were described in fewer studies, but in generally high levels (frequencies reported between 12 and 67% of exposed individuals, ORs ranged between 0.9 and 20).

Nasal symptoms in woodworkers were reported in approximately half of the 55 studies. The prevalence in exposed workers ranged between 25 and 64% in the 26 relevant studies (ORs ranged between 0.8 and 16.4). Similarly, ocular or throat symptoms were common (ranging from 20 to 51% in the included studies; ORs of between 1.1 and 13.5).

Work-related respiratory symptoms (WRRS), suggestive but not diagnostic of OA, were inconsistently defined between differing studies, and were reported in fewer studies. Where WRRS were described, the prevalence among exposed workers ranged from 25% for work-related wheeze to 52% for work-related cough (79, 89). Two studies reported increased ORs for WRRS or work-related nasal symptoms, ranging from 1.8 to 6.0 (90, 91).

Possible relationships between wood dust exposure and the presence of symptoms (or dose-response relationships) were explored in eight studies. For example, Jacobsen *et. al.* reported significantly increased risk for both cough and chronic bronchitis in female workers in high versus low exposure categories (92). The same research group showed a dose-response relationship for nasal symptoms in two earlier studies (93, 94). In contrast, three studies found no increase in symptoms when stratified by exposure (95-97). In one study of Tanzanian woodworkers, risks for wheeze and shortness of breath were reported to be higher in workers with lower wood dust exposures (91). The authors highlighted the increased use of respiratory protective equipment among workers with higher exposures as a possible explanation for this.

2.3.3 Asthma

Asthma was defined in varying ways across studies. For example, five papers identified asthma cases using worker questionnaire responses (89, 92, 94, 97, 98), and one study used insurance data and ICD-10 codes to identify population-based asthma incidence (99). A further study included a nested case-control analysis and performed objective measures of asthma including PEFR variability, BHR, and BDIR (100). A single meta-analysis was also identified; that included asthma studies where the diagnosis was made with objective measures or was self-reported through interview (101). Asthma was reported in an additional nine studies where the method for identifying cases was unclear. These papers reported asthma prevalence of between 5 and 30%, representing ORs between 3.7 and 5.5 in comparison to lesser or non-exposed populations. Studies using serial PEF recordings to support a diagnosis of asthma included Norrish *et al.*, who identified 12% of exposed furniture workers with the condition (89). Lipscomb *et. al.* reported similar prevalence, with 10.6% of their exposed population reporting “ever asthma”, and 8% reporting an asthma attack in the last 12 months (98).

In a large Finnish epidemiological study, Heikkilä *et al.* identified a relative risk (RR) of asthma of 1.5 for both male and female woodworkers compared to the general Finnish population, similar to that reported in other non-exposed blue-collar workers (RR 1.4) (99). Pérez-Ríos *et al.* found comparable results in their meta-analysis of 19 studies, with RR of 1.5 for asthma in woodworkers (101).

Three studies found atopic woodworkers to have higher risks for both asthma and airway responsiveness, again compared to non-exposed controls (89, 92, 100). Schlünssen *et al.* also found that atopic woodworkers in the highest exposure category had an increased risk for asthma and the presence of BHR (OR 22.9). Non-atopic woodworkers in the highest exposure category had an OR of 20.3 for asthma and work-related symptoms. However, no dose-response effect was seen in the study of Heikkilä *et al.* or in that of Sripaiboonkij *et al*.

Female gender was also identified to be a risk factor for asthma in one particular study (OR of 11.3 reported for asthma symptoms in women with no baseline symptoms in the longitudinal cohort), although no other studies specifically identified gender as an independent predictor for either the presence or development of asthma in woodworkers (92).

2.3.4 Lung function and airway inflammation

No papers were found reporting data on airway inflammation using sputum cell counts or FE_{NO} in workers from wood processing or manufacturing industries. One study reported no difference in nasal eosinophil cationic protein, albumin, or neutrophils between exposed woodworkers and controls but was graded as low quality (102).

All but two studies reported some lung function measures in woodworkers; including FEV_1, FVC, FEV_1/FVC, FEF, and MEF. In addition, two papers presented data on non-specific bronchial hyperresponsiveness, and a further paper reported transfer factor (TL_{CO}). It was evident that

spirometry was performed using differing protocols, suggesting that overall quality and reproducibility may not be homogeneous, and control populations differed (Table 3 and Appendix A). Eighteen studies measured lung function at a single time point (STP), 11 measured cross-shift lung function changes, and a further three assessed longitudinal decline.

78% (14) of studies reported significantly lower STP FEV_1 or FVC in their exposed populations, with two studies reporting more obstructive spirometry (as defined by an FEV_1/FVC <0.7) among exposed versus control populations (103, 104). Shamssain *et al.* also identified that 56% of woodworkers employed between 10 and 19 years had an FEV_1/FVC of less than 70%, compared with 27% employed between 1 to 9 years. By contrast, four studies reported no difference in STP lung function between exposed and control populations (Appendix A).

Bohadana *et al.* reported higher than predicted FEV_1 and FVC in exposed woodworkers, but did show a significant increase in BHR across exposure categories, with only 8% of workers in the lowest exposure category versus 27% of workers in the highest exposure category having BHR (95). However, an earlier Italian study did not demonstrate any increase in BHR between exposure populations, with OR 0.6 (95% CI 0.3 – 1.6) in woodworkers not significantly increased from controls (105).

Ten studies that measured cross-shift changes in lung function reported lower FEV_1 or FVC values in their exposed populations. Mean cross-shift loss in FEV_1 ranged between 0.11 and 14.9%; in FVC between 0.12 and 5.85%; and in MEF and FEF values between 4.8 and 22.2%. Mandryk *et al.* found cross-shift FEV_1 and FVC decrements in joiners were significantly associated with number of years wood dust exposed, with correlation coefficients of -0.77 and -0.8 respectively (78). Conversely, however, Jacobsen *et al.* found no evidence that cross-shift change correlated with longitudinal lung function decline (106).

Two further studies (including over 1000 workers) examined longitudinal lung function decline in woodworkers. In their North American cohort, Glindmeyer *et al.* measured mean inhalable dust concentrations of 1.45 mg/m^3 (range 0.77 – 2.51 mg/m^3) and mean respirable dust concentrations of 0.18 mg/m^3 (range 0.1 – 0.21 mg/m^3) (107). In this context, they reported a negative effect of cumulative exposure to respirable dust in the milling and in the sawmill/planing/plywood industries on FEV_1, FEV_1/FVC and FEF levels, and in the sawmill/planing/plywood industry for FVC. No effect was observed in furniture and cabinet workers. Conversely, Jacobsen *et al.* found accelerated lung function decline in female furniture workers who smoked, this effect being more marked in those workers with higher wood dust exposures (108). They also found small, but significant, excess longitudinal FEV_1 decline in both male and female workers still employed in the wood industry at follow-up.

2.3.5 Atopy and specific sensitisation

Seven papers reported data on sensitisation. A variety of methods were used to test sensitisation: skin prick tests (SPT) to common aeroallergens and to specific woods and moulds; total serum IgE; and serum IgE to specific wood species. Atopy (defined as SPT positivity to common aeroallergens) was common in exposed populations (ranging between a prevalence of 9 and 80%). Skovsted *et al.* reported no difference in prevalence of SIgE to pine in atopic compared to non-atopic workers (77). In their follow-up study however, Schlünssen *et al.* reported high levels of atopy among pine and beech workers, with a significant correlation between atopy and reported respiratory symptoms (109).

Where studies reported SIgE to wood types, positive results were uncommon. Ricciardi *et al.* found no SIgE to iroko wood in a group of asthmatics, all of whom had a sustained fall in PEF on SIC to iroko extract (74). Notably, this group also demonstrated a significant increase in blood eosinophils and

positive methacholine challenge post SIC suggesting these individuals had mounted a significant allergic response to iroko inhalation.

Furthermore, Skovsted *et al.* found no difference in prevalence of SIgE between exposed and non-exposed workers (both 3%), and Schlünssen *et al.* reported only 1.7% of exposed pine workers having pine-specific IgE, and 3.1% of exposed beech workers having beech-specific IgE (77, 109). The latter study did not firmly establish a relationship between specific sensitisation and respiratory symptoms but did find an association with increasing wood exposures and higher rates of SIgE: 7.8% of workers in the highest exposure category were sensitised to pine, with 9.8% of beech workers in the highest exposure category sensitised.

2.4 Discussion

A number of high-quality observational studies were identified for this review although the majority of papers were of a lower evidence rating. Only five studies were graded SIGN 2++ or above (Table 2), although MERGE identified more highly rated studies. This finding emphasises the importance of using tools specifically designed for reviewing occupational research, where the validity and applicability of studies may otherwise be underestimated (87).

Exposure to wood dust in the furniture and wood manufacturing industries was associated with an increased risk of a variety of reported respiratory symptoms, and particularly cough. This was based on evidence from both cross-sectional and longitudinal studies. Indeed, reported symptoms were more common in higher exposed woodworkers in all but one study reporting airway symptoms. It is plausible that some of this excess relates to recall bias, but there is evidence to suggest that though respiratory illness may alter symptom reporting, workers continue to self-report with high sensitivity (106).

2.4.1 Wood dust exposure, respiratory symptoms and asthma: evidence for a dose-response relationship

There was conflicting evidence to support a dose-response relationship for respiratory symptoms in woodworkers. Neither Rongo *et al.* nor Sripaiboonkij *et al.* found increasing levels of wheeze or chest tightness with increasing dust exposure, despite large study numbers and relatively high wood dust exposures; inhalable dust ranging from 0.02 – 2.93 mg/m^3 and 1.43 – 22.76 mg/m^3 respectively (91, 97).

Woodworkers in the furniture and wood manufacturing industry were also found to be at a greater risk of asthma, despite measured exposure to dust levels being lower than may be expected in some European countries. Schlünssen *et al.* found an excess of self-reported and physician-diagnosed asthma in atopic female wood workers (100). They also reported that atopic workers in the highest exposure category had significantly more asthma symptoms and BHR. Conversely, non-atopic workers in the same exposure category had less BHR and more work-related asthma symptoms. Though confidence intervals were wide, this finding suggested that non-atopic and atopic wood workers may manifest different clinical outcomes as a result of wood dust exposure. More conclusive work is needed to better understand these responses.

The only meta-analysis included in the review reported an increased relative risk for asthma in woodworkers of 1.5 compared with the general population (101). Heikkilä *et al.* found similar relative risks (1.5 in both female and male woodworkers) comparable to blue-collar workers in the same industry, although did not identify a dose effect (99). Inclusion in the study depended on inclusion in a national registry, potentially missing workers with a past, or missed, asthma diagnosis. Jacobsen *et al.* demonstrated an increased risk of asthma for female woodworkers in the highest exposure category (92). Interestingly, they also reported significantly more asthma in female workers who had left the furniture industry at follow up, suggesting a healthy worker effect (HWE).

2.4.2 Cross-shift and longitudinal lung function in wood dust exposed workers

Cross-shift changes in lung function have been reported in other industries including dairy workers and mussel pickers (110, 111). Their utility is uncertain when diagnosing occupational respiratory problems (9). Whilst these measures evidently document an acute respiratory response to the work environment, they may be affected by the normal diurnal variation in lung function, which is classically exaggerated in asthma. They may be further confounded by shift work and poor spirometry measurement, and cross-shift change has been shown to have a high specificity but low sensitivity for identifying cases of occupational asthma (9).

Jacobsen *et al.* found no relationship between acute, cross-shift changes, and longer-term lung function loss in their study of woodworkers (106). This may reflect exposures other than wood in the workplace. For example, Mandryk *et al.* demonstrated significant cross-shift lung function change in a workplace where substantial endotoxin, bacterial and fungal exposures were measured (78).

This review identified conflicting evidence for impairment of lung function among exposed woodworkers. Certain cross-sectional studies reported a difference in measured values between exposed and control populations. However, evidence for excess longitudinal lung function decline was only demonstrated in certain subgroups. Two studies reported annual decline in lung function, but with mixed conclusions. Jacobsen *et al.* found female smokers exposed to wood dust had significantly accelerated lung function decline (108). In contrast, Glindmeyer *et al.* found no relationship between longitudinal lung function decline and employment in the furniture or cabinet making industry (107). Studies in WRC workers have demonstrated irreversible longitudinal lung function decline, findings that have been replicated with other forms of OA (14, 72).

No papers were found reporting data on non-invasive measures of airway inflammation in workers from the wood processing or manufacturing industries. Only one paper of low-quality reported data

on nasal cell counts and protein showing no difference between exposed and control populations. A rise in sputum eosinophils has previously been shown among workers with WRC asthma and a positive SIC (112). Sputum eosinophilia, but not FE_{NO}, has previously been shown to relate to FEV_1 impairment and asthma severity in WRC asthma (72). However, the same study group found that FE_{NO} correlated with sputum eosinophilia, suggesting it may be a useful non-invasive marker for eosinophilic airway inflammation in WRC workers. FE_{NO} is increasingly used in the diagnosis and management of OA and is recommended in recent BTS/SIGN and NICE asthma guidelines (4, 48). FE_{NO} has been shown to predict incident bronchial hyperresponsiveness in exposed bakers and hairdressers at risk of OA (41). FE_{NO} has also been shown to reduce following allergen exposure reduction in farmers, with a corresponding reduction in respiratory symptoms (113). Further study of airway inflammation in woodworkers is needed to examine its potential usefulness in surveillance and diagnosis of work-related asthma.

2.4.3 Mechanisms of disease

Whilst this review provides some information on mechanisms for asthma in wood dust-exposed populations, it is not the aim of this study to review all the evidence relating to mechanisms by which wood dust causes respiratory disease. Whilst the exact causative agent (or agents) responsible is not clearly identified, one study suggested that high levels of endotoxin were linked to the reporting of cough in furniture workers (79). In addition, the relationship between chronic bronchitis and wood processing was particularly strong, even when the effects of smoking had been taken into consideration. Jacobsen *et al.* reported increased risks for chronic bronchitis in a longitudinal study of female woodworkers and showed that increasing exposures in this group increased the likelihood of bronchitis (92). Whether this finding represented an “irritant” effect is not clear, but symptoms may have reflected underlying disease mechanisms not typical of type 1 hypersensitivity or eosinophilic allergy (66). In addition to intensity of exposure, the propensity of wood to cause asthma is likely to be influenced by other factors including the chemical properties of specific

species, the particle size generated, the route of exposure, and individual susceptibility to disease (114).

Whilst certain studies supported the development of sensitisation to wood dust as being a potentially important process in the development of respiratory symptoms and asthma, the mechanism remains unclear. Skovsted *et al.* identified a potentially pathogenic protein band in beech and pine, but reported very low levels of pine and beech specific IgE in exposed workers (77). Similarly, Ricciardi *et al.* reported 100% SIC positivity in patients with iroko asthma, but 0% specific IgE positivity, despite these patients demonstrating a sustained asthmatic response to iroko extract (74). Overall, IgE positivity was low throughout the included studies. A number of non-IgE mechanisms for sensitisation have been reported: further research in this field is important to better understand the both micro- and macroenvironmental circumstances that give rise to allergic disease in order to improve controls, reduce risk to workforces, and develop more targeted therapies (114).

2.4.4 Limitations

This review is the only systematic review of asthma in workers employed in the furniture or wood manufacturing industries. Its findings are comparable to a previous review of the dry wood industry (82). Since a greater number of papers were included in the current review, and a wide range of search terms employed over a long period of interest, it is unlikely that a significant number of relevant studies were missed. Although there was heterogeneity between studies, consideration of a range of endpoints allowed us to extract data relevant to several areas of interest and raise points for further investigation.

There are, however, certain downsides to consider. Our review findings were difficult to compare for a number of reasons. Exposed populations were drawn from a variety of sources including worksites, outbreaks, or from hospital cohorts. Population size, country, exposure level and design also varied

between papers. A number of studies were published prior to the year 2000, and before mandatory changes to exposure limits in both North America and Europe. As such, some data may not accurately reflect current practice; although recent international data suggests that exposure to wood dust continues to differ both within and between countries (50).

In addition, exposure misclassification represents an important potential source of bias within this review. Some studies reported data on inhalable or respirable dust, whereas others reported only total or non-specific dust readings. Variation in measurement techniques could also have influenced clinical risk attributed to exposure and bias results, potentially underestimating (or overestimating) the magnitude of past exposures especially where workers have subsequently moved to lower exposed tasks (88).

2.4.5 Summary

In summary, this review found an increased risk of respiratory symptoms and asthma in people working in the wood processing and furniture manufacturing industries. Accelerated lung function decline due to ongoing exposure was evident for specific study groups. It is important to note that although much of the more robust research included was conducted in lower exposure environments, ill-health occurred across all exposure groups. Further study exploring measures of airway inflammation in the workplace may help explore the mechanisms through which wood dust causes respiratory disease and asthma, and more importantly how to define interventions to reduce wood-related respiratory ill-health.

3 Respiratory symptoms, airway inflammation and asthma in British woodworkers

3.1 Background

3.1.1 Asthma and wood dust exposure

Between 2000 and 2003 an estimated 200,000 people were annually exposed to wood dust in the UK, working in diverse sectors such as forestry, furniture manufacture, boat building, and construction (52). Exposures to wood dust have changed significantly over the last two decades, with increased mechanisation of tasks, reduction in hard wood use, increase in use of wood composites, and an overall reduction in exposure (25). However, despite exposure reduction and a smaller population of workers exposed, OA remains prevalent among woodworkers. Over the last decade, SWORD data has shown an increase in the incidence of OA caused by wood dust, with wood dust among the top five causes of OA in Great Britain in 2019 (26). Data from reporting schemes in France and Australia also identify wood dust as a leading cause of occupational asthma (115, 116). Despite this, few recent studies have studied the risk of OA in woodworkers (117).

Wood dust exposure is the key risk factor for asthma in woodworkers. Population-based studies have demonstrated woodworkers have a 50% increased risk of asthma compared to the general population (101). Wood dust exposure is associated with an increased risk of respiratory symptoms, particularly cough (117). Population-based studies have demonstrated a link between hard and soft wood dust exposure and airway responsiveness (100), excess lung function decline (108), and asthma (99). Both immediate and dual asthmatic reactions have been reported following specific inhalation challenge to hardwoods including oak (118), iroko (74), and chengal wood (119). Western red cedar is a well-documented cause of OA, and studies have reported increased presence of inflammatory cells including eosinophils in bronchioalveolar lavage of patients with WRC OA (120).

3.1.2 Mechanisms of wood dust OA

A variety of wood species, including both hard and soft woods, have been implicated in causing OA (117). However, the immunological mechanism through which wood dust causes OA remains unclear. Wood dust has been categorised as both an HMW and LMW allergen (121). IgE mechanisms of disease have been inconsistently described. Some wood species, in particular tropical hardwoods, have been associated with detectable SIgE and associated HMW proteins (122). However, most cases of wood dust OA have not been associated with detectable SIgE. For WRC OA, an IgE-independent process where plicatic acid acts as a hapten and initiates histamine release and basophil degranulation has been described (70). Other studies have demonstrated hard and soft wood constituents to have both inflammatory and genotoxic effects on pulmonary epithelium *in vivo*, suggesting wood may induce asthma through direct bronchial injury rather than through an immunological mechanism (67). Since OA is commonly described by its causative exposure, characterising relationships between wood dust exposure and OA is challenging, particularly since workers may be exposed to multiple wood species.

3.1.3 Measures of airway inflammation in wood dust exposed workers

Exposure to wood dust is regulated by HSE and where exposures cannot be reduced to levels as low as reasonably practicable, health surveillance is mandatory (29). Low-level health surveillance includes worker respiratory questionnaires, with those at higher risk of exposure also requiring serial spirometry (123). In addition, measurements of airway inflammation such as non-specific BHR and FE_{NO} may be used for diagnosing wood dust OA, but their use in such settings is not established (117). Since assessment of BHR requires specialist delivery, it has not been routinely used in health surveillance programmes. In contrast, FE_{NO} is non-invasive, easy to use, and reproducible, and therefore has been explored as a potentially useful tool for identifying workers at risk of OA (124). FE_{NO} is recommended in the diagnosis and monitoring of asthma in the UK (4). Its presence relates to Th-2 inflammation and it is particularly associated with other inflammatory markers including

eosinophils, IL-4, IL-13, and antigen-specific IgE (125). This inflammatory response pattern has been more strongly associated with HMW rather than LMW exposures in OA (126). In WRC workers, rises in FE_{NO} have been shown to correlate strongly with increases in sputum eosinophilia but not with positive SIC (112). Since wood dust is varyingly characterised as both an HMW and LMW allergen, the utility of FE_{NO} as a measure of airway inflammation in wood dust OA has yet to be determined.

3.1.4 Exposure thresholds for asthma in wood dust exposure

Respiratory symptoms, SIgE, NSBHR and OA have all been reported at wood dust exposures lower than the current UK WEL of 5mg/m^3 (117). Exposures of less than 2 mg/m^3 inhalable dust have been associated with increased respiratory symptoms, bronchial hyperresponsiveness, and reduced lung function compared to lower or non-exposed groups (100, 108). There is no safe limit for exposure to an asthmagen, and this suggests that wood dust OA occurs at levels lower than the current UK WEL.

There have been no recent studies of respiratory ill-health in British woodworkers despite wood dust being consistently reported as a common cause of OA in UK for the past decade, the changing shape of the industry due to economic pressures, and the increasing use of wood composite materials. No previous studies have explored widely available measures of airway inflammation such as FE_{NO} and its relationship with measured workplace exposures, respiratory symptoms, and asthma outcomes in woodworkers. The aim of this study is to examine how wood dust exposure influences respiratory symptoms, airway inflammation, sensitisation, and abnormal lung function among British woodworkers.

This study was reviewed and approved by the NHS Health Research Authority National Research Ethics Committee: London (REC reference 14/LO/1262).

3.2 Methods

Research into woodworkers was conducted by CWH. I developed the study protocol, obtained ethical approval, and conducted data collection visits alongside my CWH colleagues Jade Sumner and Ed Robinson. I developed a data analysis plan alongside Charlotte Young, HSE statistician. I was responsible for data management, analysis, and interpretation. The data from this study has not been reported elsewhere.

3.2.1 Site identification

British woodworkers were recruited to a cross-sectional study conducted as part of HSE's Health SRP. All registered worksites manufacturing or processing wood or timber within Great Britain were potentially eligible to participate. Worksites that had recent (within three months) or ongoing HSE investigations were not approached to take part in the study. Initially woodworking companies participating in an HSE occupational hygiene study in 1999/2000 were approached to take part (52). The original hygiene study included worksites from the following sectors involving wood dust exposure:

- timber and sawmills;
- furniture manufacture;
- wood processing;
- boat building.

No construction sites were included in the 1999/2000 study.

Further sites were identified using the HSE Corporate Operational Information System (COIN) database. Every intervention by a HSE duty holder is recorded on the COIN system, which holds an address and contact number of most worksites in Great Britain (127).

Managers or health and safety representatives at worksites were contacted via telephone. Interested worksites were sent information sheets about the study for circulation to management and employees (Appendix B). Each worksite then received a recruitment visit from the study team where a detailed explanation of the study was provided to workers and site managers, and workers and managerial staff were given the opportunity to ask questions about the research. Worksites were then re-contacted the following week and asked whether they agreed to participate. Arrangements were then made to re-visit participating worksites at mutually agreeable times.

3.2.2 Power calculation

Since there were no recent studies reporting asthma rates in British woodworkers, a study reporting equivalent exposures was used to determine the number of workers required to achieve 80% study power at a 5% significance level. The most recent British exposure data in woodworkers reported median inhalable wood dust exposures of 2.6 mg/m^3 (52). In their study of New Zealand woodworkers, Norrish *et. al.* reported 25% of exposed woodworkers had work-related chest tightness or wheeze at a mean inhalable dust exposure of 3.6 mg/m^3 (89). The same research group reported a prevalence of asthma of 18% in sawmill workers, with an odds ratio (OR) of 1.4 between high and low exposure groups (128). This research compares to more recent evidence from British bakery workers where work-related respiratory symptoms were reported in 18.7% of workers, with mean OR for medium and high exposure versus low exposure groups of 2.3 and 2.0 respectively (129). Mean exposures to flour dust in three groups were 2.1, 3.6 and 5.0 mg/m^3 respectively.

On this basis, a power calculation was made using logistic regression in MATLAB (130). A sample of 240 workers was estimated to provide 80% power to detect an association between wood dust exposure and work-related respiratory symptoms with an expected 95% CI of 13.4% to 23.5%, assuming a two-sided 5% significance level, a true underlying symptom prevalence of 18% and an OR of 1.4 per doubling of exposure.

3.2.3 Worker recruitment

All workers exposed to wood dust and over the age of 16 at were eligible to participate. All workers on site during the visit were invited to take part. Worksites varied in size and shift pattern. Data collection visits included extended day, evening, or night visits to ensure the maximum number of workers, including late and night shift workers, were able to participate. Multiple visits were made to larger worksites to ensure saturation of participation.

3.2.4 Questionnaire

Each worker underwent an interviewer-administered validated questionnaire detailing upper and lower respiratory symptoms, nasal symptoms, and ocular symptoms based on the Medical Research Council (MRC) and European Community Respiratory Health Survey (ECRHS) questionnaires (Appendix B) (12, 131). Questions on asthma control and medication use were asked using the asthma control test (ACT) (132). A detailed job history was taken including current and previous jobs and processes and materials involved. Use of any respiratory protective equipment (RPE) was recorded, including type (if any) and length of time used.

3.2.5 Spirometry and fractional exhaled nitric oxide measurement

All participating workers were invited to undertake spirometry and FE_{NO} measurement. FE_{NO} was performed before spirometry using a NOBreath device (Bedfont Scientific, Kent) according to ATS/ERS standards (46). A minimum of two technically acceptable blows at 50mls.s^{-1} were recorded to within 10% of one another (or 1 part per billion (ppb) for readings below 10ppb) and before spirometry wherever possible. Spirometry was measured using an NDD Easy-On spirometer (Zurich, Switzerland) according to ATS/ERS standards (133). Subjects were examined sitting and without a nose clip. A minimum of three technically acceptable manoeuvres were made, and the best two measurements of FEV_1 and FVC measured to within 150mls of one another. FEV_1, FVC, PEF, and

FEV_1/FVC ratio were recorded, and percentage predicted values generated using age, sex, height, and ethnicity standardised reference values included in the spirometry software.

3.2.6 Total IgE and specific IgE to hard and soft wood

Blood was obtained from consenting workers at worksites and kept on ice after drawing. Samples were centrifuged and frozen at HSL and underwent immunological testing for TIgE and SIgE to hard (oak, mahogany and obeche) and soft (beech, pine, cedar and silver fir) woods using standard ImmunoCAP testing (Phadia, Sweden, 2012).

3.2.7 Occupational hygiene assessment

An assessment of wood dust exposure was conducted at each participating worksite by a certified HSE occupational hygienist. Workers underwent personal exposure monitoring using the Methods for the Determination of Hazardous Substances (MDHS) 14/4 (134). In brief, a passive sampling device was worn on the lapel for the duration of a working shift. Standard eight-hour time-weighted averages (8-hr TWA) were calculated for inhalable wood dust for the tasks sampled. Worksites also underwent comprehensive occupational hygiene assessment including evaluation of material exposures, processes undertaken, exposure controls employed, application of the COSHH guidelines, and use of any RPE.

3.2.8 Job exposure matrix

Each worker was assigned a job code using task-specific codes developed for the woodworking industry (52). Where workers held more than one current position (for example a managerial and a manufacturing role), a ratio of time spent in each job was applied. The proportion of time in all positions did not exceed one. In addition, Standard Occupational Classification 2010 (SOC 2010) codes were applied to current jobs using a Computer Assisted Structured Coding Tool (CASCOT)

(135, 136). Historical jobs were assigned a three or five-digit SOC 2010 code based on the level of information provided by workers.

Inhalable wood dust exposures for each worker were estimated based on a linear mixed effects model fitted to the logarithm of the measured 8-hour TWA values (88). In total, 168 valid measurements were taken across all sites. Site was treated as a random effect and task as a mixed effect, with between-worker variation treated as normally distributed residual errors. Mean exposure for each task at each site was calculated from the corresponding mean of log-exposure estimated by the model and the residual error standard deviation. For individual workers, model exposures were then weighted by the fraction of time each worker spent undertaking each task.

3.2.9 Definitions

Work-related respiratory symptoms (WRRS) were identified where workers reported deterioration in cough, shortness of breath, wheeze, or chest tightness at work, or an improvement away from work or on holiday. Self-reported asthma was defined by individuals reporting a physician diagnosis of asthma. Current asthma was defined as either a current or past physician-diagnosis of asthma and current asthma symptoms (CAS - wheezing, nocturnal chest tightness, breathlessness on exertion, at rest, or at night, or asthma medication use within the last 12 months) as per ERCHS criteria (12).

Only technically acceptable spirometry and FE_{NO} measurements were used in the final analysis. FEV_1, FVC, PEF and FEV_1/FVC values falling two standard deviations below the mean (lower limit of normal or LLN) were considered abnormal (137). FE_{NO} values above 40ppb were considered high (4).

Participants were considered sensitised if their IgE to hard or soft wood exceeded $0.35 Ku_A/L$ (109). Atopy was defined as a TIgE above 100 kU/L (138).

3.2.10 Data analysis

Demographic details were displayed as means and standard deviations. Normally distributed data were compared using independent t-tests and ANOVA analyses for continuous variables of interest, and chi squared tests were used for categorical variables. FE_{NO} and TIgE data did not meet normality assumptions. Mann-Whitney-U tests were used to compare means, and data were log transformed for use in regression models.

The outcomes of interest including current asthma symptoms, WRRS, current asthma, FE_{NO} >40ppb, and FEV_1/FVC <LLN were used as dependent variables in logistic regression models, with quartiles of exposure as the key independent variable. Workers in the lowest exposure quartile were used as the control group. Models were adjusted for ever smoking, BMI, atopy, age, sex, and RPE use. FE_{NO} models were controlled for current smoking and height (46). Odds ratios with associated 95% confidence intervals were reported.

Linear regression models were constructed to analyse lung function and FE_{NO} data in relation to wood dust exposure. Because FE_{NO} was not normally distributed, log10 FE_{NO} was used as the dependent variable with current wood dust exposure as the key independent variable. Data were back-transformed and expressed as the geometric mean ratio (GMR) of FE_{NO} per 1mg/m^3 increase in wood dust exposure (45, 139). Models were adjusted for atopy, age, sex, current smoking, and height. Absolute FEV_1 and FVC in millilitres were used as dependents with current exposure to wood dust as the key independent variable. Models were controlled for age, sex, height, and ever having smoked more than one pack year of cigarettes.

Linear regression models were used to examine the presence of a healthy worker effect (HWE). Self-reported years of exposure to vapours, dust, gases and fumes (VDGF) was used as the dependent variable, where possible to account for exposure to wood dust in previous jobs within the

woodworking industry. Respiratory symptoms, work-related respiratory symptoms, and current asthma were used as independent variables. Models were adjusted for age, ever smoking, atopy, RPE use, and BMI.

Since asthma is a heterogeneous condition with variable manifestations, relationships between exposure and combinations of asthma markers (asthma indices) were explored (100). Logistic regression models were constructed examining the composite asthma indices: (i) CAS+atopy; (ii) CAS+FE_{NO} >40ppb; (iii) current asthma+atopy; and (iv) current asthma+FE_{NO} >40ppb across exposure quartiles. Odds ratios with associated 95% confidence intervals were reported. Finally, contingency analyses were conducted between workers with CAS, FE_{NO} >40ppb and airflow obstruction (FEV_1/FVC <LLN) using intersecting Venn diagrams created using Venny, a freely available bioinformatics tool (140). All statistical analysis was performed using SPSS Statistics, v23 (141).

3.3 Results

3.3.1 Study population

Thirty-seven companies were contacted to participate. From the original 1999/2000 list, six companies had gone out of business. A further 18 companies contacted declined to take part. If reasons for non-participation were given, they included lack of time for research, concerns about productivity loss, and an unwillingness to allow HSE into the workplace. Thirteen companies were subsequently recruited across 14 sites. Companies from the furniture manufacture, boat building, and wood processing industries were included in the study. Two-hundred-and-sixty-nine out of a possible 376 workers participated (participation rate 72%). Reasons for non-participation included inability to capture workers due to shift work, inability of workers to take time away from shift, worker annual leave, and worker refusal.

Table four lists the key demographic and exposure characteristics of the study population. The population was mostly male (n=261, 97%) with an average age of 42.4 (SD 12.6) years. Seventy (26%) workers were current smokers, with 140 (52%) ever having smoked more than one pack year. Workers had spent an average 18.9 (SD 12.8) years exposed to wood dust, with 7.9 (SD 7) years spent in their current job. The majority of workers (63%) used a mixture of hardwood, softwood, and composites, and most (70%) performed more than one task at their worksite.

Mean current exposure to wood dust was 1.9mg/m^3 (SD 0.9, IQR 1.4), with a mean cumulative exposure to wood dust of 13.9mg/m^3 (SD 14.9, IQR 19.3). Mean current exposures to wood dust were 0.69 (0.37) mg/m^3, 1.74 (0.27) mg/m^3, 2.12 (0.09) mg/m^3, and 2.98 (0.80) mg/m^3 in the lowest, low, high, and highest exposure quartiles respectively (Table 4). One third of the highest exposed workers were involved in sanding and assembly (30% vs 13% in lowest exposed, $p < 0.01$). Workers in all but the lowest exposure quartile were more likely to use RPE (p for trend <0.05). Workers in the high exposure quartile were significantly more likely to work with hardwood than workers in other exposure quartiles (54% versus 30% in lowest exposure quartile, $p < 0.01$).

Table 4: Key demographic and exposure characteristics of 269 of British woodworkers, stratified by exposure quartile. Exposure range for each quartile is reported in parentheses in mg/m^3. Number of workers in each quartile is reported in parentheses.

	Lowest exposure (0-1.19) (n=67)	Low exposure (1.20-2.00) (n=72)	High exposure (2.01-2.32) (n=63)	Highest exposure (2.33-5.44) (n=67)	Total (n=269)
		Demographics			
Age, years (SD)	42.45 (10.51)	44.64 (13.69)	41.31 (13.00)	40.93 (12.68)	42.4 (12.6)
Sex, m (%)	65 (97)	71 (99)	62 (98)	63 (94)	261 (97)
Height, cm (SD)	176.13 (8.11)	175.80 (6.25)	174.80 (10.14)	177.16 (8.20)	175.99 (8.22)
Current smoker, n (%)	17 (25)	22 (30)	14 (22)	17 (25)	70 (26)
Smoked >1 pack year, n (%)	34 (51)	46 (64)	27 (43)	33 (49)	140 (52)
		Exposure			
Hardwood/softwood/mix, n (%)	20/3/44 (30/4/66)	6/11/55 (8/15/76)	34/6/23 (54/9/27)**	16/5/46 (24/7/69)	76/25/168 (28/9/63)
Uses RPE, n (%)	44 (66)	55 (76)*	51 (81)*	57 (85)*	207 (77)
Multitasking, n (%)	38 (57)	54 (75)	56 (89)	39 (58)	187 (70)
Sawing, n (%)	4 (6)	7 (10)	1 (2)	1 (2)	13 (5)
Moulding/shaping, n (%)	1 (2)	2 (3)	0 (0)	0 (0)	3 (1)
Sanding/assembly, n (%)	9 (13)	4 (6)	1 (2)	20 (30)**	34 (13)
Cleaning/maintenance, n (%)	5 (8)	5 (7)	5 (8)	7 (10)	22 (8)
Other non-woodworking, n (%)	10 (15)	0 (0)**	0 (0)**	0 (0)**	10 (4)
Time in current job, years (SD)	7.40 (6.02)	7.91 (6.57)	8.94 (8.00)	7.28 (7.30)	7.9 (7.0)
Time in woodworking industry, years (SD)	17.95 (11.03)	19.71 (12.52)	20.86 (14.58)	17.11 (12.90)	18.9 (12.8)
Mean current exposure, mg/m^3 (SD)	0.69 (0.37)	1.74 (0.27)**	2.12 (0.09)**	2.98 (0.80)**	1.9 (0.9)
Cumulative exposure, mg/m^3/yrs (SD)	5.13 (5.95)	13.50 (11.40)**	17.93 (16.21)	19.22(19.04)	13.9 (14.9)

*p <0.05, **p <0.01. RPE = respiratory protective equipment.

Table five shows the key health characteristics of the study population, stratified by exposure quartile. Overall, CAS were common, reported by almost half the study population (n=123, 46%). CAS rates were high across exposure quartiles, although were slightly lower in the highest exposed group (39%). WRRS were less frequent, reported by 29 (11%) workers. WRRS tended to be commoner among the lower exposed, with 16% in the lowest exposed versus 3% in the highest exposed groups reporting any WRRS. There was no significant difference in prevalence of CAS or

WRRS across exposure quartiles. Work-related ocular symptoms were significantly more common in the higher exposed groups, with 22% in the highest exposed versus 9% in the lowest exposed reporting symptoms ($p<0.05$). No difference was observed for work-related nasal symptoms.

One sixth of the study population (n=40, 15%) fulfilled ECRHS criteria for current asthma, although a smaller proportion (n=22, 8%) self-reported a current diagnosis (Table 5). There were no significant differences between self-reported or ECRHS asthma across exposure quartiles. Those in lower exposure quartiles tended to have higher rates of asthma, with 9% of the lowest and 14% of the low exposed versus only 3% of the highest exposed reporting a diagnosis. Similarly, prevalence of current asthma tended to be higher in lower exposure quartiles; prevalence was 16% in the lowest exposed versus 7% in the highest exposed. Nine workers reported recent steroid inhaler use (3%), with no significant differences between exposure groups.

TIgE levels varied widely and were not normally distributed. Geometric mean (GM) TIgE was 35.08 (4.43) kU/L. There was no significant difference between either arithmetic or geometric mean TIgE across exposure groups. Mean rank TIgE was significantly higher in workers with a history of allergy (Mann-Whitney-U mean rank TIgE 132 IU/kL versus 114 IU/kL, p 0.04, Figure 3). Atopy (TIgE >100 IU/L) affected 20% of workers. Rates of SIgE sensitisation were very low, with only one worker having a positive SIgE to soft wood (<1%) at a low level of detection.

Table 5: Respiratory symptoms, asthma, atopy and specific sensitisation among 269 British woodworkers, stratified by exposure quartile. Exposure range for each quartile is reported in parentheses in mg/m^3. Number of workers in each quartile is reported in parentheses.

	Lowest exposure (0-1.19) (n=67)	Low exposure (1.20-2.00) (n=72)	High exposure (2.01-2.32) (n=63)	Highest exposure (2.33-5.44) (n=67)	Total (n=269)
		Symptoms			
Current asthma symptoms, n (%)[a]	34 (50)	34 (47)	27 (46)	26 (39)	123 (46)
Any work-related respiratory symptom, n (%)	11 (16)	8 (11)	8 (13)	2 (3)	29 (11)
Work-related cough, n (%)	7 (10)	4 (6)	2 (3)	1 (2)	14 (5)
Work-related wheeze, n (%)	5 (8)	5 (7)	1 (2)	0 (0)	11 (4)
Work-related chest-tightness, n (%)	2 (3)	2 (3)	3 (5)	1 (2)	8 (3)
Work-related breathlessness, n (%)	4 (6)	3 (4)	3 (5)	0 (0)	10 (4)
Work-related nasal symptoms, n (%)	5 (8)	8 (11)	11 (18)	11 (16)	35 (13)
Work-related ocular symptoms, n (%)	6 (9)	13 (18)	3 (5)	15 (22)*	37 (14)
		Asthma and COPD			
Current inhaled steroid use, n (%)	3 (5)	2 (3)	3 (5)	1 (2)	9 (3)
Self-reported asthma, n (%)[b]	6 (9)	10 (14)	4 (6)	2 (3)	22 (8)
Current asthma, n (%)[c]	11 (16)	16 (22)	8 (13)	5 (7)	40 (15)
Physician diagnosed COPD, n (%)	3 (4)	0 (0)	1 (2)	0 (0)	4 (1)
		Atopy and sensitisation			
AM TIgE, kU/L (SD)	138.72 (305.23)	95.95 (148.18)	236.75 (1223.90)	61.40 (108.90)	133.5 (632.2)
GM TIgE, kU/L (SD)	36.18 (4.96)	37.97 (4.28)	40.23 (4.72)	26.65 (3.76)	35.08 (4.43)
Atopic, n (%)[d]	13 (19)	17 (23)	16 (25)	7 (10)	53 (20)
Positive SIgE to hardwood, n (%)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Positive SIgE to softwood, n (%)	0 (0)	0 (0)	1(1)	0 (0)	1 (1)

*p <0.05, **p <0.01

a= current asthma symptoms defined by wheezing, nocturnal chest tightness, breathlessness on exertion, at rest, or at night, or asthma medication use within the last 12 months as per ECRHS. b= self-reported asthma defined by workers self-reporting a current doctor diagnosis of asthma. c= Current asthma defined by current asthma symptoms plus a past or present diagnosis of asthma. d= atopy defined by total IgE >100kU/L.

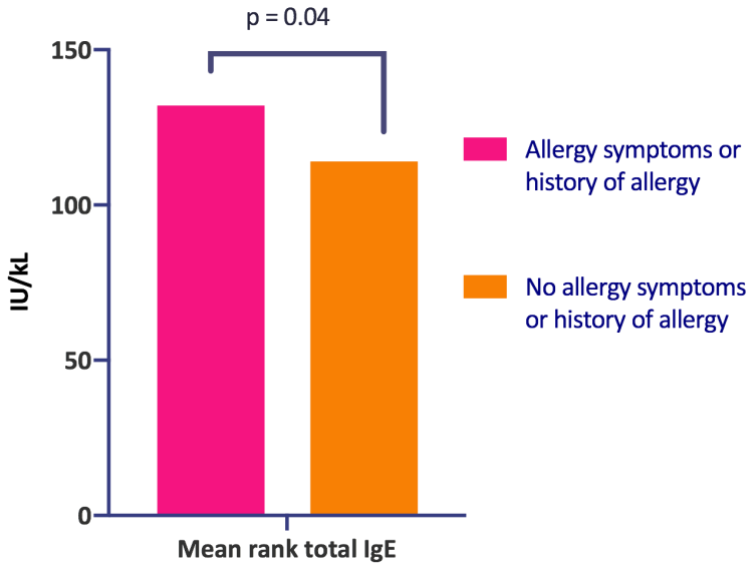

Figure 3: Mean rank total IgE between workers with or without a self-reported history of allergy (eczema, rhinitis, or asthma).

Table six shows FE_{NO} and spirometry data across the four exposure groups. Spirometry met ATS criteria for FEV_1 and FVC in 228 (85%) cases. Of those cases where spirometry was not reproducible, 59% failed to meet FEV_1 criteria and 81% failed to meet FVC criteria. Valid FE_{NO} measurements were obtained in 225 (84%) workers. Reasons for failure to obtain a valid FE_{NO} measurement included inability to perform technically acceptable manoeuvre (59%), inability to perform reproducible manoeuvre within 10 ppb (14%), lack of availability for testing (25%), and testing declined (2%).

FE_{NO} data was not normally distributed. AM FE_{NO} was 27.2 (27.7) ppb, and GM FE_{NO} was 18.4 (2.5) ppb. Median FE_{NO} was 18.71 ppb with 5^{th} and 95^{th} percentiles of 3.50 and 85.44 ppb respectively. When stratified by smoking, median FE_{NO} was 20.17 ppb (5^{th} and 95^{th} percentile 6.00 - 87.62 ppb) for non-smokers, and 10.75 ppb (5^{th} and 95^{th} percentiles 2.08 - 50.25 ppb) for smokers (Mann-Whitney-U, $p < 0.01$, Figure 4). No differences were observed between mean AM or GM FE_{NO} across exposure groups in univariate analyses (table 6). Over a sixth (n=41, 18%) of workers met the BTS/SIGN criteria for a high FE_{NO}, with no difference between exposure quartiles.

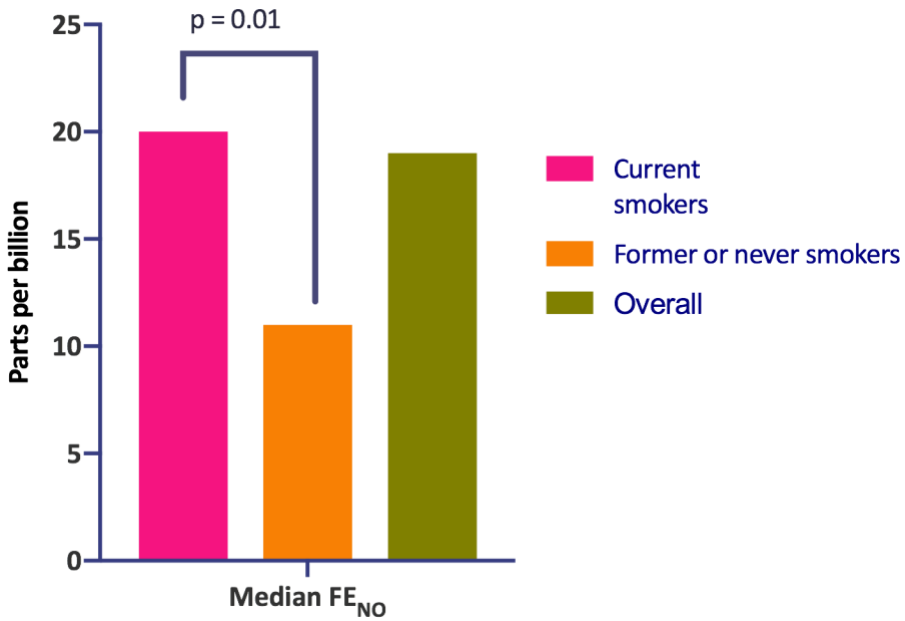

Figure 4: Median FE_{NO} in British woodworkers stratified by smoking status.

Four workers (2%) self-reported a doctor diagnosis of COPD, although 10 (4%) had an FEV_1/FVC ratio less than the LLN (Table 6). Mean percentage predicted FEV_1, FVC and PEF did not vary across exposures. Mean FVC and PEF readings were better than predicted in all exposure groups. There was no significant difference in FEV_1 <LLN, FVC <LLN, PEF <LLN, or FEV_1/FVC <LLN across exposure quartiles.

Table 6: FE_{NO} and lung function among 269 British woodworkers, stratified by exposure quartile. Exposure range for each quartile is reported in parentheses in mg/m^3. Number of workers in each quartile is reported in parentheses.

	Lowest exposure (0-1.19) (n=67)	Low exposure (1.20-2.00) (n=72)	High exposure (2.01-2.32) (n=63)	Highest exposure (2.33-5.44) (n=67)	Total (n=269)
		FE_{NO}			
AM FE_{NO} [a]	24.40 (23.90)	32.45 (32.31)	24.90 (20.29)	26.39 (32.45)	27.2 (27.7)
GM FE_{NO}	18.20 (2.19)	21.38 (2.57)	17.78 (2.45)	16.98 (2.63)	18.4 (2.5)
FE_{NO} >40ppb, n (%)	7(10)	15 (21)	9 (14)	10 (15)	41 (18)
		Spirometry			
Mean % predicted FEV_1 (SD)[b]	99.90 (13.45)	98.61 (12.34)	99.35 (12.25)	101.41 (13.09)	99.8 (12.7)
Mean % predicted FVC (SD)	103.48 (12.52)	101.16 (12.16)	101.63 (12.81)	105.41 (13.59)	102.9 (12.8)
Mean % predicted PEF (SD)	108.16 (16.91)	109.80 (16.63)	107.56 (13.86)	108.27 (16.14)	108.5 (15.9)
Mean FEV_1, mls (SD)	3809 (658)	3697 (720)	3823 (656)	3986 (739)	3824 (699)
Mean FVC, mls (SD)	4825 (818)	4658 (899)	4759 (750)	5027 (905)	4813 (854)
Mean PEF, mls (SD)	584 (116)	587 (98)	581 (95)	588 (102)	588 (102)
FEV_1 <LLN, n (%)	2 (3)	4 (6)	3 (5)	3 (4)	12 (4)
FVC <LLN, n (%)	1 (1)	1 (1)	2 (3)	2 (3)	6 (2)
PEF <LLN, n (%)	1 (1)	3 (4)	2 (3)	4 (6)	10 (4)
FEV1/FVC <LLN, n (%)	4 (6)	3 (4)	1 (2)	2 (3)	10 (4)

*p <0.05, **p <0.01

a=valid FE_{NO} in 225 workers. b=valid spirometry in 228 participants. AM = arithmetic mean. GM = geometric mean. LLN=lower limit of normal.

3.3.2 Respiratory symptoms, allergy and asthma across exposure group

Logistic regression analyses for associations between exposure, symptoms and asthma are presented in table seven. No significant relationships were observed for age, BMI, inhaled steroid use and RPE. Risk of WRRS was significantly lower in the highest versus lowest exposed group (OR 0.16, 95% CI 0.03-0.81, p <0.05). Increasing exposure was not significantly associated with risk of any other work-related respiratory, nasal, or ocular symptom, although there was a tendency for work-related nasal and ocular symptoms to be more common in the highest compared to the lowest exposed group. There was no significant difference in the risk for CAS, self-reported or current

asthma across exposure groups. However, higher exposures tended to decrease the risk for either self-reported or current asthma (Table 7).

Atopy was significantly associated with risk of work-related symptoms and asthma among woodworkers. Even after controlling for confounders, atopic workers were at significantly increased risk of CAS (2.07, 1.07-4.03), self-reported (3.95, 1.45-10.79) and current asthma (4.3, 1.87-9.88). In addition, atopic woodworkers were at significantly increased risk of WRRS (OR 3.31, 1.29-8.48), particularly work-related chest tightness (14.18, 2.28-88.2). Atopy was also associated with a two-fold increased risk of work-related ocular symptoms (2.73, 1.12-6.64), although was not related to an increased risk of work-related nasal symptoms. In contrast, ever smoking was associated with a significantly lower risk of WRRS but increased risk of CAS.

Table 7: Associations between work-related respiratory symptoms, current asthma symptoms, asthma across different quartiles of wood dust exposure. Odds ratios are displayed with 95% confidence intervals in parentheses.

	Any WRRS (n=239)	WR nasal symptoms (n=239)	WR ocular symptoms (n=239)	Current asthma symptoms (n=240)	Self-reported asthma (n=239)	Current asthma (n=239)
Low vs lowest exposure	0.39 (0.12-1.22)	1.19 (0.34-4.17)	1.34 (0.43-4.17)	0.79 (0.37-1.68)	1.90 (0.57-6.37)	1.88 (0.69-5.11)
High vs lowest exposure	0.45 (0.15-1.38)	2.67 (0.86-8.35)	0.39 (0.09-1.73)	0.85 (0.39-1.86)	0.78 (0.19-3.18)	0.75 (0.24-2.33)
Highest vs lowest exposure	0.16 (0.03-0.81)*	2.44 (0.77-7.79)	2.15 (0.68-6.84)	0.83 (0.38-1.82)	0.38 (0.07-2.19)	0.55 (0.16-1.93)
Atopy yes vs no	3.31 (1.29-8.48)*	0.87 (0.33-2.29)	2.73 (1.12-6.64)*	2.07 (1.07-4.03)*	3.95 (1.45-10.79)**	4.30 (1.87-9.88)**
Ever smoker yes vs no	0.34 (0.13-0.87)*	0.77 (0.36-1.68)	1.45 (0.63-3.36)	2.14 (1.22-3.75)**	0.65 (0.24-1.78)	0.92 (0.40-2.07)

*p<0.05

Models also adjusted for age, BMI, inhaled steroid and RPE use, only significant predictors shown. Numbers included in models shown in parentheses: in total, 239 workers had valid atopy, smoking and exposure data.

3.3.3 Airway inflammation and lung function across exposure quartiles

Table eight shows the results of the logistic regression analysis for categorical FE_{NO} and lung function outcomes, stratified by exposure group. Inclusion of inhaled steroid use in models did not influence the outcome. Compared with the lowest exposure group, low exposure to wood dust was associated with an increased risk of significant airway inflammation (OR for FE_{NO} >40ppb 3.43, 95% CI 1.06-11.15). Any exposure was associated with an increased risk of airway inflammation compared with the lowest exposure group, although risk tended to attenuate in the highest exposed (FE_{NO} in highest versus lowest 1.45, 0.40-5.19), and relationships were not significant. Both smoking and atopy had a significant impact on risk of airway inflammation. Current smoking dramatically reduced the risk of high FE_{NO} (0.13, 0.03-0.57). Atopy significantly increased the risk of airway inflammation, with over a doubling of risk of eosinophilic airway inflammation among atopics (2.58, 1.07-6.19).

Table 8: Associations between categorical FE_{NO} and lung function variables with wood dust exposure, atopy and smoking. Odds ratios are displayed with 95% confidence intervals in parentheses.

	FE_{NO} >40 ppb (n=200)[a]	FEV_1 <LLN (n=200)[b]	FVC <LLN (n=200)[b]	PEF <LLN (n=200)[b]	FEV_1/FVC<LLN (n=200)[b]
Low vs lowest	3.43 (1.06-11.15)*	2.02 (0.29-13.93)	-	2.89 (0.22-37.95)	0.95 (0.15-5.95)
High vs lowest	1.28 (0.36-4.55)	1.10 (0.13-9.12)	1.94 (0.16-23.24)	1.54 (0.08-28.91)	0.26 (0.02-2.95)
Highest vs lowest	1.45 (0.40-5.19)	2.84 (0.39-20.45)	1.90 (0.16-22.58)	6.21 (0.52-74.72)	0.94 (0.13-7.01)
Atopy yes vs no	2.58 (1.07-6.19)*	3.55 (0.83-15.29)	0.81 (0.08-8.02)	0.81 (0.08-7.93)	1.47 (0.25-8.85)
Smoking yes vs no	0.13 (0.03-0.57)**	1.83 (0.36-9.37)	1.02 (0.15-6.82)	2.56 (0.38-17.45)	8.25 (0.79-85.95)

*p<0.05.
–: no workers in the low exposure group had an FVC <LLN.
a: FE_{NO} models adjusted for current smoking, atopy, height, age and RPE use. b: Spirometry models adjusted for ever having smoked more than one pack year, atopy, height, age, and RPE use. Only significant predictors shown. Numbers included in models shown in parentheses: in total, 200 workers had valid atopy, smoking FE_{NO} and spirometry data.

No clear relationships were observed between lung function and exposure quartiles in logistic regression models. Higher exposed workers tended to be more likely to have an FEV_1 and PEF <LLN.

There was no trend observed across exposure groups for FEV_1/FVC <LLN. Ever having smoked more than one pack year was non-significantly associated with an increased risk of having lung function <LLN across all categories, but especially for FEV_1/FVC <LLN (8.25, 0.79-85.95).

3.3.4 Examining a healthy worker effect

Relationships between years' exposure to VDGF, respiratory symptoms and asthma are shown in Table nine. Models were adjusted for age, ever smoking, atopy, RPE use, and BMI. Work-related wheeze was associated with a small but significant reduction in years' exposure to VDGF (unstandardised β -0.004 years, p=0.02). A similar trend was seen for work-related cough and WRRS but did not reach significance at the 5% level. No significant relationships between increasing years' VDGF exposure and CAS or current asthma were observed.

Table 9: Linear regression models for cumulative years' exposure to VDGF, respiratory symptoms, and asthma. Unstandardised β coefficients are reported with 95% confidence intervals.

	WRRS	WR cough	WR CT	WR wheeze	WR SOB	WR NS	WR OS	CAS	Current asthma (ERCHS)
Years' exposure to VDGF	-.004 (-0.008 - 0.001)	-0.003 (-0.007 - 0.001)^	-0.001 (-0.004 - 0.001)	-0.004 (-0.007 - -0.001)*	-0.001 (-0.003 - 0.002)	0.004 (-0.002 - 0.009)	0.0002 (-0.005 - 0.005)	-0.005 (-0.013 - 0.002)	-0.002 (-0.007 - 0.004)

*p=0.02, ^p=0.1

VDGF = vapours, dusts, gases, and fumes. WRRS = work-related respiratory symptoms. WR cough = work-related cough. WR CT = work-related chest tightness. WR SOB = work-related shortness of breath. WR NS = work-related nasal symptoms. WR OS = work-related ocular symptoms. CAS = current asthma symptoms.

3.3.5 Indices of asthma across exposure quartile

Relationships between wood dust exposure and composite asthma indices are shown in Table ten. Models were adjusted for age, height, current smoking, RPE use and atopy (where not included in the composite index). Low exposed workers tended to be at increased risk of all composite asthma indices compared with the lowest exposed, with odds ratios of 2.02 (95% CI 0.47-8.74) for CAS+FE_{NO}

>40ppb, and 10.97 (0.79-152.70) for current asthma+FE_{NO} >40ppb respectively. When exposure was dichotomised, high exposure to wood dust (the highest three exposure quartiles) was non-significantly associated with an increased risk of CAS+FE_{NO}, current asthma+atopy and current asthma+FE_{NO} >40ppb. In the high versus low exposed, risk was increased for CAS+FE_{NO} >40ppb (1.86, 0.51-6.78) and for current asthma+FE_{NO} >40ppb (1.47, 0.30-7.27).

Atopic woodworkers were at significantly increased risk for CAS+FE_{NO} >40ppb and current asthma+FE_{NO} >40ppb when compared with non-atopic woodworkers, with OR of 4.10 (1.38-12.19) and 21.21 (3.89-115.56) respectively. Effect sizes for composite indices exceeded those seen between FE_{NO} >40ppb and either atopy or CAS alone. In contrast, current smoking was associated with significantly lower risk of current asthma+FE_{NO} >40 ppb (0.07, 0.01-0.70).

Table 10: Logistic regression models showing relationships between composite asthma indices and exposure quartile. Odds ratios are displayed with 95% confidence intervals in parentheses.

	Current asthma symptoms+atopy (n=269)	Current asthma symptoms+FE_{NO} >40ppb (n=200)	ECRHS current asthma+atopy (n=269)	ECRHS current asthma+FE_{NO} >40ppb (n=200)
Low vs lowest	1.29 (0.47-3.56)	2.02 (0.47-8.74)	3.05 (0.58-16.03)	10.97 (0.79-152.70)
Medium vs lowest	1.32 (0.47-3.67)	2.15 (0.49-9.38)	1.96 (0.34-11.42)	2.55 (0.13-49.60)
High vs lowest	0.34 (0.08-1.36)	1.46 (0.32-6.64)	1.35 (0.21-8.60)	1.53 (0.10-24.53)
Highest 3 quartiles vs lowest quartile	0.96 (0.40-2.31)	1.86 (0.51-6.78)	2.12 (0.45-9.83)	1.47 (0.30-7.27)
Atopy yes vs no	-	4.10 (1.38-12.19)*	-	21.21 (3.89-115.56)**
Smoking yes vs no	0.75 (0.3-1.85)	0.29 (0.07-1.35)	0.38 (0.08-1.74)	0.07 (0.01-0.70)*

*p<0.05, **p<0.01.
Models also adjusted for age, height and RPE use, only significant predictors shown. Numbers included in models shown in parentheses: in total, 200 workers had valid atopy, smoking FE_{NO} and spirometry data.

3.3.6 Linear relationships between wood dust exposure, FE_{NO} and lung function

Table 11 shows data from linear regression models for relationships between inhalable wood dust exposure and FEV_1 in mls, FVC in mls, and GMR FE_{NO}. R squared for all models was significant ($p<0.01$).

Atopy and smoking had a greater impact on GMR FE_{NO} than exposure to wood dust. Smoking was negatively associated with GMR FE_{NO} and atopy was positively associated. Atopic workers had a GMR FE_{NO} 50% higher than non-atopic workers, and smokers had a GMR FE_{NO} 40% lower than never smokers. An increase of 1mg/m^3 in exposure was non-significantly associated with increases in GMR FE_{NO} of 1%. Exposure was not a significant predictor of either FEV_1 or FVC. Both FEV_1 and FVC were positively associated with exposure to wood dust, with increases of 18.31mls and 12.90mls per 1 mg/m^3 increase respectively. Ever having smoked one pack year was negatively associated with FEV_1 (unstandardised β -42.37mls) but positively associated with FVC (unstandardised β 36.67mls). Interestingly, RPE use appeared to have a sizeable effect on both FEV_1 and FVC. Using RPE was associated with a significantly higher FEV_1 (unstandardised β 240.36mls, p <0.01) and FVC (β 167.71mls, p 0.12) compared with not using RPE.

Table 11: Linear regression models for inhalable wood dust exposure and airway inflammation (GMR FE_{NO}), FEV_1 (millilitres) and FVC (millilitres). FE_{NO} is reported as geometric mean ratio data with 95% confidence intervals. For spirometry, unstandardised β coefficients are reported with 95% confidence intervals.

	GMR FE_{NO} (n=200)	FEV_1, mls (n=200)	FVC, mls (n=200)
Current exposure mg/m³	1.01 (0.90-1.15)	18.31 (-58.43 – 95.05)	12.90 (-80.37 – 101.17)
Atopy yes vs no	1.52** (1.13-2.05)	16.35 (-151.23 – 183.94)	30.74 (-172.94 – 211.93)
Smoking yes vs no	0.63 (0.49 – 0.80)**	-42.37 (-186.57 – 101.82)	36.67 (-138.58 – 211.93)
RPE use yes vs no	1.16 (0.88 – 1.55)	240.36** (68.59 – 412.12)	167.71 (-41.06 – 376.47)
R^2; p for model	0.08; <0.01	0.50; <0.01	0.50; <0.01

**p=<0.01
Models adjusted for age, smoking, height, and RPE use. FE_{NO} models additionally adjusted for atopy. Numbers included in models shown in parentheses: in total 200 workers had valid atopy, smoking FE_{NO} and spirometry data.

3.3.7 Relationships between current asthma symptoms, airway inflammation, and airflow obstruction in British woodworkers

Intersections between participants with CAS, airway inflammation (FE_{NO} >40 ppb), and airflow obstruction (FEV_1/FVC <LLN) were explored using contingency analysis. Figure five shows data from 147 workers with either CAS, airway inflammation and/or airflow obstruction. The remaining workers reported no CAS and had no airway inflammation or airflow obstruction.

Two-thirds of those with CAS (n=85, 58% of total) had neither evidence of airway inflammation measured by FE_{NO} nor obstructive spirometry using LLN criteria. Over twice as many workers had CAS and airway inflammation (n=26, 18%) compared with CAS and obstructive spirometry (n=11, 8%). Twenty-three (16%) workers had evidence of airway inflammation but no symptoms of current asthma. There was little overlap between airway inflammation and obstructive lung function. Only one worker had CAS, airway inflammation, and obstructive spirometry with no workers having obstructive spirometry and airway inflammation alone.

Figure six shows intersections between 84 participants with WRRS, airway inflammation and airflow obstruction. Most workers with WRRS had neither airway inflammation nor airflow obstruction. In total, six (7%) workers with WRRS had airway inflammation; the remaining 23 (27%) had no airway inflammation. Even fewer (n=1, 1%) had airflow obstruction. There was little overlap between workers with airflow obstruction and airway inflammation, with only one worker having both airway inflammation and airflow obstruction, and no participants having WRRS, airway inflammation and airflow obstruction.

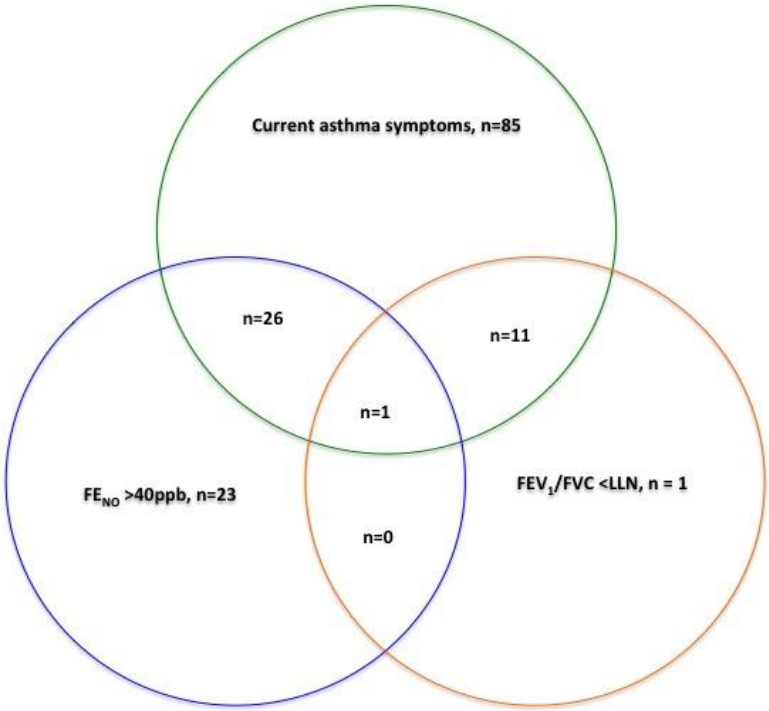

Figure 5: Venn diagram showing intersections between workers with current asthma symptoms, airway inflammation (FE_{NO} >40ppb) and airflow obstruction (FEV_1/FVC <LLN) among 147 study participants

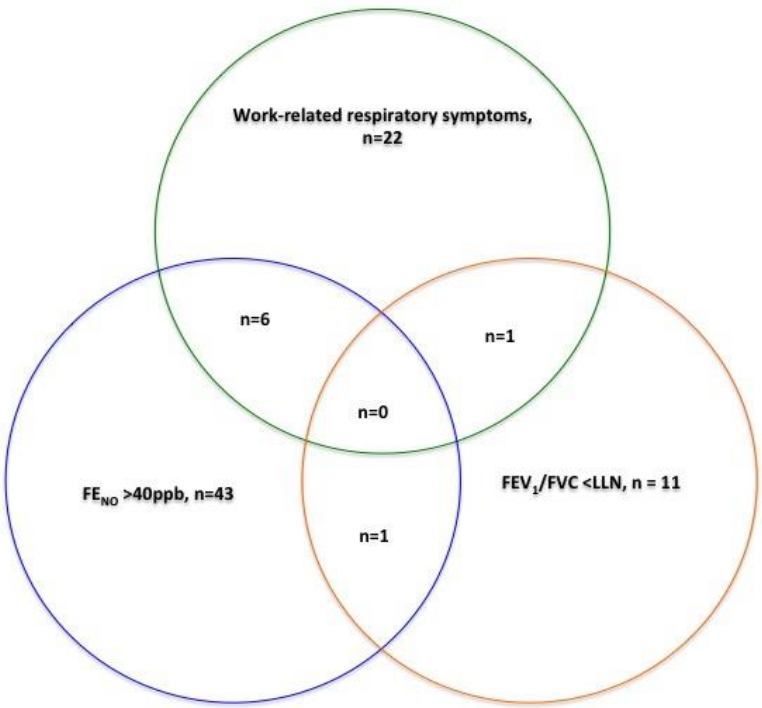

Figure 6: Venn diagram showing intersections between current asthma symptoms, airway inflammation (FE_{NO} >40ppb) and airflow obstruction (FEV_1/FVC<LLN) among 84 study participants.

Figure seven shows asthma diagnoses among the six workers with WRRS and airway inflammation and the 27 workers with CAS and airway inflammation. Only 13 (48%) of symptomatic workers with a FE_{NO} >40ppb had an asthma diagnosis: the figure for symptomatic workers with WRRS was lower (n=4, 66%). However, the presence CAS and airway inflammation was significantly associated with a diagnosis of current asthma: 34% of workers with current asthma had CAS and airway inflammation versus 6% of those without a current asthma diagnosis (Chi squared for difference 27.96, p <0.01).

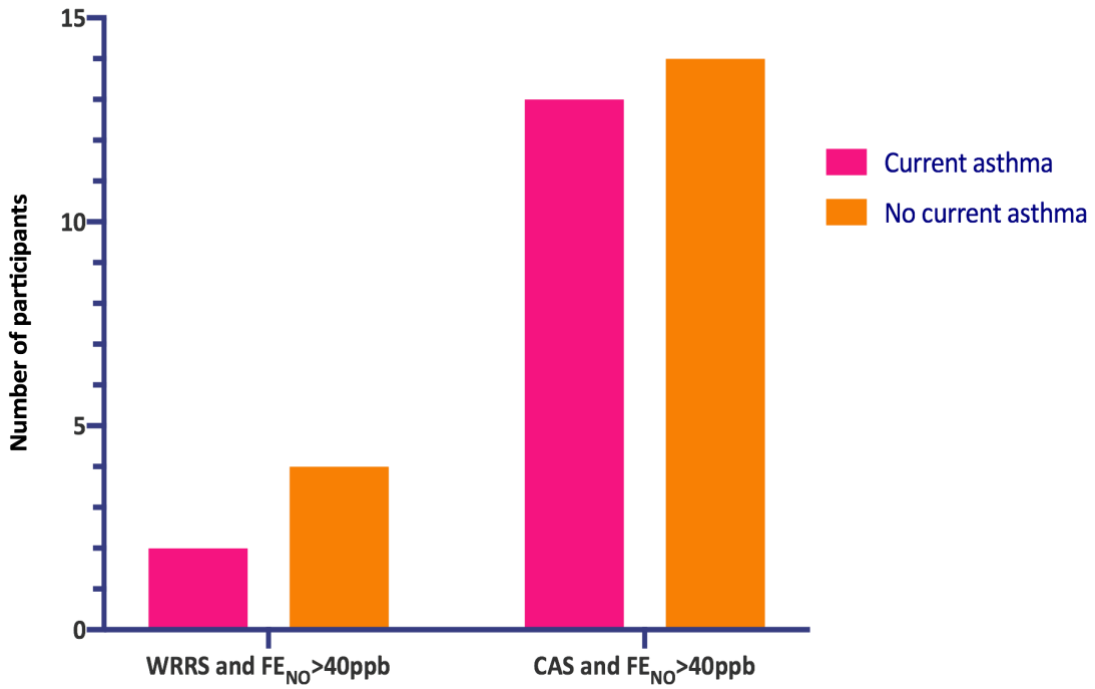

Figure 7: Data for six workers with WRRS and airway inflammation (FE_{NO} >40ppb) and 27 workers with CAS and airway inflammation, split by current asthma diagnosis.

3.4 Discussion

Overall, asthma symptoms were common among woodworkers and were prevalent even at levels of exposure half the current UK WEL. Furthermore, current asthma (defined by ECRHS criteria) was present in one-sixth of the study population, twice the UK prevalence of one in 12 (1). Rates of specific IgE sensitisation were less than 1%. Atopy was a significant modifier of risk for asthma symptoms, WRRS, current asthma, and airway inflammation, even after controlling for confounders.

Furthermore, atopic woodworkers were at significantly increased risk for composite asthma indices including CAS+FE_{NO} >40 ppb and current asthma+FE_{NO} >40 ppb, with effect sizes exceeding those seen for atopic workers with CAS, current asthma, or airway inflammation alone.

Wood dust exposure was associated with an increased risk of asthma symptoms, work-related respiratory symptoms, and asthma, although with no clear dose-response effect. Risk for airway inflammation was significantly higher among the low versus the lowest exposed, although a similar lack of dose-response effect was observed across exposure groups. Similarly, risk for composite asthma indices, including current asthma and airway inflammation, tended to be higher in all but the lowest exposed, although no dose-response was seen. In linear regression models, clear dose-response relationships between current dust exposure, spirometry, or FE_{NO} were not observed.

Set analysis revealed little overlap between airway inflammation and airflow obstruction in symptomatic workers. More workers were identified with CAS and airway inflammation than with CAS and airflow obstruction. The majority of those with respiratory symptoms and airway inflammation did not have an asthma diagnosis, suggesting FE_{NO} may be a useful addition to spirometry in identifying workers at risk of OA.

3.4.1 Limitations

There have been no epidemiological studies of asthma in British woodworkers for over 30 years. Wood dust exposure in the current population was relatively well controlled, with a 95% CI of 1.76-1.99 mg/m^3 and range of 0-5.44 mg/m^3. Only 3% of workers had exposures above the UK WEL of 5 mg/ m^3 and only 17% above half the WEL of 2.5 mg/m^3. As this study was powered to detect a significant difference at levels of exposure at the UK WEL, it may have lacked power to detect significant differences at lower levels of exposure. This is reflected in the finding of positive but not significant effect of exposure on symptoms, FE_{NO} and spirometry.

Cross-sectional studies of this kind are liable to exposure misclassification, as individual exposures vary over time and are influenced by other factors such as workforce, factory size, and health and safety practices (88). Exposure measurements in the current study were made over a single time period and therefore could not capture changes in day-to-day or week-to-week exposure. Exposure misclassification on polytomous scales may dilute effect sizes at higher exposures, and thus lead to error biases towards the null (142). Such non-differential errors may also reduce study power, reducing the ability of a study to find a significant result.

Worksite recruitment may provide another source of bias within this study. As part of this research, worksites underwent a full occupational hygiene assessment. This may expose poor or even illegal practice where workers were placed at unacceptable risk. Since worksites were aware of this hygiene assessment prior to agreeing to take part, they may have avoided participation if they were concerned about exposing poor working practices and subjecting themselves to potential investigation. This is likely to have resulted in 'better' worksites participating in the study, falsely underestimating exposures within the whole population. Thus, the relatively high rates of respiratory symptoms and asthma reported in the current study may be an underestimate of those seen within the wider woodworking industry.

It is possible that some of the respiratory symptoms reported in the current study were caused by irritant, not allergic, disorders. Woodworkers are at risk of exposure to irritant dusts, vapours, gases and fumes, as well as heat, cold, or moisture (52). Incorrectly attributing irritant symptoms to wood dust exposures may explain the absence of an exposure-response relationship observed between increasing wood dust exposure and respiratory symptoms. Furthermore, woodworkers may be exposed to respiratory allergens other than wood dust including isocyanates, formaldehyde, and epoxy resins (50). Exposure to allergens other than wood dust may lead to an increase in symptom

prevalence and an overestimation of work-related or respiratory symptoms. As respiratory symptoms and WRRS were less frequent than previously reported in a UK study, overestimation of symptoms in the current study is unlikely (104).

Skin prick tests were not performed in the current study. As such, a total IgE above 100 IU/l was used to identify atopic workers. Since atopy had a significant modifying effect on risk of respiratory symptoms, asthma, and airway inflammation, misclassifying atopic workers could have a significant impact on the study results. An IgE over 100 IU/l IgE has been shown to have good positive predictive value but only modest negative predictive value for at least one positive allergy test in individuals with asthma symptoms (138). Compared to skin-prick tests, total IgE correlates better with the total allergic component of asthma and is more easily comparable between populations (143). Furthermore, there is evidence that total IgE may be useful in predicting atopy in individuals where a specific IgE for an inhalant is negative (144). Since rates of SIgE were low among the population, it is unlikely that this interfered with TIgE results.

3.4.2 Respiratory symptoms are common among woodworkers even at low exposures

Our observed mean wood dust exposures of 1.88 (1.76 – 1.99) mg/m^3 were substantially lower than the 5mg/m^3 WEL currently set out by HSE (24). Furthermore, although the data range was 5.44 mg/m^3, the 25% to 75% range lay close to the mean with 25% of exposure measurements exceeding the 2.33 mg/m^3 75th percentile. Overall dust exposures were significantly lower than reported in the most recent UK hygiene data. In 2009, Galea *et al.* reported mean exposures of 5.2 mg/m^3 among their study population (25). In their national survey of British woodworking sites, Black *et al.* reported 50% of workers were occupationally exposed to levels of wood dust higher than 2mg/m^3 (52). Despite lower exposures, symptoms, asthma, and airway inflammation were still prevalent in the current study, with rates comparing to recent reviews on asthma in woodworkers (82, 117). This

suggests that symptoms and airway inflammation are still present at levels of wood dust exposure substantially lower than the current UK WEL.

3.4.3 Work-related symptoms in British woodworkers

One-in-ten workers in the current study reported WRRS. This prevalence is lower than has been reported in other studies where levels of exposure to wood dust were comparable (89, 91). Furthermore, we found workers in the highest exposure quartile to be at significantly reduced risk of WRRS versus those in the lowest exposure quartile (OR 0.16, 95% CI 0.03-0.81, p <0.05). Factors that may influence a reduction in symptom prevalence and exposure-response effect include an overall reduction in wood-dust exposure (25), increasing use of RPE (145), or relocation of symptomatic workers away from high exposures through either health surveillance or exit from the workplace (146). In addition, there is evidence to support underreporting of work-related symptoms among exposed workers. In their study of bakers, Gordon *et al.* found a proportion of workers who reported no work-related symptoms but were subsequently found to have OA (30). In their qualitative study of workers with OA, Bradshaw *et al.* described asthmatic workers reluctant to divulge symptoms to employees, citing fear of loss of earnings and job security as key explanatory factors (16). Underreporting of symptoms may explain the discrepancy between the lower rates of work-related symptoms in the current study and the recent national increase in cases of wood dust OA (27).

Work-related nasal and ocular symptoms were more prevalent than WRRS in the current study and showed a stronger relationship with wood dust exposure. Prevalence was similar to a study of carpenters exposed to a variety of hard and soft woods, where Campo *et al.* reported nasal symptoms in 40% (147). Risk of work-related nasal and ocular symptoms was twice as high in the highest versus lowest exposure quartiles. Rhinoconjunctivitis may precede the onset of OA in up to 50% of workers, and work-related upper airway symptoms are an important risk factor for developing disease (11). Higher rates of nasal and ocular symptoms in the current study may point

to individuals at risk of developing OA who should undergo enhanced health surveillance. Longitudinal studies are required to identify the proportion of such workers who go on to develop allergic airways disease.

3.4.4 Asthma symptoms and evidence for a dose-response effect

This study reflects findings elsewhere of an increased risk of asthma symptoms in woodworkers, with conflicting evidence for a dose-response effect (91, 92, 97, 107). Asthma symptoms were common among the current study population, with almost half of all workers reporting CAS. Prevalence of CAS did not differ across exposure groups, suggesting additional drivers for respiratory symptoms than wood dust exposure alone. As well as wood dust, workers in the current study were exposed to glues, solvents, paints, and resins, and a small number also worked with isocyanates. These substances are both irritant and allergenic and may explain the high rates of respiratory symptoms across exposure groups and the lack of a dose-dependent relationship. In addition, workers in the lowest exposure quartile were more likely to perform cleaning and maintenance tasks. Cleaning and maintenance are associated with higher single time-point exposures, but a lower overall exposure, compared with other job tasks. Previous research has suggested that intensity of exposure to wood dust may be as important as overall exposure when assessing risk for asthma, and individuals with multiple high-dose exposures are at increased asthma risk even if overall exposures are lower (148). Longitudinal studies are needed to understand how the nature of exposure modifies future asthma risk among woodworkers.

Woodworkers have been previously been found to be at significantly increased risk of asthma symptoms compared to non-exposed controls (79, 107). However, evidence for increasing risk of asthma symptoms across exposure groups has been conflicting, particularly where wood dust exposure is low. Female, but not male, woodworkers have been previously reported to be at increased risk for chronic bronchitis at higher wood dust exposures (92). Since female

representation in the current study was very low (3%), analysis on the effects of gender was not possible. When considered alone, no clear dose-response relationship between wood dust exposure and CAS was found. However, when FE_{NO} was included in analysis, risk ratios became more positive, albeit non-significantly so. This suggests an effect of exposure on asthma symptoms that is more pronounced in the presence of eosinophilic airway inflammation.

Two studies have found paradoxical relationships between wood dust exposure and CAS, with higher symptom risk in lower exposed groups (91, 97). The respective authors of these studies suggested variation in dust exposure and RPE use as potential reasons for the inverse dose-response effect. Overall levels of RPE use were high in the study population, with between 65% and 88% of participants saying they used RPE. Indeed, workers wearing RPE had significantly better FEV_1 and FVC than those without. This could explain the relative reduction in symptoms observed among the current population.

The lack of a clear dose-response relationship between wood dust exposure, symptoms and asthma may additionally be explained by a healthy worker effect (HWE). The HWE has been well described as a source of confounding among occupational epidemiological studies, particularly cross-sectional studies, where symptomatic or 'unhealthy' workers may be pre-selected out of higher exposure jobs or be relocated or removed from high exposure jobs as a result of ill-health (146, 149). In the current study, workers with WR wheeze had small but significantly less time exposed to VDGF, and similar trends were seen for WRRS and WR cough, supporting earlier symptom onset in workers at risk for occupational asthma (146). There was a non-significant trend towards younger age and fewer years in woodworking among workers in the highest exposure group in the current study. This may reflect recent entry to, or premature exit from, the workplace, providing further evidence for a possible HWE. Overall, workers had spent an average of eight years exposed to wood dust with a further 19 years within the woodworking industry. Although OA can occur after many years exposure to an

occupational allergen, it is more common in the first years of exposure (11). The long tenure of the workers in the current study is further evidence for a HWE, where susceptible workers with fewer years' exposure may have become symptomatic and exited the workplace prematurely. Cohort studies are required to further examine the HWE phenomenon among woodworkers, especially at lower exposure levels.

3.4.5 Atopy modifies disease risk in woodworkers but specific sensitisation to wood dust is uncommon

Atopic woodworkers were at significantly increased risk of WRRS, CAS, asthma, and airway inflammation in the current study. Risk ratios for CAS were 2.07 (95% CI 1.07 – 4.03), for WRRS 3.31 (1.29 – 8.48), and for asthma 4.30 (1.87 – 9.88) in atopic versus non-atopic workers. In addition, atopic workers were at an increased risk of significant eosinophilic airway inflammation using BTS/SIGN criteria: risk ratio for FE_{NO} >40ppb was 2.58 (1.07-6.19) in atopic versus non-atopic workers. In contrast, atopy did not increase the risk for airflow obstruction, or modify relationships between exposure and FEV_1 or FVC in linear regression models.

The modifying effect of atopy in asthma risk among woodworkers has been described previously. In the current study workers were exposed to a variety of wood species, with the majority using a mixture of hard and soft woods. Atopic workers exposed to pine have been found to be at increased risk for asthma symptoms and airway hyperresponsiveness across exposure groups when compared with non-atopic workers (100). In contrast, studies in WRC asthma have suggested no modifying effect of atopy (150). However, although atopy increases the risk of OA, no direct causal link for OA among atopic individuals has been established (9). Prevalence of atopy in industrialised nations may be as high as 40%, with false positive rates as high as 20%, making pre-employment screening problematic for all workers (151). Only a small proportion of atopic workers will go on to develop disease, and there is currently no way to identify such individuals. As with any screening programme,

there must be benefit to the individual, and those benefits must be balanced with costs to society (35). Since pre-employment screening on the basis of atopy is unlikely to offer advantages to individual workers and is likely to be costly and possibly misleading, pre-selection of workers away from woodworking on the basis of atopic status should not be recommended. Further studies exploring gene-environment interactions in the causation of OA will help to define risk factors for developing disease and may help to inform surveillance for susceptible individuals at high risk of occupational diseases.

Rates of SIgE sensitisation in the current study were less than 1%. Low IgE sensitisation rates have been reported among woodworkers, suggesting that sensitisation among woodworkers is not measurable with SIgE, that tests are not accurate enough, or that disease occurs through a non-IgE mechanism (77, 152). It is possible the sensitivity of the panel tests used was reduced as workers were exposed to a number of different wood species in varying quantities. However, even where bespoke tests to a specific allergen have been used in patients with confirmed OA, specific sensitisation rates have been low suggesting at least some patients do not raise an IgE response (109). Where positive SIgE results are present they may be helpful in supporting a diagnosis of wood dust OA, but the current findings suggest that negative tests are not helpful in identifying woodworkers at risk of, or suffering from, OA.

3.4.6 Eosinophilic airway inflammation in woodworkers

Overall, airway inflammation was common among the study population, with almost a fifth of workers having a FE_{NO} above 40ppb, the cut-point suggested by BTS/SIGN for significant eosinophilic airway inflammation (4). In the current study, mean FE_{NO} was 27.59 ppb (95% CI 24.18 – 31.00 ppb), and median FE_{NO} was 18.71 ppb with a 95^{th} percentile of 85.44 ppb. In comparison, Torén *et. al.* reported a lower median FE_{NO} 18.2 ppb with a 95^{th} percentile of 41.3 ppb from their population-based study of European adults (153). Furthermore, the ATS/ERS guideline suggests a FE_{NO} above

25ppb is 'predictive of eosinophilic airway inflammation, and that predictive values for FE_{NO} are higher than spirometry and peak flow and similar to bronchial challenge testing' (46). Eighty (36%) of our study population had a FE_{NO} above 25 ppb, suggesting a significant number of workers with airway inflammation, even accounting for a false positive rate of 20% (154). The higher rates of eosinophilic airway inflammation observed in woodworkers may be a consequence of exposure to wood dust, may represent early allergic airways disease, or be a combination of the two. Only one previous study has examined FE_{NO} in WRC workers with OA, finding no relationship with positive SIC (72). However, this is the first workplace study to examine FE_{NO} in a population of exposed woodworkers. Better understanding of the role of FE_{NO} in exposed, but not necessarily diseased, woodworkers will help to determine its use as a diagnostic and screening tool for OA in future.

Interestingly, over half the workers with symptoms (either WRRS or CAS) and airway inflammation had no asthma diagnosis, suggesting a number of workers may have had undiagnosed airways disease. NICE have previously reported levels above 40 ppb as having a sensitivity of 78.6 - 88.3% and specificity of 82.6 - 89.5% for predicting asthma in symptomatic individuals (48). Furthermore, among symptomatic workers FE_{NO} provided additional information than spirometry alone. More symptomatic workers had airway inflammation than airflow obstruction (18% versus 8%), and there was very little overlap between airway inflammation and airflow obstruction suggesting the two tests provide information on distinct groups. Although some workers may have had falsely positive FE_{NO}, some may also have had early allergic airway disease. FE_{NO} has been demonstrated to identify at-risk groups of occupationally exposed apprentices, even when spirometry is normal (41, 124). The development of obstructive lung function is a late sign in OA and, when present, may be irreversible (14). Identifying individuals with OA early in their disease process is vital to ensuring good health outcomes for affected workers (11). This finding suggests there may be individuals identified by high FE_{NO} who are not picked up by spirometry and supports further exploration of FE_{NO} as a tool for identifying OA in wood-dust exposed workers.

Few studies have examined whether FE_{NO} is related to increasing exposure in workplace settings. In woodworkers, exposure was associated with an increased risk of eosinophilic airway inflammation, albeit without a clear dose-response. Risk ratios for FE_{NO} >40ppb were significantly raised in the low versus lowest exposed (3.43, 1.06-11.15), with positive, but non-significant effects seen in higher exposure groups. Since rates of both current and ever smoking, age, and atopy were not significantly different between exposure quartiles, this difference may be further explained by the HWE. Consensus is lacking to show FE_{NO} is useful in occupational settings (155). However, the current study demonstrates FE_{NO} to be as reproducible as spirometry with a similar number of people able to perform the required manoeuvre. Cohort studies are needed in order to examine how FE_{NO} relates to allergen exposures over time, and better understand its utility in a programme of health surveillance.

3.4.7 Lung function in British woodworkers

No significant differences were found in lung function across exposure groups. Although post-bronchodilator spirometry was not performed, the 4% prevalence of obstructive lung disease (FEV_1/FVC <LLN) reported here is lower than the European prevalence range of 9-26% recently reported by Blanco *et al* (156). In contrast, rates of current smoking among woodworkers were 26%, comparable to the recent national average for men (157). The current study population was younger than those included in the COPD prevalence studies, which may account for the lower rates of airflow obstruction. Interestingly, positive associations with wood dust exposure were found for both FEV_1 and FVC, reflecting findings elsewhere (106, 107). The reduced prevalence of airflow obstruction and positive associations with FEV_1 and wood dust exposure is likely to be a further example of a healthy worker effect.

The most significant effect on lung function in the current study was the use of RPE, an effect even more remarkable considering the relatively low exposures recorded. Workers using RPE had an FEV_1 240mls higher than those who did not (unstandardised β=240.36mls, 95% CI 68.59-412.12). Properly used RPE is an important part of the hierarchy of controls, and can help to reduce individual exposures after appropriate risk assessment and elimination has taken place (23). Conversely, RPE may not be available in workplaces with poorer health and safety practices; workers may not use it due to lack of education about its importance or beliefs about its effects; or it may not be tolerated in people with pre-existing respiratory disease where symptoms make its use more difficult (158). Further studies are needed to explore in more detail how and why RPE is used in workplaces, its impact on personal exposure, and its influence on long-term lung function decline in woodworkers.

3.4.8 Summary

Current asthma symptoms are common among woodworkers at levels of wood dust exposure significantly lower than the current UK WEL, raising the possibility of an allergic, rather than irritant, mechanism. Among woodworkers, atopy was the most significant predictor of current asthma symptoms, asthma, and airway inflammation. Rates of SIgE sensitisation to wood dust were very low, suggesting specific sensitisation in woodworkers is not measurable by routinely available IgE tests, and allergy in such populations may not be IgE mediated. Eosinophilic airway inflammation was common among the study population and provided additional information on asthma risk to spirometry alone in symptomatic workers. Workers in higher exposure categories tended to be at increased risk of airway inflammation and asthma, although a clear dose-response was not observed. A significant proportion of symptomatic workers with airway inflammation reported no asthma diagnosis, suggesting a possible role for the measurement of eosinophilic airway inflammation in health surveillance. Further exploration of the utility of FE_{NO} in LMW exposure environments is required to better understand how eosinophilic airway inflammation relates to exposure, respiratory symptoms and lung function in these settings.

4 Respiratory symptoms, airway inflammation, and asthma in British foundry workers

4.1 Background

4.1.1 Definition and history of the foundry process

The casting of metal items dates back thousands of years. The earliest records of copper castings date from ancient Mesopotamia in the period between 4000 and 3000 B.C (159). The addition of tin

to create bronze alloys enabled ancient civilisations to cast metals for a wide variety of objects including weapons, jewellery and ornaments.

Figure 8: Bronze age spear tip mould, France. Photograph by Rama, Wikimedia Commons, Cc-by-sa-2.0-fr [CeCILL (http://www.cecill.info/licences/Licence_CeCILL_V2-en.html) or CC BY-SA 2.0 fr (https://creativecommons.org/licenses/by-sa/2.0/fr/deed.en)], from Wikimedia Commons

Iron castings were first produced in China around 1000 B.C., and by the 1500s iron-smelting foundries were common. The foundry industry was rapidly industrialised in the 19th Century following the invention of the Bessemer process in Sheffield, which revolutionised the oxidization of pig iron and allowed mass production of steel for the first time (160). Today the foundry industry employs around 17,000 people in the UK, with an annual turnover of £2.2 billion (161).

Most foundries employ a similar process for casting metal items (Figure 9) (162). Pattern making is the first part of the process: patterns for items to be cast are made from wood, metal, wax, or plastics. Subsequently, moulds and cores are made. Sand (or greensand) was historically used for moulding, but increasingly new moulding materials such as urethane or phenol binder systems are employed due to their ease of use and reuse and their improved casting results. A wide variety of metals are used for melting and pouring. Castings are then 'knocked-out' of their moulds and cooled

and mould materials are reclaimed. Finally, finishing processes including fettling, shot blasting, and welding may be employed before castings are cleaned and coated ready for use. The foundry industry serves sectors including transport, energy, medical, defence, and aerospace, involving a variety of ferrous and non-ferrous foundry processes and employing a wide range of moulding methods.

Figure 9: Process map showing the main stages of the foundry process

4.1.2 Exposure to respiratory hazards and asthma risk in foundry work

Foundry work may involve exposure to a number of respiratory allergens (Figure 10). Urethane resin moulding systems can release isocyanates, most commonly methyldiisocyanate (MDI), during thermal degradation (163). Isocyanates are among the commonest causes of OA in many industrialised countries including Great Britain (26), France (164), Belgium (20), and Canada (21). Reported annual incidence in these studies varies between 6 and 15% of all cases of OA. Sensitisation to a number of volatile isocyanate species including toluene diisocyanate (TDI), hexamethylene diisocyanate (HDI), and MDI, has been widely described (165).

Figure 10: Infographic showing the main parts of the foundry process associated with exposure to respiratory hazards. Photographs used with permission from the Health and Safety Executive British Survey of Foundry workers 2017.

Alkali-phenolic binder systems containing a phenol-formaldehyde resin are also used in foundries, leading to formaldehyde release during the heating process (166). Formaldehyde has been identified as an occupational asthmagen in a number of settings including healthcare work (167), manufacture of wooden products (168), plastics (169), and clothing (170). Older furan moulds or cores used furfuryl alcohol (FFA), which has also been shown to be a respiratory sensitiser (171). Exposure to allergenic metals such as nickel sulphate, hexavalent chromium, and cobalt may also occur during parts of the finishing process such as shot-blasting or welding, and metal particulate may be present in ferrous or non-ferrous foundry particulate (162, 172).

Though isocyanates and formaldehyde are both well-documented in their association with asthma, studies in foundry workers are limited in number. Foundry workers with at least one WRRS have

been found to have higher rates of histamine reactivity compared with non-exposed workers (173). In foundry workers exposed to MDI, workers with higher MDI exposures were more likely to have asthma symptoms and had a 50ml cross-shift decrease in FEV_1 compared to non-exposed workers (174). More recently foundry workers have been found to be at increased risk of OA (defined by an onset of asthma symptoms after first foundry exposure, work-related symptoms, and PEF variability of at least 20%) versus non-exposed workers (175). Data from cohort studies are scarce: a single study of foundry workers, conducted in Scandinavia at lower levels of allergen exposure, demonstrated a high prevalence of nasal and ocular symptoms but no significant difference in cross-shift lung function between the two exposure groups (176).

Additional malignant and non-malignant respiratory diseases are reported in excess in foundry workers, who are exposed to hazards other than respiratory allergens. Respirable crystalline silica (RCS) exposure is common in foundries, and silica-related disease such as COPD, silicosis, lung cancer and tuberculosis is more prevalent in foundry workers (177-179). Cadmium exposure has also been reported in foundry workers leading to an increased risk of COPD (180). Lung cancer is more prevalent among foundry workers, even after accounting for smoking, attributed to silica, polyaromatic hydrocarbon and other carcinogen exposure (181). Additional irritant symptoms may be caused by benzene, toluene, carbon monoxide, foundry particulate and fume, as well as extreme heat or dryness (162).

4.1.3 Fractional exhaled nitric oxide in foundry workers

FE_{NO} is increasingly used in the diagnosis and treatment of asthma (48). Its use is attractive in occupational settings as an adjunct to spirometry in health surveillance, where it could provide additional information to lung function in at-risk workers and help to diagnose workers with asthma earlier in their disease process (124). Some studies have shown an increase in airway inflammation following exposure to isocyanates (182, 183). Even fewer studies have reported eosinophilic airway

inflammation in association with formaldehyde exposure. A case-report of occupational eosinophilic bronchitis due to formaldehyde has been published (184), but case-reports for OA due to formaldehyde have been less strongly associated with eosinophilic airway inflammation (185, 186). Fewer population-based studies looking at airway inflammation in isocyanate or formaldehyde workers have been published. One study has reported increasing exposure to foundry dust is associated with increasing airway inflammation (187). Only one paper has demonstrated an increase in FE_{NO} in children following higher domestic exposure to formaldehyde (188).

Although foundry workers may be exposed to a number of well-documented sensitisers such as isocyanates and formaldehyde, few studies have previously considered the relationships between FE_{NO} and allergen exposure in foundry populations. In addition, recent data on respiratory health in foundry workers is lacking, with few studies exploring the relationship between foundry exposures and asthma symptoms. This study aims to evaluate the prevalence of respiratory symptoms, airway inflammation, asthma, and abnormal lung function in foundry workers, and to understand relationships between these key variables and allergen exposure.

This study was reviewed and approved by the NHS Health Research Authority National Research Ethics Committee: North West – Greater Manchester East (REC reference 12/NW/0048).

4.2 Methods

Data for foundry workers was collected by CWH between 2011 and 2014. As part of the longitudinal project and by way of contribution to the data described here, I: constructed a database; coded, cleaned and refined the data; co-developed a job-exposure matrix (JEM); designed and undertook an analysis plan; and conducted further recruitment visits to foundries. This represents the first analysis of this data, which has not yet been reported elsewhere.

4.2.1 Worker recruitment

British foundries were recruited to a longitudinal study through the Cast Metals Federation (CMF). CMF is the main industry body for the UK foundry industry and represents a large group of both ferrous and non-ferrous foundries. An occupational hygienist from HSE worked with the CMF to identify foundry sites employing processes with a high potential for exposure to respiratory hazards. These exposure scenarios included:

- whether the foundry was ferrous or non-ferrous (specifically with exposure to iron, steel, or aluminium);
- the binder system used to create cores and moulds (greensand, furan, urethane, or alkali phenolic);
- and whether the foundry was jobbing or automated (162).

The study was publicised to all attendees at a national health and safety meeting conducted by CMF and HSE. Interested worksites were given information regarding the study and followed up with a telephone call from the study team (Appendix C). Prior to data collection, the study was explained in detail to managers, and sites received a recruitment visit by one of the study team to directly discuss the research with employees.

4.2.2 Power calculation

A Monte Carlo-based power calculation was performed as part of the longitudinal Health SRP examining lung function decline in foundry workers. Individual three-year exposures were simulated using a random effects model based on data from Casting Technologies International published in an HSE report in 2009 (189). This is a large dataset with over 9000 measurements from 245 foundries: mean respirable dust exposure in the dataset was 1.7 mg/m^3, with a between-company SD of 0.58 (log scale) and a within-company SD of 0.95 (log scale). Assuming a normal FEV_1 range of 100 ± 20% predicted and a within person SD for spirometry of 100ml (190), a cross-sectional sample of 400 and a longitudinal sample of 300 people was estimated to be able to detect a loss of 10.5 ml/yr per

mg/m^3 increase in respirable dust exposure with 80% power using a two-way significance of 5% (130). Similar losses in FEV_1 have been observed in construction and wood workers at risk of asthma (108, 190).

4.2.3 Study population

All exposed workers over the age of 16 employed in any part of the foundry process were invited to take part. This included workers with indirect exposures such as maintenance or cleaning. Visits were conducted during all working shifts to ensure the maximum number of workers were able to participate. Workers were excluded if they worked entirely outside the foundry process in an unexposed job (for example, in a wholly office-based role).

4.2.4 Respiratory questionnaire

Each worker was administered an interviewer-led questionnaire, including detailed information about current and previous jobs, change of jobs due to vapours, dusts, gases, or fumes (VDGF) exposure, and use of RPE as previously described in section 3.2.4 (Appendix C). Self-reported current and past respiratory illnesses such as asthma, COPD, silicosis, and lung cancer were also documented.

4.2.5 FE_{NO} and spirometry measurement

All workers were invited to undergo FE_{NO} and spirometry measurement, as described previously in section 3.2.5, according to ATS/ERS guidelines (46, 133).

4.2.6 Exposure assessment of respiratory sensitisers

A detailed assessment of foundry exposures was undertaken by HSE occupational hygienists. Individual methods of measurement and exposures across surveyed sites are reported in detail elsewhere (162). In brief, respirable and inhalable ferrous foundry particulate or, in non-ferrous foundries, foundry dust (hereafter referred to as foundry particulate or FP); respirable crystalline

silica (RCS); volatile organic compounds (VOCs) including FFA and formaldehyde; isocyanates; metal dust and fume; and toluene dyes; were measured in surveyed foundries. Air sampling was carried out in accordance with the MDHS 14/4 (134). In addition, a full assessment of foundry processes, including the use of local exhaust ventilation (LEV) and personal protective equipment (PPE) was also undertaken.

Potential respiratory allergens were identified based on publications of respiratory allergy in foundry workers. Isocyanates, formaldehyde, FFA, chromium VI (CrVI), cobalt, and nickel sulphate were all considered as possible allergens (191). FFA, nickel sulphate, cobalt and CrVI were either not utilised in recruited foundries or had levels below the limit of detection, so were not considered significant allergens in the current study (162). As such, isocyanates and formaldehyde were treated as the main foundry allergens of interest in the current study.

4.2.7 Foundry-specific job-exposure matrix

A foundry-specific job-exposure matrix (JEM) was created alongside an HSE statistician and an experienced HSE occupational scientist with an interest in foundry work. The JEM incorporated:

- current exposure to bonding systems containing isocyanate, formaldehyde, or both, as determined by the hygiene assessment at each worksite;
- current foundry process undertaken by each individual worker as reported within the health study and evaluated by the hygienist;
- empirical airborne exposures measurements made at each participating site by HSE occupational hygienists.

Participants worked in one of 12 foundry processes (Table 12). These processes were grouped into four common work areas, based on the stage of the foundry process (Figure 9):

1. moulding (including mould or core making and sand reclamation);
2. casting (furnace work; melting and pouring of metals);

3. finishing (including knockout, fettling, shot blasting and any subsequent welding);
4. general foundry work (including housekeeping processes such as cleaning and maintenance).

Table 12: List of 12 foundry processes evaluated in the current study, grouped by common work area.

Moulding	Casting	Finishing	General foundry work*
Moulding	Casting	Knockout	Foundry work
Core making	Furnace work: ferrous metals	Fettling	Foundry work: ferrous metals
	Furnace work: non-ferrous metals	Finishing	Foundry work: non-ferrous metals
			Pattern making
* Foundry work included cleaning, general maintenance, labouring and other ancillary jobs.			

Weighted exposure estimations were used to assign individual exposures to each of the study participants (88). Eight-hour TWA measurements for isocyanates and formaldehyde captured data from the 12 foundry tasks undertaken by participants (Table 12). Data were not normally distributed so were log-transformed for further analysis. For both formaldehyde and isocyanate exposure, task-specific averages were estimated based on the mean task-specific 8-hr TWA across the eight sites. Similarly, site-specific averages were estimated based on the mean site average across all tasks at each of the eight participating worksites. Individual exposures were then estimated for both isocyanate and formaldehyde using the product of the task-specific average and the site-specific average and expressed as a ratio of overall mean exposures. Where the number of workers performing a particular task at an individual site was less than five, exposures were assigned using the mean exposure for that particular task taken across all work sites.

Lowest, middle, and highest exposure categories were determined *a priori* for the study population by stratification of isocyanate and formaldehyde exposure into tertiles. First, formaldehyde exposures were ranked, with the lowest tertile being lowest exposed, middle tertile medium exposed, and highest tertile highest exposed. Individuals who were not exposed to formaldehyde (in other words where formaldehyde was not used at that particular worksite) were assigned missing values. The same process was repeated for isocyanate exposure, with workers not exposed to

isocyanates assigned missing values. A four-by-four contingency table was used to establish tertiles of exposure for the whole population (Figure 11). Workers with low-low or low-missing exposures were designated the lowest exposed. Workers with medium-low, medium-medium, or medium-missing exposures became the medium exposure category. Workers with high-medium, high-high, or high-missing exposures were included in the highest exposure category.

		Formaldehyde exposure			
		Low	**Medium**	**High**	**Missing**
Isocyanate exposure	**Low**	*Lowest*	*Medium*	*Highest*	*Lowest*
	Medium	*Medium*	*Medium*	*Highest*	*Medium*
	High	*Highest*	*Highest*	*Highest*	*Highest*
	Missing	*Lowest*	*Medium*	*Highest*	

Figure 11: Four-by-four contingency table used to establish an overall exposure category for the 351 foundry workers in the study. Workers were exposed either to formaldehyde, isocyanates, or both.

4.2.8 Definitions

Respiratory symptoms evaluated included cough, wheeze, breathlessness, and chest tightness. Work-related nasal or ocular symptoms, WRRS, CAS, ever, and current asthma were defined as previously described in section 3.2.9. Atopy was defined as a self-reported history of eczema, hay fever, or asthma. Data previously reported in section 3.3.2 demonstrated a significantly higher TIgE in woodworkers with a history of allergy defined by self-report. High FE_{NO} was defined as a FE_{NO} above 40 parts per billion (ppb) and LLN criteria were used for abnormal spirometry, as previously described in section 3.2.9.

4.2.9 Statistical analysis

Demographic data was presented as means ± SD where data was continuous, and as numbers with percentages for categorical data. Univariate analyses were conducted across the three exposure tertiles for demographic, exposure, and health data, using independent t-tests, ANOVA and Chi-

squared testing. Non-normally distributed data were analysed using Mann-Whitney U testing. Where FE_{NO} and spirometry values did not meet reproducibility criteria they were excluded from analysis. FE_{NO} data were not normally distributed so expressed as both arithmetic and geometric means.

Multiple logistic regression was used to evaluate associations between exposure to respiratory sensitisers (formaldehyde and isocyanates) and the six dependent variables of interest: any WRRS; CAS; current asthma; FE_{NO} >40 ppb; FEV_1 <LLN, FVC <LLN; and FEV_1/FVC <LLN, as per section 3.2.10. The lowest exposure category was used as the reference group. Models were adjusted for age, sex, smoking, RPE use, height, and atopy (for FE_{NO} models). Respirable FP was included as an obligate confounder in models as a function of overall dust exposure across worksites (162).

Linear regression modelling was used to examine relationships between exposure to sensitisers, FE_{NO}, and spirometry. Since FE_{NO} data were not normally distributed, data were log-transformed and back transformed into GMR FE_{NO}, as described in section 3.2.10. Ratios for FE_{NO} were reported separately per 1 $\mu g/m^3$ in isocyanate exposure and per 1mg/m^3 increase in formaldehyde exposure, in accordance with the standard units of measurement in the UK (24). Models were adjusted for age, smoking, sex, height, RPE use, atopy (for FE_{NO}), and respirable FP.

Linear regression models were constructed to explore a potential healthy worker effect among foundry workers, as described in section 3.2.10. Years of exposure to VDGF were used as the dependent variable to account for movement in jobs within and between foundries. Data were adjusted for age, ever smoking, atopy, RPE, FP, and height.

Relationships between workers with CAS or WRRS, FE_{NO} >40ppb and airflow obstruction (FEV_1/FVC <LLN) were explored using intersecting Venn diagrams as described in section 3.2.10. Only significant

variables were reported. A *p* value of less than 0.05 was considered statistically significant. All statistical analyses were performed using IBM SPSS Statistics v23 (IBM, New York, 2015).

4.3 Results

4.3.1 Study population

Eight foundries agreed to take part in the study, employing a total of 510 workers. Three-hundred-and-fifty-one workers participated, with a participation rate of 69%. Reasons for non-participation included individual workers declining, unavailability due to shift work, and failure to attend the study appointment. No demographic data was available on those who declined to participate.

Seven foundries were ferrous with only one non-ferrous foundry included. Exposure to formaldehyde binders was more common among the study population. In total, 309 workers were potentially exposed to formaldehyde binder systems and 186 were potentially exposed to isocyanate binder systems (Table 13). Three foundries (including a total of 165 workers) used only alkali-phenolic (formaldehyde containing) binders, two foundries (42 workers) used only urethane (isocyanate) binders, and a further three foundries (144 workers) used both binder systems.

Table 13: Number of workers exposed to either NCO-based, formaldehyde-based, or both NCO and formaldehyde-based binder systems by employment in either ferrous or non-ferrous foundries

	Isocyanate binder only	Formaldehyde binder only	Both systems used
Ferrous foundry	Foundry A: n=29 Foundry B: n=13	Foundry E: n=64 Foundry H: n=93	Foundry C: n=38 Foundry D: n=83 Foundry F: n=23
Non-ferrous foundry		Foundry G: n=8	
Total	**N=42**	**N=165**	**N=144**

4.3.2 Allergen exposure in foundry workers

Mean exposure to formaldehyde across the study population was 0.14 (0.11) mg/m^3, median exposure was 0.10 mg/m^3 with a 5th and 95th centile of 0.02 and 0.16 mg/m^3 respectively (Table 14). Mean exposure to isocyanates was 1.61 (1.44) microgrammes/m^3 (µg/m^3), median exposure was 1.31 µg/m^3, with a 5th and 95th centile of 1.10 and 1.52 µg/m^3 respectively.

Following exposure stratification, 76 people were in the lowest exposure tertile, 124 in the medium exposure tertile, and 151 in the highest exposure tertile (Table 14). Mean exposures for formaldehyde and isocyanates were, respectively: 0.05 mg/m^3 and 0.12 µg/m^3 in the lowest exposed; 0.09 mg/m^3 and 0.70 µg/m^3 in the medium exposed; and 0.24 mg/m^3 and 2.90 µg/m^3 for the highest exposed. For formaldehyde-exposed workers, mean exposures were almost double in the medium compared to the lowest exposure group, and exposures in the highest exposure group were five times those in the lowest exposure group.

For isocyanate-exposed workers, mean exposures were six-times higher in the medium compared to the lowest exposures, and highest exposures were 24 times the lowest exposures. All exposures were an order of magnitude below the current UK WEL of 2.5 mg/m^3 for formaldehyde and 20 µg/m^3 for isocyanates: maximum formaldehyde exposure reached one-fifth of the WEL and maximum isocyanate exposure reached one-third of the WEL. Respirable FP exposures increased across tertiles of formaldehyde exposure but were lower in the highest exposure tertile for isocyanate exposure. All respirable FP exposures were lower than the current WEL of 4mg/m^3.

Table 14: Range of formaldehyde and isocyanate exposures in the lowest, medium, and highest exposure groups, with respirable FP exposure in each tertile. The current UK 8-hr TWA WEL for isocyanates is 20 µg/m^3, and for formaldehyde is 2.5 mg/m^3. Respirable FP was measured in mg/m^3.

	Formaldehyde exposed, mg/m^3 (SD)	Isocyanate exposed, µg/m^3 (SD)

	N in group	Mean exposure	Exposure range	FP	N in group	Mean exposure	Exposure range	FP
Lowest tertile (n=76)	89	0.05 (0.02)	0-0.07	1.22 (1.48)	61	0.12 (0.05)	0-0.18	1.20 (1.74)
Medium tertile (n=124)	129	0.09 (0.03)	0.071 - 0.15	1.91 (2.76)	63	0.70 (0.54)	0.19-2.24	1.33 (2.59)
Highest tertile (n=151)	91	0.24 (0.121)	0.16-0.56	3.32 (5.99)	62	2.90 (0.94)	2.25-6.36	0.46 (0.49)
Total (n=351)	**309**	0.14 (0.11)			**186**	1.61 (1.44)		

4.3.3 Population demographics

Table 15 shows the demographic characteristics of the study population across exposure tertiles. Of the 351 participants, 99% were men with a mean age of 42.3 (SD 12.4) years and 8.9 (9.9) years working in their current role. There were no differences in age, smoking history, sex, and job duration across the three exposure categories. Workers in the highest exposure category were more likely to use RPE than those in the medium exposure category and had worked longer in the foundry industry compared to those in lower exposure groups. As expected, exposure to formaldehyde and isocyanates increased significantly across the groups.

Table 15: Study characteristics of 351 British foundry workers, stratified by exposure tertile

	Lowest exposure, n=76	Medium exposure, n=124	Highest exposure, n=151	Total, n=351
		Demographics		
Male, n (%)	76 (100)	124 (100)	150 (99)	350 (99)
Current smokers, n (%)	24 (32)	33 (27)	39 (26)	96 (27)
Ever smoked more than one pack year, n (%)	40 (53)	60 (48)	78 (52)	178 (51)
Ethnicity, Caucasian (%)	73 (96)	121 (98)	148 (98)	342 (97)
Age, years (SD)	42.76 (11.48)	40.99 (12.64)	43.04 (12.69)	42.3 (12.4)
BMI, kg/m^2 (SD)	28.86 (4.38)	28.16 (4.42)	28.10 (4.50)	28.29 (4.44)
Height, cm (SD)	176.61 (5.94)	175.77 (6.15)	176.55 (6.74)	176.29 (6.36)

	Exposure			
Moulding, n (%)	13 (4)	31 (9)	56 (16)	100 (28)
Casting, n (%)	7 (2)	5 (1)	5 (1)	17 (5)
Finishing, n (%)	35 (10)	37 (11)	28 (8)	100 (28)
General foundry work, n (%)	21 (6)	51 (15)	62 (18)	134 (38)
Wears RPE, n (%)	60 (79)	81 (65)	124** (82)	265 (76)
Time spent in current job, years (SD)	9.45 (10.54)	8.56 (9.63)	8.86 (9.78)	8.9 (9.9)
Total time worked in foundry, years (SD)	12.33 (11.83)	14.00 (12.19)	16.64 (13.25)*	14.76 (12.66)
Ever left job due to health, n (%)	0 (0)	2 (2)	1 (1)	3 (1)
Mean exposure to formaldehyde, mg/m³ (SD)	0.05 (0.02)	0.09 (0.03)	0.24 (0.11)**	0.14 (0.11)
Mean exposure to isocyanate, µg/m³ (SD)	0.12 (0.05)	0.70 (0.54)	2.91 (0.94)**	1.61 (1.44)
Mean exposure to respirable FP, mg/m³ (SD)	1.20 (1.55)	2.21 (2.99)	2.18 (4.86)**	1.98 (3.73)

*p <0.05, **p <0.01.
RPE = respiratory protective equipment. Lowest exposure group = those with low/low or low/missing exposures from JEM. Medium exposure group = medium/medium or medium/missing exposures in JEM. Highest exposure group = high/high or high/missing exposures in JEM.

Respiratory symptoms, WRRS, CAS, asthma, and atopy across exposure tertiles are presented in Table 16. Over a third of workers (40%) reported at least one respiratory symptom. CAS were also common, reported by 37%. Prevalence of respiratory symptoms across the whole population varied between 12% (chest tightness without cold) to 27% (wheeze). A third (35%) of the study population were atopic. Nineteen (5%) of the study population were taking an inhaled corticosteroid, with no significant difference across exposure groups. A self-reported diagnosis of asthma was reported by 14%, with current asthma by ECRHS criteria less prevalent at 9%. Six workers (2%) self-reported a diagnosis of COPD. No workers reported a diagnosis of silicosis.

Table 16: Respiratory symptoms, work-related respiratory symptoms, atopy, and asthma across the three exposure tertiles in 351 British foundry workers

	Lowest exposure, n=76	Medium exposure, n=124	Highest exposure, n=151	Total, n=351
Atopy, respiratory symptoms and asthma				
Atopy, n (%)	26 (34)	54 (44)	43 (28)*	123 (35)
Cough, n (%)	20 (26)	31 (25)	29 (19)	80 (23)
Chest tightness without cold, n (%)	15 (20)	14 (11)	14 (9)	43 (12)
Wheezing without colds, n (%)	13 (17)	24 (19)	36 (24)	73 (21)
Shortness of breath, n (%)	17 (22)	22 (18)	17 (11)	56 (16)
More than one respiratory symptom, n (%)	30 (39)	55 (44)	56 (37)	141 (40)
Ever diagnosed with asthma, n (%)	12 (16)	15 (12)	23 (15)	50 (14)
Current steroid inhaler use, n (%)	4 (5)	7 (6)	8 (5)	19 (5)
Current asthma, n (%)	7 (9)	11 (9)	14 (9)	32 (9)
Current asthma symptoms, n (%)	33 (43)	48 (39)	47 (31)	128 (37)
Work-related respiratory symptoms				
Changed job due to breathing problems, n (%)	0 (0)	2 (2)	1 (1)	3 (1)
Work-related cough, n (%)	14 (18)	15 (12)	16 (11)	45 (13)
Work-related chest tightness, n (%)	2 (3)	4 (3)	7 (5)	28 (8)
Work-related wheeze, n (%)	7 (9)	11 (9)	9 (6)	27 (8)
Work-related breathlessness, n (%)	0 (0)	0 (0)	1 (1)	1 (1)
Work-related eye irritation, n (%)	14 (18)	17 (14)	20 (13)	51 (15)
Work-related nasal irritation, n (%)	9 (12)	16 (13)	31 (21)	56 (16)
WRRS, n (%)	17 (22)	26 (21)	26 (17)	69 (20)
More than one work-related eye or nasal symptom, n (%)	19 (25)	23 (19)	41 (27)	83 (24)

*p<0.05, **p<0.01.
Atopy = history of allergy (rhinitis, eczema or asthma). Lowest exposure group = those with low/low or low/missing exposures from JEM. Medium exposure group = medium/medium or medium/missing exposures in JEM. Highest exposure group = high/high or high/missing exposures in JEM.

Prevalence of individual WRRS varied from 1% (work-related breathlessness) to 13% (work-related cough). Work-related chest tightness and work-related wheeze were reported by 8% respectively. Overall, one-fifth (n=69) reported at least one WRRS. Work-related nasal or ocular symptoms were reported by 24%. Less than 1% (n=3) of workers reported ever having to change their job due to breathing problems.

Workers in the highest exposure group were significantly less likely to be atopic than those in the medium exposure group (28% versus 44%, p=<0.05, Table 16). Respiratory symptoms including cough, chest tightness without cold, wheeze, and breathlessness were all more common among the lowest exposed group, but not significantly so. There was no significant difference between any work-related symptom, CAS, or asthma between exposure groups.

FE_{NO} and spirometry data for the study population are presented in Table 17. Valid FE_{NO} readings were obtained in 297 workers (85%). One-quarter (n=75) of the study population had a FE_{NO} above 40ppb. Valid spirometry was performed in 313 (89%) participants. Mean percent predicted FEV_1, FVC, and PEF were 98.58 (13.71), 103.46 (12.29), and 108.42 (17.37) respectively. LLN criteria for FEV_1 were met in 6% of workers, for FVC in 2%, and for FEV_1/FVC in 7%.

GM FE_{NO} was significantly higher in the medium exposure group compared to the highest exposure group (25.70 versus 16.98 ppb, p<0.01). Workers in the medium exposure group were significantly more likely to have a FE_{NO} above 40 ppb compared to other groups (34% versus 20% in the lowest and 12% in the highest group, p <0.01). Absolute and percent predicted FEV_1, FVC, and PEF did not differ significantly between groups. However, medium exposed participants were significantly more likely to have an FEV_1 <LLN versus the lowest exposed.

Table 17: FE_{NO} and spirometry across the three exposure tertiles in 351 British foundry workers

	Lowest exposure, n=76	Medium exposure, n=124	Highest exposure, n=151	Total, n=351
	Fractional exhaled nitric oxide [a]			
AM FE_{NO}, ppb (SD)	28.7 (30.64)	34.36 (24.30)*	24.25 (21.93)	29.04 (25.39)
GM FE_{NO}, ppb (SD)	17.78 (2.81)	25.70 (2.29)**	16.98 (2.51)	20.42 (2.51)
FE_{NO} above 40ppb, n (%)	15 (20)	42 (34)**	18 (12)	75 (25)

Spirometry[b]				
Mean FEV_1, mls (SD)	3715.8 (680.77)	3749.8 (751.92)	3849.9 (680.49)	3787.20 (705.75)
Mean FVC, mls (SD)	4892.5 (804.65)	4823.4 (794.26)	4856.6 (800.83)	4852.50 (797.17)
Mean PEF, mls (SD)	570.74 (94.74)	591.86 (108.37)	593.38 (93.23)	592.30 (98.75)
Mean % predicted FEV_1 (SD)	96.49 (12.18)	96.81 (15.95)	100.95 (12.13)	98.58 (13.71)
Mean % predicted FVC (SD)	103.97 (12.60)	102.39 (13.44)	104.06 (11.19)	103.46 (12.29)
Mean % predicted PEF (SD)	108.20 (17.76)	107.63 (18.99)	109.14 (15.89)	108.42 (17.37)
FEV_1 < LLN, n (%)	4 (6)	12 (11)*	3 (2)	19 (6)
FVC < LLN, n (%)	1 (2)	4 (4)	1 (1)	6 (2)
FEV_1/FVC < LLN, n (%)	6 (9)	11 (10)	4 (3)	21 (7)

*p<0.05, **p<0.01. a: Valid FE_{NO} obtained in 297 (85%) workers. b: Valid spirometry obtained in 313 (89%) workers. Lowest exposure group = those with low/low or low/missing exposures from JEM. Medium exposure group = medium/medium or medium/missing exposures in JEM. Highest exposure group = high/high or high/missing exposures in JEM.

Mean arithmetic mean (AM) FE_{NO} was 29.04 (25.39), with mean geometric mean (GM) FE_{NO} 20.42 (2.51). Median FE_{NO} was 22.0 ppb and was significantly lower in current versus ex- or never smokers (Figure 12). Median FE_{NO} in current smokers was 12.50 ppb (5^{th} and 95^{th} centile 1.47 and 72.90 ppb respectively) versus 25.67 ppb (5^{th} and 95^{th} centile 6.27 and 83.47 ppb) in ex- or never smokers (Mann-Whitney-U for difference <0.01).

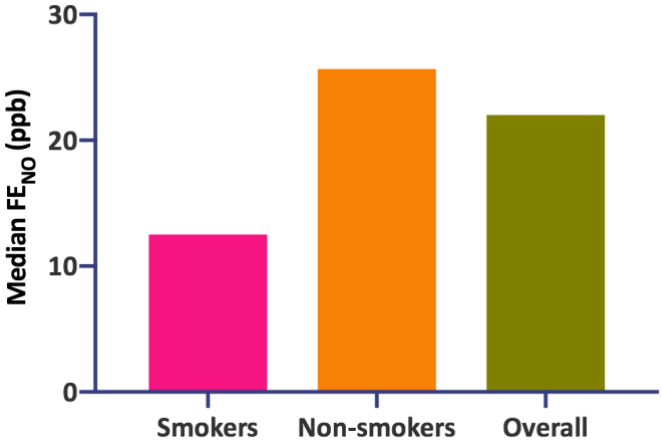

Figure 12: Median FE_{NO} across the study population, stratified by current versus former or never smokers.

4.3.4 Respiratory symptoms, work-related respiratory symptoms, and asthma across exposure groups

Table 18 shows data from logistic regression models for respiratory symptoms, WRRS, CAS, and asthma. There was no clear increase in risk for any respiratory symptom or WRRS across exposure tertiles. Workers in the highest exposure tertile were at increased risk of work-related nasal symptoms, though this was not significant (OR 2.13, 95% CI 0.93 – 4.87, p 0.07). Ever smoking was associated with an increased risk of any respiratory symptom and any upper airway symptom (OR for any respiratory symptom 2.28, 1.46 – 3.57, p=0.01 and for any upper airway symptoms 1.66, 1.05-2.6, p=0.03). Ever smoking was also significantly associated with an increased risk of WRRS (OR 2.04, 1.17-3.55, p=0.01). Atopic workers were twice as likely to report any upper respiratory symptom than non-atopic workers (2.05, 1.27-3.31, p=0.01).

There was a tendency for workers in the highest exposure category to have a lower risk for CAS, although this did not reach statistical significance (0.59, 0.33-1.06, p=0.08). Ever smokers were twice as likely to report CAS than those who had never smoked (OR 2.22, 1.4-3.51). Risk for either self-reported or current asthma did not vary significantly across exposure tertiles. In contrast, atopy significantly increased the risk for both self-reported (OR 4.31, 2.22-8.36, p <0.01) and current

asthma (3.56, 1.62-7.85, p <0.01) compared to non-atopic workers. Respirable FP had no effect on any model.

Table 18: Logistic regression models for respiratory symptoms, work-related respiratory symptoms, current asthma symptoms, and asthma stratified by exposure tertile. Odds ratios are displayed with 95% confidence intervals in parentheses.

	Any respiratory symptom (n=345)	Any upper airway symptom (n=345)	WR ocular symptoms (n=345)	WR nasal symptoms (n=345)	WRRS (n=345)	CAS (n=345)	Self-reported asthma (n=345)	Current asthma (n=345)
Medium vs lowest	1.27 (0.69 – 2.33)	0.57 (0.31 – 1.08)	0.69 (0.31 – 1.53)	0.98 (0.41 – 2.54)	0.91 (0.45 – 1.89)	0.87 (0.47 – 1.60)	0.57 (0.23 – 1.38)	0.71 (0.24 – 2.07)
Highest vs lowest	0.97 (0.54 – 1.74)	1.19 (0.67 – 2.13)	0.73 (0.34 – 1.55)	2.13 (0.93 – 4.87)^	0.76 (0.38 – 1.53)	0.59 (0.33 – 1.06)#	1.04 (0.46 – 2.32)	0.98 (0.36 – 2.67)
Atopy yes vs no	1.13 (0.71 – 1.81)	2.05 (1.27 – 3.31)**	1.33 (0.71 – 2.46)	1.54 (0.82 – 2.90)	0.8 (0.45 – 1.44)	1.15 (0.71 – 1.86)	4.31 (2.22 – 8.36)**	3.56 (1.62 – 7.85)**
Ever smoked >1 Pack year yes vs no	2.28 (1.46 – 3.57)**	1.66 (1.05 – 2.60)*	1.11 (0.61 – 2.02)	1.72 (0.93 – 3.19)#	2.04 (1.17 – 3.55)	2.22 (1.4 – 3.51)**	0.83 (0.44 – 1.58)	0.7 (0.4 – 1.88)
Respirable FP (mg/m³)	0.98 (0.92 – 1.04)	0.94 (0.88 – 1.01)	0.96 (0.88 – 1.07)	0.89 (0.78 – 1.02)	1.00 (0.93 – 1.08)	1.00 (0.94 – 1.06)	0.99 (0.91 – 1.09)	1.01 (0.92 – 1.12)

*p<0.05 **p<0.01 ^p=0.07 #p=0.08.
WRRS = work-related respiratory symptoms. CAS = current asthma symptoms.
Models also adjusted for age, BMI and RPE use, only significant predictors shown. Numbers included in models shown in parentheses: in total, 345 workers had valid atopy, smoking and exposure data.

4.3.5 Airway inflammation and spirometry across exposure groups

Table 19 shows risk ratios for airway inflammation and spirometry across exposure groups. Models including FE_{NO} were adjusted for current smoking, atopy, age, sex, and RPE use: the addition of ICS use to models did not affect the outcome. Medium exposure was associated with an increased risk of FE_{NO} >40ppb (OR 1.82, 0.87 – 3.82) but this was not statistically significant (Table 19). However, increasing exposure to respirable FP was significantly associated with FE_{NO} >40ppb (OR 1.09, 1.01-1.18, p=0.04). Current smokers were at reduced risk of airway inflammation (0.45, 0.22-0.90, p<0.05), whereas risk was increased in atopics (OR 1.70, 0.95 – 3.04, p=0.07).

Table 19: Odds ratios for categorical FE_{NO} and spirometry outcomes of interest, stratified by exposure tertile.

Risk ratios and 95% confidence intervals are reported.

	FE_{NO} >40ppb (n=297)[a]	FEV_1 <LLN (n=313)[b]	FVC <LLN (n=313)	FEV_1/FVC <LLN (n=313)
Medium vs lowest	1.82 (0.87 – 3.82)	2.15 (0.63 – 7.41)	1.38 (0.13 – 15.06)	1.48 (0.48 – 4.56)
Highest vs lowest	0.63 (0.28 – 1.39)	0.32 (0.07 – 1.55)	0.22 (0.01 – 6.15)	0.28 (0.07 – 1.08)^
Atopy yes vs no	1.71 (0.95 – 3.04)^	1.76 (0.65 – 4.76)	2.68 (0.43 – 16.56	1.71 (0.65 – 4.55)
Smoking yes vs no	0.45 (0.22 – 0.90)*	1.01 (0.38 – 2.70)	-	1.4 (0.53 – 3.74)
Respirable FFP (mg/m³)	1.09 (1.01 – 1.19)*	1.01 (0.89 – 1.16)	1.13 (0.93 – 1.38)	0.87 (0.69 – 1.11)

*p=0.05, ^ p = 0.07

a: Model adjusted for age, current smoking, height, RPE use, atopy, and sex.

b: Model adjusted for age, ever having smoked more than one pack year, RPE use, atopy, height and sex.

- No data generated for FVC due to small numbers in models.

Associations between FEV_1<LLN and exposure disappeared in adjusted models (Table 19). No significant observations were seen for any of the spirometric outcomes considered and either exposure, atopy, smoking, or respirable FP. There was a tendency for medium exposure to be associated with an increased risk of low FEV_1, FVC, and FEV_1/FVC. In contrast, exposure in the highest group tended to be associated with a lower risk of abnormal spirometry. Atopy tended to be associated with an increased risk of abnormal spirometry. Clear associations between smoking and abnormal spirometry were not observed, and no data was generated for FVC <LLN due to small numbers of smokers in the model.

4.3.6 Evidence of a healthy worker effect among foundry workers

Table 20 shows relationships between respiratory symptoms and asthma and years' exposure to VDGF among British foundry workers. A current asthma diagnosis was associated with increasing years' exposure to VDGF among foundry workers (unstandardised β 0.003, p=0.05). Similarly, work-related cough and wheeze were associated with increasing years' exposure to VDGF. In contrast, work-related breathlessness was associated with fewer years' exposure to VDGF.

Table 20: Linear regression models for cumulative years' exposure to VDGF, respiratory symptoms, and asthma among British foundry workers. Unstandardised β coefficients are reported with 95% confidence intervals.

	WR cough	WR CT	WR wheeze	WR SOB	WR NS	WR OS	WRRS	CAS	Current asthma (ERCHS)
Years' exposure to VDGF	0.003 (-0.001-0.007)^	0.001 (-0.002-0.004)	0.003 (-0.001-0.005)^	-0.001 (-0.002 - 0.001)#	0.001 (-0.003 - 0.005)	-0.001 (-0.005 - 0.003)	0.003 (-0.002 - 0.007)	0.004 (-0.001 - 0.010)	0.003 (-0.001 - 0.007)*

*p=0.05, ^p=0.09, #p=0.08

VDGF = vapours, dusts, gases, and fumes. WRRS = work-related respiratory symptoms. WR cough = work-related cough. WR CT = work-related chest tightness. WR SOB = work-related shortness of breath. WR NS = work-related nasal symptoms. WR OS = work-related ocular symptoms. CAS = current asthma symptoms.

4.3.7 Associations between continuous measurements of exposure, airway inflammation and lung function

Multiple linear regression models were constructed to examine relationships between exposure to either isocyanates or formaldehyde and GMR FE_{NO}, percent predicted FEV_1, percent predicted FVC, and percent predicted PEF (Tables 21 and 22). All models were significant although explained only around 5% of variance within the outcome variable (R^2 varied between 0.03 and 0.11). Increasing isocyanate exposure was positively associated with GMR FE_{NO} (Table 21). For each 1µg/m^3 increase in isocyanate exposure, there was a 11% increase in GMR FE_{NO} (GMR 1.11, 95% CI 1.01 – 1.21, $p<0.05$). In contrast, increasing exposure to formaldehyde was not significantly associated with GMR FE_{NO} (0.89, 0.34 – 2.33, p=0.81, Table 22). Increasing respirable FP exposure was associated with small but significant increases in GMR FE_{NO} in formaldehyde-exposed workers. Each 1mg/m^3 increase in respirable FP related to a 3% increase in GMR FE_{NO} (1.03, 1.00 – 1.06, $p<0.01$) in workers co-exposed to formaldehyde. Similar trends were observed in isocyanate models but were not significant. In linear models, smoking was negatively associated with GMR FE_{NO} for both formaldehyde and isocyanate exposed workers, whereas no significant association was observed for atopic workers.

Table 21: Linear regression models showing the association between current exposure to isocyanates with GMR FE_{NO} and % predicted FEV1, FVC, PEF and FEV1/FVC. FE_{NO} is reported as geometric mean ratio data with 95% confidence intervals. For spirometry, unstandardised β coefficients are reported with 95% confidence intervals.

	GMR FE_{NO}[a]	%FEV_1[b]	%FVC[b]	%PEF[b]
Current exposure to isocyanates, (µg/m³)	1.11 (1.01 – 1.21)*	1.77 (0.31-3.23)*	0.65 (-0.72-2.02)	1.93 (1.22-3.73)
Atopy, yes vs no	1.04 (0.80 - 1.37)			
Smoking, yes vs no	0.68 (0.53 – 0.89)**	-1.99 (-6.16 – 2.18)	0.75 (-3.17-4.67)	-2.74 (-7.90-2.42)
Respirable FFP (mg/m³)	1.06 (0.99 – 1.14)	-0.21 (-1.49-1.07)	-0.67 (-1.88-0.53)	1.28 (-0.30-2.86)
R2; p for model	0.11; <0.01	0.03; 0.09	-0.01; 0.65	0.03; 0.05

[a] Regression coefficient showing relationship between log10 FE_{NO} and current exposure to isocyanates. Models were adjusted for age, ever smoking, RPE use, atopic symptoms, and sex. Only reproducible FE_{NO} data was included. FE_{NO} data is back-transformed.
[b] Regression coefficients showing relationship between spirometric outcomes of interest and current exposure to isocyanates. Models adjusted for age, ever smoking, RPE use, sex, and height.

Table 22: Linear regression models showing the association between current exposure to formaldehyde with GMR FE_{NO} and % predicted FEV1, FVC, PEF and FEV1/FVC. FE_{NO} is reported as geometric mean ratio data with 95% confidence intervals. For spirometry, unstandardised β coefficients are reported with 95% confidence intervals.

	FE_{NO}[a]	%FEV_{1b}	%FVC[b]	%PEF[b]
Current exposure to formaldehyde (mg/m³)	0.89 (0.34 – 2.33)	12.05 (-2.80-26.89)	-0.29 (-13.36-13.36)	-2.14 (-21.26-16.97)
Atopic, yes vs no	1.12 (0.99 – 1.40)			
Smoking, yes vs no	0.73 (0.59 – 0.90)**	0.56 (-2.79-3.86)	2.17 (-0.87-5.20)	-0.75 (-5.0-3.51)
Respirable FFP (mg/m³)	1.03 (1.00 – 1.06)**	0.38 (-0.06-0.82)^	0.35 (-.05-0.75)^	0.25 (-0.311-0.82)
R2; p for model	0.07; < 0.01	0.02; 0.06	0.01; 0.28	0.01; 0.25

[a] Regression coefficient showing relationship between log10 FE_{NO} and current exposure to isocyanates. Models were adjusted for age, ever smoking, RPE use, atopic symptoms, and sex. Only reproducible FE_{NO} data was included. FE_{NO} data is back-transformed.
[b] Regression coefficients showing relationship between spirometric outcomes of interest and current exposure to isocyanates. Models adjusted for age, ever smoking, RPE use, sex, and height.

Isocyanate exposure was associated with small but significant increases in FEV_1 and PEF (Table 21). For each 1µg/m³ isocyanate rise there was a 1.77% increase in percent predicted FEV_1 (1.77, 0.31-

3.23, p=0.02). Percent predicted PEF increased by 1.93% with each 1µg/m^3 increase in isocyanates (1.93, 1.22-3.73, p=0.04). No trend was seen for FVC. In formaldehyde models, no significant relationships between exposure and the six spirometric outcomes of interest were observed.

Stratified analyses were performed to evaluate whether exposure-response effects were more evident in workers with WRRS or current asthma. Stratification by WRRS or current asthma made no difference to relationships between FE_{NO} and exposure to either isocyanates or formaldehyde, and models were not significant.

4.3.8 Relationships between current asthma symptoms, high FE_{NO} and abnormal spirometry among the study population

In order to understand the relationships between airway inflammation (FE_{NO} >40ppb) and other clinical features of work-related asthma (CAS or WRRS and airflow obstruction), contingency analyses were performed. Figure 13 shows intersections between CAS, airway inflammation and airflow obstruction among 187 foundry workers. Workers without CAS, airway inflammation or airway obstruction were excluded. Of the 128 workers with CAS, 37 (19% of total) also had airway inflammation whereas only 12 (6%) had airflow obstruction. There was little overlap between airway inflammation and airflow obstruction among symptomatic workers: only five symptomatic workers (3%) had both airway inflammation and airflow obstruction. A further two workers had both airway inflammation and airflow obstruction but reported no CAS.

Figure 14 shows intersections between 153 participants with WRRS, airway inflammation and airflow obstruction. A quarter of workers (n=17, 11%) with WRRS had significant airway inflammation. Only three workers (2%) with WRRS had airflow obstruction. Again, little overlap was observed between airway inflammation and airflow obstruction among symptomatic workers: of the seven workers with both airway inflammation and airflow obstruction, only one (1%) had WRRS.

Thus, there were 6 workers with both airflow obstruction and airway inflammation who reported no WRRS.

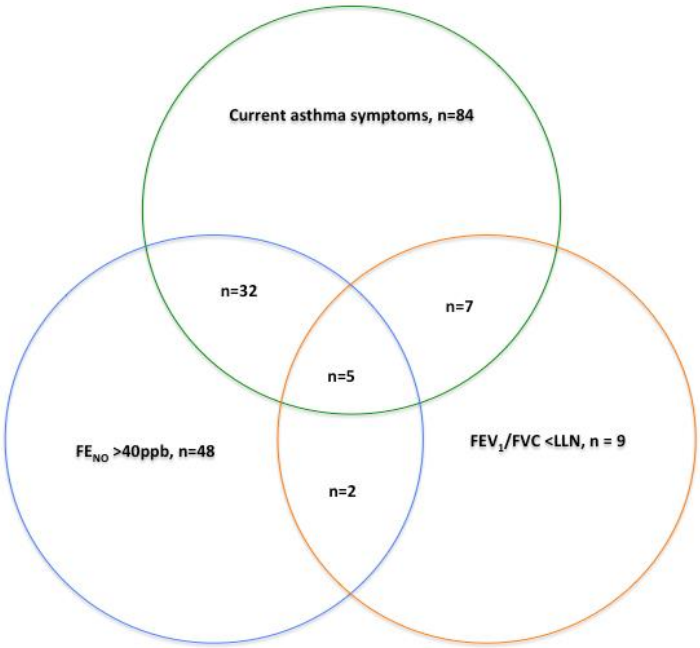

Figure 13: Intersections between the three asthma indices current asthma symptoms, airway inflammation (defined by FE_{NO} >40ppb), and airflow obstruction (FEV_1/FVC <LLN) across 187 foundry workers.

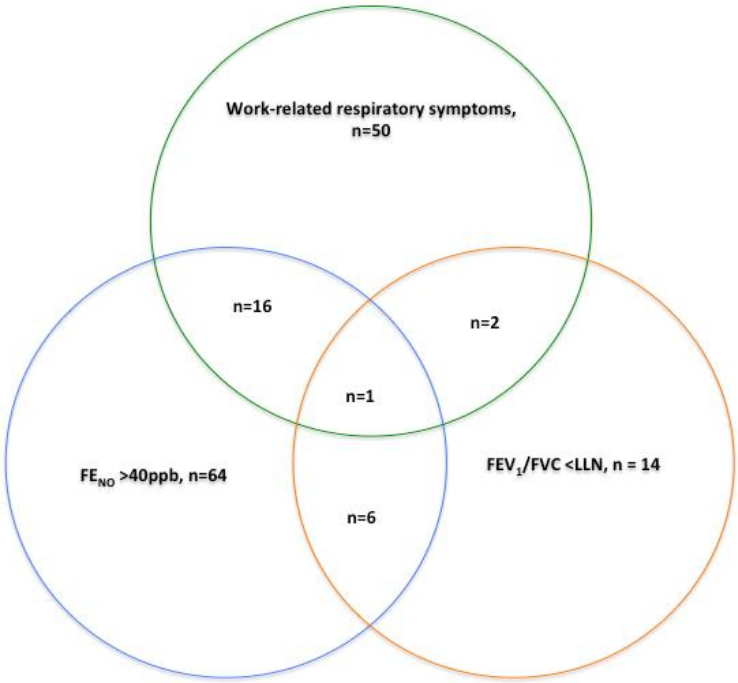

Figure 14: Intersections between work-related respiratory symptoms, airway inflammation (defined by FE_{NO} >40 ppb) and airflow obstruction (defined by FEV_1/FVC <LLN) in 153 foundry workers.

Figure 15 shows the proportion of workers diagnosed with asthma who had either CAS+airway inflammation or WRRS +airway inflammation. Nearly two-thirds (n=23, 62%) of those with CAS and airway inflammation did not have an asthma diagnosis. Two-thirds (n=10, 67%) of those with WRRS and airway inflammation did not have an asthma diagnosis. As a composite variable, CAS and airway inflammation had a 43.8% (95% CI 26.4 – 62.3%) sensitivity and 92.3% (88.7 - 95.1%) specificity for predicting a diagnosis of asthma, with a positive predictive value (PPV) of 37.84% (25.9 - 51.8%) and a negative predictive value (NPV) of 93.9 % (91.9 - 95.4%).

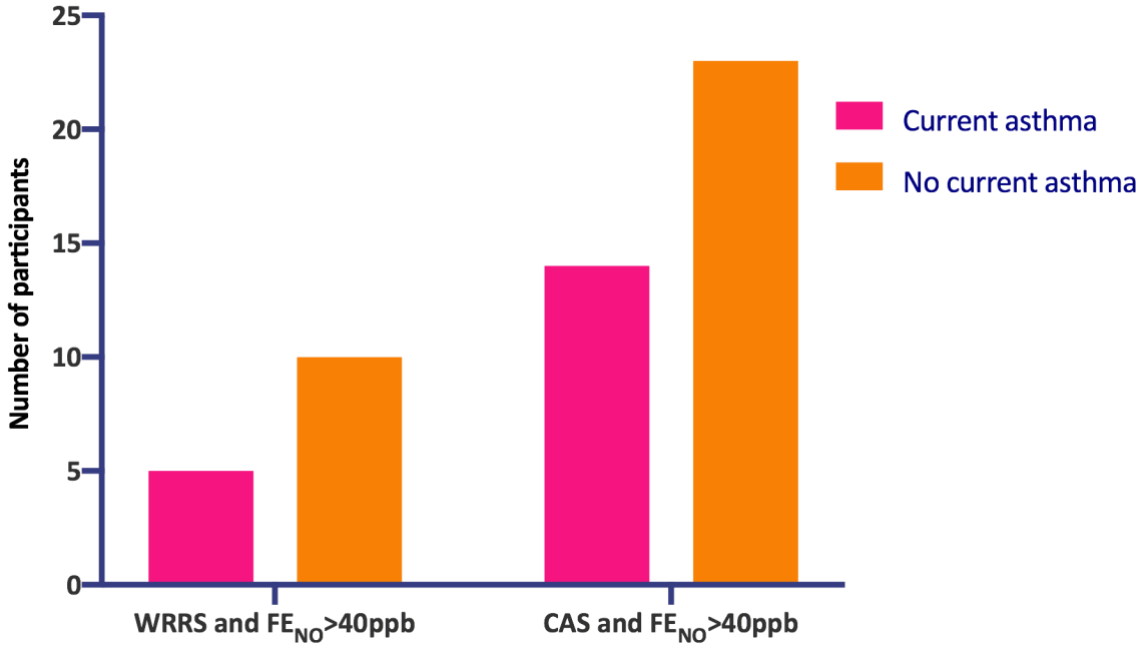

Figure 15: Proportion of study participants with and without a current asthma diagnosis among workers with WRRS and airway inflammation (defined by FE_{NO} >40ppb) and CAS and airway inflammation.

4.4 Discussion

This cross-sectional study of British foundry workers found isocyanate, but not formaldehyde, exposure was associated with small but significant increases in eosinophilic airway inflammation, despite low exposures. Overall, foundry workers had high levels of airway inflammation measured using FE_{NO}, with 25% of the workforce having a FE_{NO} exceeding 40ppb. Isocyanate exposure was associated with increasing airway inflammation in linear regression models, at levels substantially lower than the current UK WEL, even after controlling for common confounders such as smoking or atopy. However, evidence for a dose-response across exposure groups was less clear, with medium, but not high, exposed workers at increased risk for FE_{NO} above 40ppb and FEV_1 <LLN compared with the lowest exposed group. Using a foundry-specific JEM to attribute exposures, no clear exposure-response relationship was found between exposure to formaldehyde or isocyanates and risk of CAS, WRRS, or self-reported asthma. Unexpectedly, increasing isocyanate exposure was associated with an increase in FEV_1 and PEF in linear regression models. A significant proportion of symptomatic

workers (with either CAS or WRRS) had evidence of airway inflammation, but there was little overlap between airway inflammation and airflow obstruction among the study population. Most of these workers had no diagnosis of asthma, suggesting FE_{NO} may provide additional information to spirometry alone when screening at-risk populations for OA.

4.4.1 Limitations

Few recent studies have investigated allergic respiratory disease in foundry workers. The current study considered the effect of exposure to two respiratory sensitisers – formaldehyde and isocyanates – based on the processes and exposures present in the foundries recruited. Other potential respiratory sensitisers employed in foundries include, but are not limited to, FFA, cobalt, nickel sulphate, chromium VI, stainless steel, and welding fume (192). A detailed exposure assessment allowed exclusion of these other sensitisers as either not presently used or detected (162). However, this does not account for historical exposure to sensitisers nor exclude their occasional use. Past or occasional exposures may contribute to the risk of OA in exposed populations, and the magnitude of their effect can be difficult to quantify in observational studies (9).

Surprisingly, increasing exposure to FP was associated with small but significant increases in FE_{NO}. Whilst FP was quantitatively measured, its constituent parts were not examined in detail. A recent study has shown foundry particulate to contain fragments of metals such as copper, nickel, chrome, tin and zinc (187). Such metal particles are also known to cause occupational asthma. Observed increases in FE_{NO} may therefore be due to allergen, rather than irritant, exposures.

FP may also contain irritants such as smoke, dust, or caustic substances, that are directly toxic to the pulmonary epithelium (162, 193). Increasing airway inflammation has previously been reported following exposure to cement dust; the authors suggested that direct epithelial injury rather than an

immune-mediated process was causative (194). FE_{NO} rises following irritant exposures may falsely overestimate the degree of asthmatic airway inflammation in the study population. However, even after controlling for FP exposures in regression models, an exposure-response effect for isocyanates and FE_{NO} remained. Further work is needed to explore the extent to which allergenic or irritant components of FP contribute to increasing airway inflammation in foundry workers.

This study differs from recent studies of foundry workers in its exposure assessment and assignment. Worksites underwent contemporaneous measurements of a number of respiratory allergens by a qualified occupational hygienist. A foundry-specific site-process JEM was utilised to assign exposures to workers based on job title, process exposures, and workplace exposures. Even using this method, exposure misclassification is possible if individuals were assigned exposures based on a small number of measurements for a particular site-process combination. However, any misclassification is likely to bias the results towards the null; that is to say, exposures are likely to be under- not overestimated meaning any effects observed underestimate any exposure-response relationships (142).

A standardized and validated questionnaire was used to determine atopy, respiratory symptoms, WRRS, and history of respiratory diseases. However, these questionnaires rely on self-report and may be affected by recall bias (195). Using a history of atopic symptoms may overestimate the 'true' prevalence of atopy among a group of workers. Conversely, where definitions rely on self-report, underreporting may underestimate the true incidence of symptoms or disease within a population (30). Underreporting of symptoms in workplace studies may underestimate the true symptom or disease burden of a population.

4.4.2 Exposure assessment in British foundry workers

Few studies have reported exposures to isocyanates and formaldehyde in foundry workers. In their longitudinal study of foundry workers, Löfstedt and colleagues described mean isocyanic acid exposures of 18 µg/m^3, methylisocyanate exposures of 3.7 µg/m^3, and formaldehyde exposures of 0.05 mg/m^3 (192). In the current study, workers were exposed to diisocyanates, predominantly MDI. In comparison to the UK WEL, isocyanate exposures were approximately one-twelfth the 20 µg/m^3 8 hr-TWA and formaldehyde exposures were one-eighteenth the 2.5 mg/m^3 8-hr TWA. The current study was powered to detect evidence of disease at respirable dust levels close to the UK WEL, requiring 400 workers in the cross-sectional arm in order to achieve adequate power to detect an effect. As such, the power to detect evidence of disease among workers may have been reduced. The observation of lower exposures among foundry workers could be related to selection bias among participating worksites with better exposure controls. However, it is an encouraging sign and suggests use of adequate control measures in order to reduce the risk of exposure to respiratory hazards.

Although exposure levels were below the WEL, symptoms have been reported in foundry workers even at low exposure levels. A Swedish longitudinal study showed increased cough, nasal and ocular symptoms in exposed versus non-exposed workers at levels of formaldehyde exposure lower than in the current study (176). Furthermore, studies of isocyanate-exposed foundry workers have suggested that respiratory symptoms and asthma may occur at exposure levels significantly lower than the UK WEL, and a previous British study reported symptoms where isocyanate exposures were undetectable (173). There is no safe exposure threshold for respiratory sensitisers, and susceptible individuals may develop disease even at very low exposures. Current guidance aims, therefore, to achieve levels of exposure that are as low as reasonably practicable, in order to minimise the risk to entire workforce. This is one of the first studies to model foundry exposures in detail and evaluate their relationship with allergic respiratory disease. The longitudinal arm of this research programme

will generate valuable information on how allergen exposure affects asthma risk in foundry workers over time, even at low exposures.

4.4.3 Respiratory symptoms and asthma in British foundry workers

Symptom prevalence was lower in the current study compared to a previous study of British foundry workers. Cherry and colleagues reported symptom prevalence of 33% for WRRS, 9% for work-related wheeze, 15% for work-related cough, 16% for work-related chest tightness, and 8% for work-related breathlessness among workers with a mean age of 43.9 years, of whom 33% were current smokers (173). Mean age among foundry workers in the current study was similar (42.3 years) with slightly lower rates of current smoking (27%). We found overall rates of WRRS of 20%: 8% for work-related wheeze, 13% for work-related cough, 8% for work-related chest tightness, and 1% for work-related breathlessness. This comparative analysis suggests a reduction in WRRS among foundry workers over time, particularly in work-related breathlessness and chest tightness. This may be explained by a reduction in foundry allergen exposures over the last 15 years and suggests an improvement in respiratory symptoms among foundry workers.

There was no clear relationship between medium or higher exposures and risk of respiratory symptoms or asthma. In fact, higher exposures tended to be associated with a lower risk of current asthma symptoms, with borderline significance (OR 0.59 in highest versus lowest exposure, 95% CI 0.33-1.06, p=0.08). Few studies have attempted to evaluate risk of respiratory symptoms and asthma across a spectrum of foundry exposures; most have focussed on comparing exposed to non-exposed groups. Kuo *et. al.* reported no difference in symptom prevalence among foundry workers where exposures varied with job description, although did report higher symptom prevalence among smokers (196). In contrast, Löfstedt *et. al.* reported a higher risk of nasal symptoms with increasing exposure gradient in foundry workers exposed to isocyanates and formaldehyde when compared with controls (176). In the current study workers in the highest exposed category were

significantly more likely to use RPE, which may have attenuated any exposure-response. Furthermore, workers may not perceive, may underreport, or may not experience respiratory symptoms for a variety of reasons that may attenuate any exposure-response relationships (30). Examining symptoms in changing exposure environments, and over time, may reveal clearer evidence of an exposure-response relationship.

There was some suggestion of a remaining 'healthy worker' population in the current study, with workers in the highest exposure group tending to have a lower risk of respiratory symptoms and asthma than those in the lowest exposure group. In specific regression analyses, workers with a current asthma diagnosis and had significantly more years' exposure to foundry work, although the magnitude of effect was small. This contrasts with what may be expected with a healthy worker survivor effect, where one sees a reduction in asthma risk with increasing years' exposure reflecting individuals' early departure from, or lack of recruitment to, the workforce. The short latency of OA means the healthy worker effect is often difficult to detect in cross-sectional studies, and there is no single measure of healthy worker effect bias in asthma (146, 197). The study was unable to capture recent movement of workers from a high to a low exposure environment, and only identified three workers who previously left a job for health reasons (the 'healthy worker effect') (146). Longitudinal studies of foundry workers are required to examine whether respiratory disease is associated with premature exit or relocation within the foundry industry.

4.4.4 Impact of smoking and atopy on respiratory symptoms and asthma in foundry workers

Both smoking and atopy impacted respiratory symptoms and asthma in foundry workers. Ever smokers were at increased risk of CAS (OR 2.22, 95% CI 1.4-3.51), any respiratory symptom (OR 2.28, 95% CI 1.46 – 3.57) and upper airway symptoms (1.66, 1.05-2.6) compared with never smokers. However, no increased risk was seen among ever smokers for WRRS, self-reported or current

asthma. Foundry workers are well-documented to be at increased risk of COPD (179, 180). This may explain the increase in symptoms among smokers in the absence of a clear association to work or asthma. However, lung function among the current population was within normal limits, and rates of obstructive airways disease were low. In addition, workers may not recognise or report work-relatedness to their symptoms, which would point to more variability in their condition and a potential diagnosis of allergic airways disease (198). Longitudinal study of these symptomatic foundry workers will help understand whether these symptoms are explained by early airways disease.

In contrast, workers with atopy were four times more likely to self-report asthma (4.31, 2.22-8.36) and three times more likely to fulfil ECRHS asthma criteria (3.56, 1.62-7.85), but significant relationships between atopy and either WRRS or CAS were not observed. Atopy has been shown to increase the risk of isocyanate OA in a case-referent study of British foundry workers, with a more modest increase in OA risk observed for smokers (199). Atopy is well recognised to increase OA risk in workers exposed to HMW and some LMW agents, although the mechanisms through which atopy modifies OA risk in LMW-exposed workers is unclear (9, 10). The relationship between smoking and asthma is less well established. Some studies report exposure to pollutants such as cigarette smoke may alter asthma phenotype and treatment efficacy, but fewer studies have shown a direct causal link (20, 200). In the current study over a quarter of the population were current smokers and over half had ever smoked. Further examination of how exposure to smoke and other pollutants augment asthma risk is important in helping to reduce the future risk of asthma in foundry workers.

4.4.5 Relationships between spirometry and foundry exposures

Among our study population, FEV_1, FVC, and PEF were similar to the expected percentage predicted values. Mean percent predicted FEV_1, FVC, and PEF did not differ significantly across exposure groups, and in adjusted regression models no significant associations between exposure and

categorical spirometric outcomes were observed. Foundry workers have previously been shown to have significantly poorer predicted FEV_1 compared to lesser or non-exposed controls (201). RPE use was more prevalent among those with higher exposures (82% versus 65% in medium exposed, $p<0.01$). This may explain the absence of abnormal lung function in higher exposed groups and supports its use in such populations.

Increasing $\mu g/m^3$ isocyanate exposure was associated with small but significant increases in percent predicted FEV_1 in linear regression models (unstandardised β 1.77%, 0.31-3.23). This was an unexpected finding, and contradicts previous research showing foundry workers are at increased risk for obstructive airways disease even after accounting for smoking (202). A healthy worker hire effect or healthy worker survivor effect could explain this aberrant finding. Workers in the current study had spent an average of 14.76 (12.66) years in the foundry industry, and the highest exposure group had significantly more years of service than the lowest (16.64 versus 12.33 years, $p <0.05$). Self-reported job change due to workplace exposures was very low in the population and did not differ between exposure groups. This suggests a relatively stable workforce, with those with higher exposures staying longer in the job. This is particularly important in foundry work, where hazards such as molten metal are immediate and obvious, and the working environment can be hostile. Individuals who are unable to work in foundry environments are likely to leave the workplace early, with those who remain more able to manage the working conditions. This healthy worker survival effect may attenuate any differences between lung function seen at different exposure levels.

4.4.6 Relationships between airway inflammation and foundry exposures

The current study found increasing exposure to isocyanates was associated with increasing airway inflammation. GMR FE_{NO} increased by 11% (GMR 1.11, 1.01 – 1.21) per $\mu g/m^3$ rise in isocyanate exposure. A previous study of car spray painters found no significant relationship between increasing isocyanate exposure and FE_{NO} using either continuous or dichotomized endpoints but

reported a strong relationship between FE_{NO} and risk of BHR (203). FE_{NO} has been shown to rise following positive SIC to isocyanates, rising at 8 hours and peaking around 24 hours post-exposure (182). Furthermore, the same study reported significant FE_{NO} increases in non-asthmatics exposed to isocyanates with, but not without, BHR. This suggests that FE_{NO} might be useful not only in identifying incident asthma in isocyanate exposed workers, but also in detecting workers with asymptomatic BHR at risk of becoming asthmatic, even at levels of exposure lower than the UK WEL.

In contrast, the current study found no clear relationship between airway inflammation and increasing formaldehyde exposure. Associations between formaldehyde exposure and FE_{NO} have not been widely reported. Previous studies into formaldehyde OA have failed to demonstrate an association with eosinophilic inflammation, with the authors suggesting formaldehyde may reduce thresholds for other exposures provoking asthmatic symptoms at lower exposure levels rather than directly inducing inflammation (185). A proportion of LMW-OA is thought to be non-eosinophilic (19); therefore, exposures may drive non-eosinophilic inflammation or other cause symptoms by other mechanisms (5). The potency of an occupational allergen may also influence its ability to induce airway inflammation, and statistical models that predict the hazard associated with LMW exposures suggest isocyanates have a stronger asthmatic potential than aldehydes (204).

This study supports an association between increasing FP exposure and airway inflammation using a categorical endpoint. Among foundry workers there was a 9% increased risk of FE_{NO} above 40ppb with each mg/m^3 increase in FP. Furthermore, in linear models we found respirable FP was associated with small but significant increases in GMR FE_{NO}. One study has previously explored relationships between dust exposure and airway inflammation in foundry workers, reporting increased nitric oxide levels in workers with higher cumulative exposures to RCS and foundry particulate. The authors concluded that FP exposure may induce pulmonary inflammation, measurable through FE_{NO} (187). Previous hygiene studies have shown FP to contain allergenic

substances including metal particles (205). Increasing airway inflammation in association with FP exposure may therefore be related to allergen rather than irritant exposure. Furthermore, high general exposure to vapours, dusts, gases and fumes has previously been associated with higher sensitiser exposure, suggesting that non-sensitisers may be useful as a surrogate for general hygiene measures at worksites (88).

4.4.7 Fractional exhaled nitric oxide as a health surveillance tool in foundry workers

Among symptomatic foundry workers, the current study found differences in the proportion of symptomatic workers with airway inflammation or airflow obstruction. A fifth (19%) of foundry workers with CAS also had airway inflammation, whereas only 6% had both CAS and airflow obstruction. Similar relationships were observed for foundry workers with WRRS; 11% also had airway inflammation, whereas only 2% had WRRS and airflow obstruction. Among asymptomatic workers, intersections between airway inflammation and airflow obstruction were limited. This suggests that airway inflammation and airflow obstruction identify discrete groups of symptomatic foundry workers, and the addition of measures of airway inflammation such as FE_{NO} may be useful in screening workers at risk of OA. A fall in FEV_1 is a late sign in OA, and early identification of workers with the condition is vital in improving both clinical and employment outcomes (11, 14). FE_{NO} could be useful in detecting early airway inflammation in symptomatic workers, prior to the onset of obstructive spirometry, and further investigation of this is warranted.

A number of external factors may affect FE_{NO} as a potential tool for health surveillance. Atopy and smoking are well known to moderate FE_{NO} (46). We found foundry workers with atopy to be at significantly increased risk of having a FE_{NO} above 40 ppb (OR 1.95, 1.11-3.42, p=0.02) and smokers to be at significantly reduced risk (0.47, 0.27-0.83, p=0.01). The longitudinal arm of the current study will provide further detail about the extent to which atopy and smoking influence the development

of airway inflammation and asthma in British foundry workers, and how this might be accounted for in future health surveillance programmes.

Foundry workers are largely exposed to LMW allergens. The role of FE_{NO} in LMW exposures is particularly debated, with a lack of consensus on its use in either SIC or workplace studies (126, 155). In contrast, there is more evidence supporting a relationship between FE_{NO} and exposure to HMW agents (40). Comparing the current findings to evidence from HMW environments may help to understand the extent to which the molecular weight of exposure can influence airway inflammation; and whether relationships between airway inflammation, airflow obstruction, and symptoms are more significant in HMW exposure environments.

4.4.8 Summary

This study of isocyanate and formaldehyde exposed foundry workers has demonstrated that though current asthma symptoms are common, they are not clearly related to exposure. However, airway inflammation appears to be related to increasing isocyanate exposure, even after controlling for common confounders. Further study is needed to examine how airway inflammation relates to other asthma indices over time, and to better understand the role of FE_{NO} in different exposure environments.

5 Respiratory symptoms, airway inflammation, and lung function in laboratory animal workers

5.1 Introduction

5.1.1 Laboratory animal allergy

Laboratory animal workers are exposed to HMW allergenic animal proteins such as dander, urine, or skin (206). Sensitisation to such proteins, measurable by SIgE or SPT, is present in nine per cent of laboratory animal workers and is strongly associated with the development of laboratory animal allergy (LAA) and OA (207). LAA is defined as either occupational rhinitis (OR), OA, or anaphylaxis caused by exposure to laboratory animal allergens and has been reported in up to 10% of exposed workers (207). LAA remains a common cause of OR and OA in the UK (26).

Allergen exposure is a key risk factor for the development of LAA. Both duration and intensity of exposure to laboratory animal allergens have been shown to significantly increase LAA risk. This includes the number of hours' exposure, as well as the number of animals handled at any one time (208). Recent changes in research practice, animal husbandry, and health and safety legislation have altered how animal research is approached, leading to a reduction in the duration and intensity of allergen exposure (209). Genetically modified mice models are now preferred for *in vivo* work, with animals increasingly housed in individually ventilated cages (IVCs) to prevent experimental contamination and infection of animals by airborne microbes (207). New legislative requirements necessitate individuals working in animal facilities to be registered with the UK Home Office, with workers undergoing training in animal welfare in order to achieve registration (210). Health and safety practices have been improved in facilities in order to prevent cross-contamination of infection between animals and to comply with the three Rs - replacement, reduction and refinement - of animal experimentation (211). However, most studies of LAA were undertaken prior to these

changes in working, with few studies examining the risk of sensitisation and LAA following their introduction.

5.1.2 Fractional exhaled nitric oxide in laboratory animal workers

Fractional exhaled nitric oxide (FE_{NO}) has been shown to be higher in people with LAA compared to asymptomatic exposed controls: stepwise increases in FE_{NO} have been demonstrated across groups of exposed workers, workers with early LAA (OR and sensitisation) and workers with OA (44). Sensitised laboratory animal workers have been shown to have significantly higher FE_{NO} than sensitised controls, despite no differences in spirometry between groups (212). A study of laboratory animal workers with work-related symptoms reported increases in FE_{NO} following increasing allergen exposure in sensitised individuals (213).

Relationships between HMW exposures, such as animal allergens, and eosinophilic airway inflammation are more established than for LMW exposures. Mechanisms for OA in HMW-exposed workers are well described, frequently related to Th-2 driven inflammation, associated eosinophilia, and SIgE (10). Data from SIC studies support a rise in FE_{NO} following exposure to HMW, but not LMW, allergens (40). Longitudinal data from workers exposed to flour supports the use of FE_{NO} in predicting incident BHR, suggesting it may be useful tool in screening for at-risk populations for OA in these settings (41, 214).

5.1.3 Study rationale

Data reported in this thesis has shown increasing airway inflammation, measured using FE_{NO}, with exposure to LMW agents including isocyanates and wood. However, a consistent dose-response effect has not been found. No recent studies have examined the relationship between airway inflammation and laboratory animal exposures in workers at risk of LAA. This study aims to understand relationships between exposure to laboratory animal allergens and work-related

symptoms, sensitisation, asthma, airway inflammation, and lung function in a subset of laboratory animal workers taking part in the SPIRAL (Safe Practice in Reducing Allergy in Laboratories) study (215). Further, it aims to understand whether associations between exposure and airway inflammation were present in this group, and whether these associations were influenced by sensitisation.

An NHS REC committee approved this study (reference 13/NI/0208).

5.2 Methods

Data on laboratory animal workers was collected as part of the Safe Practices in Reducing Allergy in Laboratories (SPIRAL) study, led by Dr Johanna Feary from the National Heart and Lung Institute, Imperial College London, and is reproduced here with Dr Feary's kind permission. The existing study protocol was amended to include measurement of lung function and ethical approval was agreed. Data presented here was collected alongside Dr Feary and Bernadette Fitzgerald across eight site visits over a two-year period.

5.2.1 Power calculation

No recent studies have examined LAA in Britain since the increasing introduction of mice models. Therefore, evidence from rat-exposed populations was used. In their study of rat-exposed laboratory animal workers, Cullinan *et. al.* reported 37% of workers had work-related chest symptoms at GM exposure to rat urinary allergen of 1.26 $\mu g/m^3$ (95% CI 0.86 - 1.85) (216). At the same exposure, there was an increased risk of respiratory symptoms with an OR of 3.5 (95% CI 0.7-18.7) versus controls. On this basis a power calculation was performed to achieve 80% study power at a two-sided 5% significance level (130). A sample size of 187 participants was estimated to provide adequate power, assuming a true underlying symptom prevalence of 25% and a OR of 3.5 between the highest and lowest exposed (130).

5.2.2 Study population

SPIRAL was established to determine whether the introduction of IVCs in laboratory animal facilities has led to a reduction in LAA populations of exposed workers. SPIRAL was a multicentre study of 750 laboratory animal workers across seven UK research institutions that ran between July 2014 and March 2017 (215). In brief, workers were recruited if they had at least four months of exposure. Mice were used most frequently among the participating research institutions. The primary study endpoint was sensitisation to mouse proteins in individuals working in IVC-only versus other (mixed or open) facilities. Only workers with fewer than three years' exposure were included in the primary analysis. All workers over 18, employed at participating worksites and exposed to laboratory animals were included. Individuals were excluded if they had ever worked with animals at an institution other than their current research facility.

As part of this thesis a subgroup of SPIRAL participants were recruited. Laboratory animal workers participating in SPIRAL were recruited over a 12-month period between November 2015 and October 2016 (Appendix D). All SPIRAL participants recruited within the study window were invited to undergo FE_{NO} and spirometry testing in addition to the standard SPIRAL testing. SPIRAL participants were consented for the additional tests and FE_{NO} and lung function were performed on the same day as the other study examinations.

5.2.3 Questionnaire and allergy testing

As part of the SPIRAL study protocol, participants undertook an online questionnaire detailing their work environment (open cages, mixed, or IVC-only), job histories, and current and past animal exposures (Appendix D). Work-related ocular, nasal or respiratory (chest-tightness, difficulty breathing, wheezing or whistling in chest) symptoms were self-reported. Respiratory symptoms occurring on exposure to any animals, and also to specific animal species, were documented. Individuals self-reported hay fever symptoms and current or previous diagnosis of asthma.

Standard immunological testing included in SPIRAL included SPT to common aeroallergens including grass pollen, *Dermatophagoides pteronyssius* (house dust mite), cat, and dog, and specific SPTs to mouse and rat epithelium. A negative saline and positive histamine control were used (Allergopharma, Diagenics, Milton Keynes, UK and Stallergenes, London, UK). SPTs were timed at 15 minutes and then allergen was removed from the skin. Visible wheals were copied and recorded in the study file.

5.2.4 FE_{NO} and spirometry

In addition to the standard SPIRAL protocol, participants underwent FE_{NO} and lung function testing, as described previously in section 3.2.5, according to ATS/ERS guidelines (46, 133).

5.2.5 Exposure assessment

Background and personal exposure to the primary mouse allergen Mus-m-1 was determined in IVC-only versus open or mixed facilities (215). In brief, inhalable particulate was collected from selected individuals working in either IVC-only or open facilities and analysed using a commercial enzyme-linked immunosorbent assay (ELISA). Background measures were collected using static samplers. Both background and personal Mus-m-1 levels were significantly lower in IVC-only facilities: background geometric mean (GM) Mus-m-1 was 0.04 ng/m^3 in IVC-only versus 0.53 ng/m^3 in open or mixed facilities, and personal GM Mus-m-1 was 1.00 ng/m^3 versus 8.35 ng/m^3 in open or mixed facilities .

Most workers with LAA will develop symptoms within three years of starting work, and the risk is highest in the first two years (217). Thus, the primary analysis in SPIRAL was limited to participants with less than three years' exposure. Individual exposures were not modelled in the SPIRAL study and are therefore not included in the current analysis.

5.2.6 Definitions

Work-related nasal symptoms (WRNS), work-related ocular symptoms (WROS) and WRRS were defined respectively as nasal, ocular, or respiratory symptoms worsening on exposure to animals in the workplace or improving when away from work at weekends or on holidays. Current asthma was defined as self-reported asthma within the last 12 months.

Individuals were considered atopic if they had positive SPTs to any common aeroallergen, defined as a saline adjusted wheal diameter equal or greater than 2mm. Specific sensitisation to mouse or rat was defined by a positive SPT to mouse or rat epithelium if a saline adjusted wheal size was equal to or greater to 2mm (215). A self-reported doctor diagnosis of LAA was used to define the LAA group. A high FE_{NO} was defined as a mean FE_{NO} exceeding 40ppb and spirometry was considered abnormal if FEV_1, FVC, PEF or FEV_1/FVC fell below their age, ethnicity, and height adjusted LLN value, as previously described in section 3.2.9.

5.2.7 Data analysis

Univariate analyses were conducted using independent student's t-tests for continuous and chi-squared analyses for categorical data. Continuous data were expressed as mean ± SD with categorical data expressed numerically with associated percentages. FE_{NO} data did not meet normality criteria so were log transformed and data expressed as GM and GSD for averages and as GMR in regression models, as described in section 3.2.10.

Binary logistic regression models were constructed to examine associations between years of exposure and the key categorical variables of interest: work-related symptoms; current asthma; specific sensitisation to mouse or rat epithelium; FE_{NO} above 40ppb; and FEV_1, FVC or PEF less than LLN. A three-year exposure threshold was used, and workers with less than three years' exposure were used as the control group. Relationships between years' exposure, FE_{NO} and spirometry were

also explored using multiple linear regression, with log FE_{NO} and percent predicted spirometry being the key dependent variables. Regression models were constructed and controlled for as previously reported in section 3.2.10. FE_{NO} data was further analysed by presence or absence of work-related symptoms (WRS) (44).

Linear regression models were constructed to explore a potential healthy worker effect among laboratory animal workers, as described in section 3.2.10. Years of exposure to laboratory animals in worker's current job were used as the dependent variable, with work-related symptoms, CAS, and current asthma as independent variables. Data were adjusted for age, sex, atopy, height, and ever smoking.

Odds ratios and unstandardised β values with associated 95% confidence intervals (CI) were reported, with two-way significance taken at the 5% level.

Stratified analyses were conducted to explore differences between workers with and without specific sensitisation (213). Finally, intersections between groups of workers with respiratory symptoms, airway inflammation, and airflow obstruction were examined using contingency analyses and Venn diagrams as previously described in section 3.2.10. All statistical analyses were conducted using SPSS (141).

5.3 Results

5.3.1 Study population

One hundred and fifty-five people were recruited to SPIRAL during the 12-month study window, of whom 153 (99%) took part in the current study (Table 23). The two non-participants declined lung function testing. Mean age of the study group was 29.21 (SD 7.96) years; just over half (n=89, 58%)

were female and a sixth (n=25, 16%) had ever smoked more than one pack year. Twenty (13%) participants were current smokers. Mean BMI was 24.28 (SD 4.4) kg/m^2. There were no significant differences in sex, age, smoking habits, BMI, or current job title between the two groups.

Table 23: Demographics of the study population stratified by duration of exposure to laboratory animals

Demographics	**Less than three years' exposure (n=66)**	**More than three years' exposure (n=87)**	**Total (n=153)**
Age, years (SD)	28.00 (7.70)	30.13 (8.08)	29.21 (7.96)
Sex, f (%)	43 (65)	46 (53)	89 (58)
Current smoker, n (%)	9 (14)	11 (13)	20 (13)
Ever smoked > 1 pack yr, n (%)	11 (17)	14 (16)	25 (16)
BMI, kg/m^2 (SD)	23.87 (4.45)	24. 56 (4.49)	24.28 (4.40)
Height, cm (SD)	168.92 (9.15)	169.28 (9.10)	169.12 (9.09)

The exposure characteristics of the study population are shown in Table 24. The majority of participants were scientists (n=124, 81%). On average, participants had spent 5.22 (SD 5.08) years in their current job. All participants worked with mice; 62 workers (41%) were also exposed to rats. Sixty-six workers (43%) had been exposed to laboratory animals for three years or less. Workers with more than three years' exposure were more likely to work in an IVC-only facility (31% versus 17%, $p < 0.05$). Participants with more than three years' exposure to laboratory animals had worked significantly longer in their current job compared to those with less than three years' exposure (7.40 versus 2.35 years, $p<0.01$). Scientists and laboratory technicians were equally represented between the two groups.

Table 24: Exposure characteristics of the study population stratified by duration of exposure to laboratory animals

Exposure	Less than three years' exposure (n=66)	More than three years' exposure (n=87)	Total (n=153)
Technician, n (%)	5 (8)	16 (18)	21 (14)
Scientist, n (%)	58 (88)	66 (76)	124 (81)
Office, n (%)	3 (5)	4 (5)	7 (5)
Maintenance, n (%)	0 (0)	1 (1)	1 (1)
Currently works in IVC-only facility, n (%)	11 (17)	27 (31)*	38 (25)
Only ever worked in IVC-only facility, n (%)	6 (9)	16 (18)	22 (14)
Mice in animal facility, n (%)	66 (100)	87 (100)	153 (100)
Rats in animal facility, n (%)	22 (14)	40 (46)	62 (41)
Number of years in current job, years (SD)	2.35 (0.67)	7.40 (5.85)**	5.22 (5.08)
*p<0.05, **p<0.01. IVC = individually ventilated cages.			

The health characteristics of the study population are shown in Table 25. Upper airway symptoms were the most frequently reported complaint. Twenty-three percent of workers reported nasal symptoms and 17% reported ocular symptoms; WRNS were reported by 16% and WROS by 13%. Respiratory symptoms on exposure to pets were reported by 16%; however, WRRS were uncommon reported by only 3%.

SPTs were refused or not interpretable due to dermatographia in three workers (2%). Atopy was common, with 41% of participants having a positive SPT to at least one common aeroallergen. Specific sensitisation was less common; 13% had a positive SPT to mouse or rat epithelium. Current hay fever symptoms were reported by 29% of the study group. Atopic symptoms correlated well to the presence of SPT positivity (Figure 16).

Table 25: Health characteristics of the study population, stratified by more or less than three years' exposure to laboratory animal allergens.

Health	Less than three years' exposure (n=66)	More than three years' exposure (n=87)	Total (n=153)
Nasal symptoms, n (%)	14 (21)	21 (24)	35 (23)
WRNS, n (%)	9 (14)	16 (18)	25 (16)
Ocular symptoms, n (%)	10 (15)	16 (18)	26 (17)
WROS, n (%)	7 (11)	13 (15)	20 (13)
Respiratory symptoms, n (%)	10 (20)	9 (13)	19 (16)
WRRS, n (%)	3 (5)	2 (2)	5 (3)
Any work-related symptom, n (%)	11 (17)	18 (21)	29 (19)
Hayfever last 12 months, n (%)	20 (30)	25 (29)	45 (29)
Atopic, n (%)	27 (42)	36 (42)	63 (41)
+ve SPT mouse/rat, n (%)	6 (9)	14 (16)	20 (13)
Current inhaled steroid use, n (%)	1 (2)	2 (2)	3 (2)
Self-reported LAA, n (%)	4 (6)	4 (5)	8 (5)
Childhood asthma, n (%)	6 (9)	5 (6)	11 (7)
Current asthma, n (%)	2 (3)	2 (2)	4 (3)
AM FE_{NO}, ppb (SD)	19.49 (19.30)	31.14 (32.56)**	26.20 (28.23)
GM FE_{NO}, ppb (GSD)	13.83 (2.32)	22.18 (2.25)**	18.15 (2.35)
FE_{NO} >40ppb, n (%)	5 (9)	17 (20)	22 (15)
Mean % predicted FEV_1 (SD)[b]	101.97 (12.78)	98.68 (9.87)	100.17 (11.36)
Mean % predicted FVC (SD)	106.22 (12.88)	102.24 (9.43)*	104.05 (11.27)
Mean % predicted PEF (SD)	108.52 (17.57)	109.15 (14.31)	108.86 (15.82)
Mean FEV_1, litres (SD)	3.63 (0.91)	3.39 (0.68)^	3.50 (0.80)
Mean FVC, litres (SD)	4.44 (1.16)	4.11 (0.84)	4.26 (1.00)
Mean PEF, litres (SD)	527.77 (133.90)	519.76 (120.58)	522.35(122.32)
FEV_1 <LLN, n (%)	3 (2)	1 (1)	4 (3)
FVC <LLN, n (%)	1 (2)	1 (1)	2 (2)
PEF <LLN, n (%)	1 (2)	1 (1)	2 (2)
FEV_1/FVC <LLN, n (%)	2 (3)	1 (1)	3 (2)

*p<0.05, **p<0.01.
WRNS= work-related nasal symptoms. WROS = work-related ocular symptoms. WRRS = work-related respiratory symptoms. SPT = skin prick tests, LAA = laboratory animal allergy, LLN = lower limit of normal.

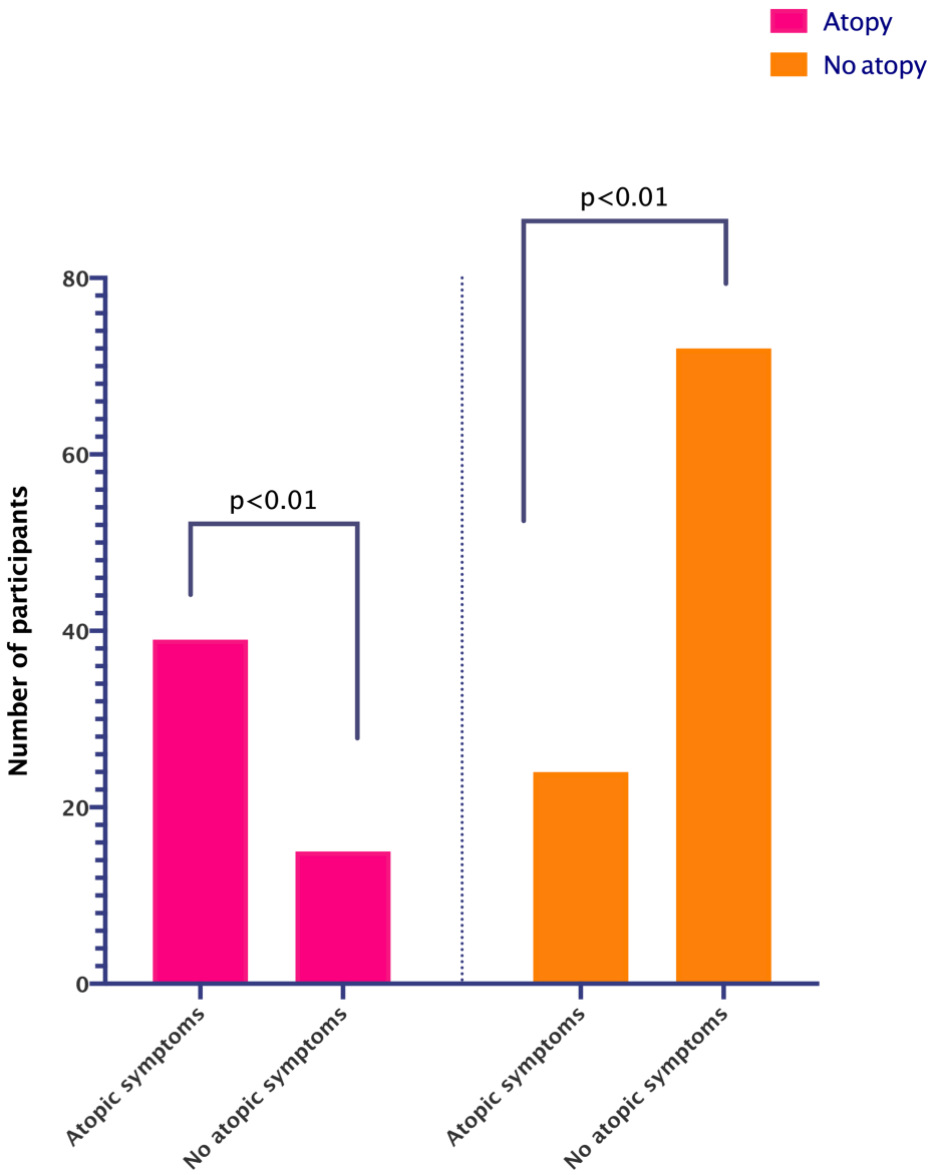

Figure 16: Presence or absence of atopy (defined by SPT positivity to a common aeroallergen) in workers with and without atopic symptoms

Seven percent of the study group reported childhood asthma. Prevalence of current asthma was low: only 4 (3%) workers self-reported a diagnosis. A further three workers (2%) were taking inhaled steroids none of whom reported a current diagnosis of asthma. Eight workers (5%) self-reported a diagnosis of LAA. There were no significant differences in prevalence of upper or lower respiratory symptoms, atopy, hayfever, or self-reported asthma or LAA between the two exposure groups.

Valid FE_{NO} measurements were obtained in 135 (91%) workers. One worker was unable to perform FE_{NO} due to recent dental surgery and a further 13 were unable to perform a reproducible test. Valid spirometry was performed in 135 (88%) participants: the remaining 16 (12%) were unable to perform a reproducible test. AM FE_{NO} was 26.20 (SD 28.23) ppb and GM FE_{NO} was 18.15 (GSD 2.35) ppb. Nineteen (14%) workers had a FE_{NO} above 40ppb. Mean percentage predicted FEV_1, FVC and PEF were all above 100% and few workers met LLN criteria for spirometry, with only 3% having an FEV_1 <LLN, 2% having an FVC <LLN, and 2% with an FEV_1/FVC <LLN.

FE_{NO} was significantly higher in workers with more than three years' exposure. GM FE_{NO} was 22.18 (GSD 2.25) ppb compared with 13.83 (2.32) in those with fewer than three years' exposure ($p<0.01$). Prevalence of FE_{NO} > 40 ppb was higher in workers with more than three years' exposure, but this was not statistically significant (18% versus 9%, p=0.13).

Percent predicted FVC was significantly lower among workers with more than three years' exposure (102% versus 106%, $p <0.05$). FEV_1 was within normal limits in both exposure groups and no significant difference was seen between FEV_1 between the two groups. The overall prevalence of abnormal spirometry using LLN criteria was low, and there was no significant difference in categorical spirometry outcomes between the two exposure groups in univariate analyses.

5.3.2 Associations between years' exposure and respiratory symptoms, atopy, and laboratory animal allergy

Associations between exposure and nasal, ocular, respiratory, and work-related symptoms were explored using logistic regression (Table 26). Non-significant relationships were observed between all symptom outcomes and exposure to laboratory animals of more than three years' duration. A non-significantly increased risk for both nasal (OR 1.07, 95% CI 0.48 – 2.41) and ocular (1.20, 0.48 – 2.97) symptoms was seen in workers with more than three years' exposure, whereas risk tended to

be reduced for respiratory symptoms (0.26, 0.05 – 1.48). A similar trend was observed for work-related symptoms, with non-significantly increased odds of WRNS (1.25, 0.49 - 3.19) and WROS (1.40, 0.50 – 3.87) and reduced risk of WRRS (0.43, 0.07 - 2.81) seen in workers with more than three years' exposure.

Atopic workers were at significantly increased risk of nasal and ocular symptoms compared to non-atopic workers (OR 2.47 and 2.92 respectively, Table 26). Similarly, risk for WRNS (2.66, 95% CI 1.05-6.79) and WROS (2.75, 0.99-7.59) was significantly increased in atopic workers. Risk for both respiratory symptoms and WRRS was higher in atopic workers although confidence intervals were wide, and data did not reach statistical significance.

Table 26: Relationships (odds ratios and 95% confidence intervals) between more than three years exposure to laboratory animals and nasal, ocular, respiratory, and work-related symptoms. All models were controlled for sex, atopy, ever smoking and BMI.

Upper and lower airway symptoms	**Nasal symptoms**	**Ocular symptoms**	**Respiratory symptoms**
> 3 years' exposure (yes=1)	1.07 (0.48 – 2.41)	1.20 (0.48 – 2.97)	0.26 (0.05 – 1.48)
Atopic (yes=1)	2.47 (1.10 – 5.57)*	2.92 (1.17 – 7.28)*	3.44 (0.59 - 19.97)
Work-related symptoms	**WRNS**	**WROS**	**WRRS**
> 3 years' exposure (yes=1)	1.25 (0.49 - 3.19)	1.40 (0.50 – 3.87)	0.43 (0.07 - 2.81)
Atopic (yes=1)	2.66 (1.05 – 6.79)*	2.75 (0.99 – 7.59)*	2.00 (0.29 - 13.58)

Data presented as odds ratios and 95% confidence intervals. > 3 years exposure = more than three years exposure to laboratory animals. Atopy defined by positive skin prick tests to more than one common aeroallergen. WRNS = work-related nasal symptoms. WROS = work-related ocular symptoms. WRRS = work-related respiratory symptoms. *p<0.05, **p<0.01

Atopic workers were over seven times more likely to have specific sensitisation than non-atopic workers (OR 7.79, 95% CI 2.29-26.83, Table 27), although confidence intervals were wide. In addition, atopic workers tended to be more likely to self-report LAA, although this just missed statistical significance (8.45, 0.95 – 75.07, p=0.06). Interestingly, ever smoking was associated with a

reduced risk of specific sensitisation (0.09, 0.01 - 0.92), whereas BMI was associated with an increased risk (1.18, 1.05 - 1.32).

More than three years' exposure was not significantly associated with increased risk of asthma, sensitisation, or self-reported LAA (Table 27). Workers with more than three years' exposure had a non-significantly increased risk of specific sensitisation (1.81, 95% CI 0.61 – 5.62). In contrast, risk of self-reported LAA (0.51, 0.10 – 2.49) and of current asthma (0.35, 0.03 – 4.39) tended to be lower in workers with more than three years' exposure.

Table 27: Associations (odds ratios and 95% confidence intervals) between asthma, sensitisation and self-reported laboratory animal allergy with years exposure, atopy, and smoking status. All models were controlled for atopy, ever smoking, BMI, and gender with significant cofactors reported.

	Childhood asthma	**Current asthma**
> 3 years' exposure (yes=1)	0.29 (0.07 - 1.27)	0.35 (0.03 – 4.39)
Atopic (yes=1)	1.40 (0.34 – 5.87)	2.56 (0.20 – 32.27)
Ever smoked (yes=1)	0.65 (0.07 – 6.14)	10.70 (0.77 – 149.32)
	Specific sensitisation	**Self-reported LAA**
> 3 years' exposure (yes=1)	1.85 (0.61 – 5.62)	0.51 (0.10 – 2.49)
Atopic (yes=1)	7.79 (2.26 – 26.83)**	8.45 (0.95 – 75.07)
Ever smoked (yes=1)	0.09 (0.01 - 0.92)*	0.70 (0.07 – 7.64)
BMI, mg/m²	1.18 (1.05 – 1.32)**	1.03 (0.85 – 1.26) 0.75

Data presented as odds ratios and 95% confidence intervals. > 3 years exposure = more than three years exposure to laboratory animals. Atopy defined by positive skin prick tests to more than one common aeroallergen. Ever smoked = ever smoked more than one pack year history. *p<0.05, **p<0.01

5.3.3 The Healthy Worker Effect in laboratory animal workers

Table 28 shows data from linear regression analyses exploring the healthy worker effect in laboratory animal workers. No significant associations were seen between years' exposure to laboratory animals and any of the independent variables studied. Overall magnitude of associations was small for all variables studied.

Table 28: Linear regression models for years' exposure to laboratory animals, respiratory symptoms, and asthma. Unstandardised β coefficients are reported with 95% confidence intervals.

	Nasal sx	Eye sx	Resp sx	WR nasal	WR eyes	WRRS	Any WRS	Current asthma
Years' exposure to laboratory animals*	-0.003 (-0.017 - 0.011)	-0.003 (-0.015 - 0.010)	-0.004 (-0.011 - 0.003)	0.001 (-0.012 - 0.013)	0.001 (-0.011 - 0.011)	-0.002 (-0.008 - 0.004)	0.004 (-0.009 -0.017)	0.003 (-0.002 - 0.008)

*self-reported years exposure to laboratory animals in current job.
WR nasal = work-related nasal symptoms. WR eyes = work-related ocular symptoms. WRRS = work-related respiratory symptoms. Any WRS = any work-related symptom.

5.3.4 Airway inflammation in workers exposed to laboratory animal allergens

Linear regression models were constructed to examine the relationship between log FE_{NO} and the dichotomised exposure variable of more or less than three years' exposure to laboratory animals (Table 29). In workers with more than three years' exposure, GMR FE_{NO} was 63% (95% CI 1.21 – 2.10) higher than in workers with less than three years' exposure. To look for an effect at earlier stages of exposure, the analysis was restricted using more or less than two years' exposure. Similar associations were observed between GMR FE_{NO} and years exposure, with workers with more than two years' exposure having a GMR FE_{NO} 51% (1.01-2.13) higher than those with less than two years' exposure.

When FE_{NO} was dichotomised in logistic regression models, significant relationships were also observed with years' exposure (Table 29). More than three years' exposure was associated with an increased risk of FE_{NO} >40ppb (OR 2.87, 0.83 – 9.93, p=0.09). When using a two-year exposure threshold, this association was stronger (OR 7.75, 0.93 – 64.88, p=0.06), although confidence intervals were wide.

Interestingly, no clear associations between current smoking and FE_{NO} were seen. Both GMR FE_{NO} and risk of FE_{NO} >40ppb were lower in smokers, but neither association was significant (Table 29).

Similarly, neither GMR FE_{NO} nor FE_{NO} >40ppb were significantly associated with atopy in workers exposed to laboratory animal allergens.

Table 29: Adjusted GMR FE_{NO} (unstandardised β and 95% confidence intervals) and FE_{NO} >40ppb (odds ratios and 95% CI) between groups with more than three and more than two years' exposure to laboratory animals. Models were adjusted for sex, ever and current smoking, height, and atopy.

	GMR FE_{NO} (n=127)	FE_{NO} > 40 ppb (n=127)
> 3 years' exposure (yes=1)	1.63 (1.21 - 2.10)**	2.87 (0.83 – 9.93)
> 2 years' exposure (yes=1)	1.51 (1.07 - 2.13)*	7.75 (0.93 – 64.88)
Atopic (yes=1)	1.13 (0.83 – 1.52)	0.68 (0.22 - 2.11)
Smoker (yes=1)	0.96 (0.62 – 1.50)	0.80 (0.15 – 4.40)

GMR FE_{NO} data presented as back transformed $\log_{10}$ FE_{NO} values in units of measurement (unstandardised β and associated 95% confidence interval). FE_{NO} >40ppb data presented as odds ratios and 95% confidence intervals. > 3 years exposure = more than three years exposure to laboratory animals. Atopy defined by positive skin prick tests to more than one common aeroallergen. Smoker = currently smokes cigarettes. *p<0.05, **p<0.01

There were no significant differences in either GM or AM FE_{NO} among workers with and without WRS (Table 30). Similarly, no difference in the prevalence of FE_{NO} >40ppb was found between the two groups.

Table 30: FE_{NO} by presence or absence of any work-related symptom*

	WRS (n=28)	No WRS (n=111)	*P* value
AM FE_{NO}, ppb (SD)	25.94 (20.64)	26.26 (29.92)	0.29
GM FE_{NO}, ppb (SD)	20.26 (2.10)	17.65 (2.41)	0.25
FE_{NO} >40ppb, n (%)	4 (14)	15 (14)	0.92

*Any work-related symptom = any work-related respiratory, nasal, or ocular symptoms

5.3.5 Association between exposure and spirometry in laboratory animal workers

Small but significant reductions in percent predicted FEV_1 and FVC were observed in workers with more than three years exposure to laboratory animals (Table 31). Percent predicted FEV_1 was 3.9% lower in workers with more versus less than three years exposure (unstandardised β -3.90%, 95% CI -7.82 - - 0.02). Percent predicted FVC was 4.4% lower in workers with more versus less than three

years exposure (β -4.41, -8.12 - -0.70). Similarly, absolute FEV_1 and FVC were significantly lower in workers with more than three years exposure, with FEV_1 333 mls lower and FVC 453mls lower in workers with more than three years exposure compared to those with less than three years exposure (for FEV_1 β = -333.48, -579.113 - -87.84; for FVC β = -453.30, -763.18 - -143.42). No effect was seen for PEF.

When years' exposure was used as a continuous variable in linear models, no significant associations were observed between increasing exposure duration and FEV_1 or FVC. However, increasing years' exposure tended to be negatively associated with both absolute and percent predicted FEV_1 and FVC (Table 31).

Table 31: Adjusted linear regression models showing associations between more than years exposure to laboratory animals as categorical (more or less than three years) and continuous predictors for absolute and percent predicted FEV1, FVC, and PEF. Unstandardised β and 95% confidence intervals in parentheses. Al models adjusted for age, gender, smoking and atopy.

	FEV_1, mls (n=128)	**FVC, mls (n=128)**	**PEF, mls (n=128)**
> 3 years' exposure (yes=1)	-333.48 (-579.11 - -87.84)**	-453.30 (-763.18 - -143.42)**	-33.41 (-71.94 - 5.13)
Exposure, years	-15.99 (-38.98 – 7.00)	-17.29 (-46.49 - 11.90)	-0.32 (-3.89 - 3.25)
	% predicted FEV_1 (n=128)	**% predicted FVC (n=128)**	**% predicted PEF (n=128)**
> 3 years' exposure (yes=1)	-3.90 (-7.82 - - 0.02)*	-4.41 (-8.12 - -0.70)*	-1.16 (-6.66 - 4.35)
Exposure, years	-0.25 (-0.61 - 0.11)	-0.20 (--0.55 - 0.14)	0.10 (-0.40 - 0.61)

Atopy defined by positive skin prick tests to more than one common aeroallergen. Smoker = currently smokes cigarettes. *p<0.05, **p<0.01

5.3.6 Relationship between specific sensitisation and nasal, ocular and respiratory symptoms

To explore the degree in which specific sensitisation influenced outcomes, univariate analyses and regression models were stratified by specific sensitisation to mouse or rat epithelium. The 20 people specifically sensitised had significantly higher BMI and had worked longer in their current job. Technicians were over-represented in the sensitised group (30%, versus 12% in the non-sensitised group, p <0.01).

Regression models were constructed to examine associations between specific sensitisation, symptoms, and airway inflammation (Table 32). Sensitised workers were 11 times more likely to report nasal symptoms (OR 11.52, 95% CI 3.62-36.31), nine times more likely to report ocular symptoms (OR 9.60, 2.98-30.96), and 14 times more likely to report respiratory symptoms (14.49, 2.36-89.04) compared to non-sensitised workers.

Table 32: Odds ratios (and associated 95% confidence intervals) from logistic regression models showing associations between specific sensitisation, nasal, ocular, and respiratory symptoms, and airway inflammation.

	Nasal symptoms (n=141)	**Ocular symptoms (n=141)**	**Respiratory symptoms (n=141)**
Specific sensitisation (sensitised = 1)	11.52 (3.62-36.31)**	9.60 (2.98-30.96)**	14.49 (2.36-89.04)**
	WRNS (n=141)	**WROS (n=141)**	**WRRS (n=141)**
Specific sensitisation (sensitised = 1)	17.41 (5.18-58.52)**	16.75 (4.64-60.54)**	37.94 (3.47-414.77)**
	FE_{NO} >40 ppb (n=127)	**LAA (n=141)**	
Specific sensitisation (sensitised = 1)	3.36 (0.94 – 12.02)	14.95 (2.35 – 95.32)**	

Controlled for age, current smoking, height, sex, and years of current exposure. No other predictors were significant. WRNS = work-related nasal symptoms. WROS = work-related ocular symptoms. WRRS = work-related respiratory symptoms. *p<0.05, **p<0.01

Similar associations were observed for work-related symptoms. Sensitised workers tended to have a higher risk of significant airway inflammation versus non-sensitised workers: OR for FE_{NO} >40ppb was 3.36 (0.94-12.02, p=0.06) among sensitised participants. Specific sensitisation was strongly associated with risk for LAA (OR 14.95, 2.34 – 95.32, p <0.01).

5.3.7 Intersections between airway inflammation, airflow obstruction and respiratory symptoms

In order to evaluate the interactions between respiratory symptoms, airway inflammation (FE_{NO} >40ppb) and obstructive spirometry (FEV_1/FVC <LLN), an intersecting Venn diagram was constructed (Figure 17). A total of 41 participants were included in the analysis: 19 with any respiratory symptoms, 22 with airway inflammation, and three with airflow obstruction. There was little association between the three groups. Only two (5% of total) participants with respiratory symptoms had evidence of airway inflammation. Only one symptomatic worker had airflow obstruction. No workers with airway inflammation also had airflow obstruction.

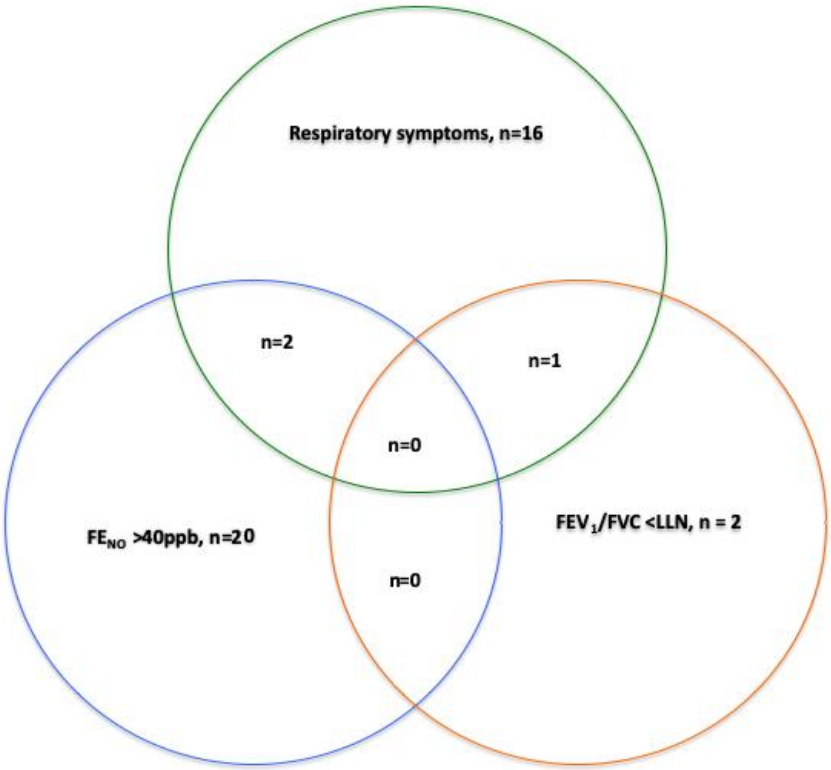

Figure 17: Venn diagram showing intersections between groups with either respiratory symptoms, airway inflammation or obstructive spirometry. In total 41 workers participants were included in the analysis.

Similarly, little association was observed between those with WRRS, airway inflammation, or airflow obstruction (Figure 18). Twenty-eight participants were included in the set analysis: five with WRRS, 22 with airway inflammation and three with airflow obstruction. Two (7%) of participants with WRRS had airway inflammation. There was no overlap between the other groups.

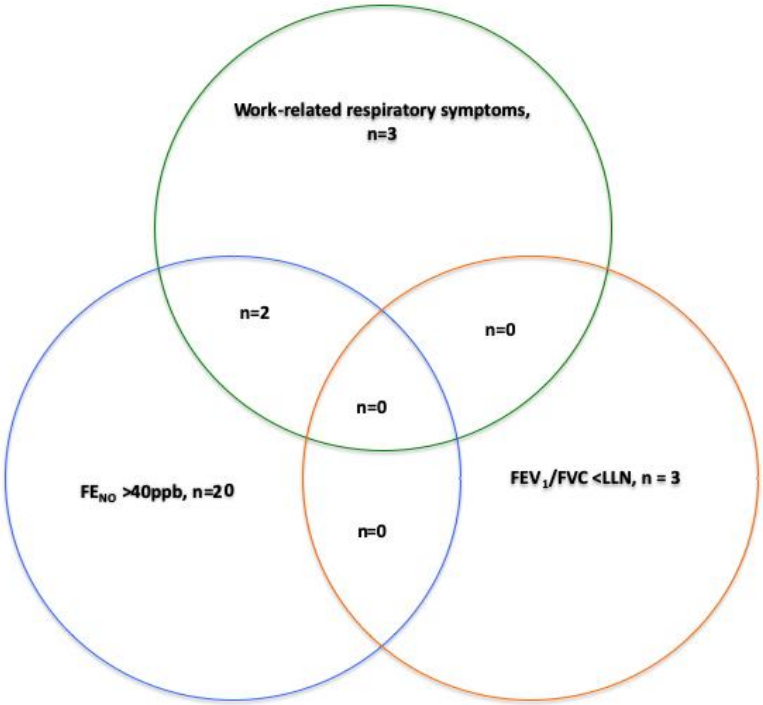

Figure 18: Intersecting Venn diagram showing relationships between participants with work-related respiratory symptoms, airway inflammation, and airflow obstruction (FEV$_1$/FVC <LLN).

5.4 Discussion

This study has demonstrated that more than three years' exposure to laboratory animal allergens is associated with significantly higher GMR FE$_{NO}$ and lower percent predicted FEV$_1$ and FVC. Risk of symptoms was highest in laboratory animal workers who were sensitised to mice. In contrast, no clear associations were seen between increasing duration of exposure and self-reported respiratory symptoms, asthma, or LAA. Among laboratory animal workers, atopy is a significant modifier of nasal and ocular symptoms, with atopic workers significantly more likely to report both work-related nasal and work-related ocular symptoms.

5.4.1 Limitations

Since case-referent studies have shown the majority of individuals who develop symptoms and sensitisation in LAA are most likely to do so within the first three years of exposure, the primary

analysis in SPIRAL was limited to those with less than three years' exposure to laboratory animal allergens (215). Exposure to laboratory animal allergens was measured through static and personal monitoring, but individual exposures were not assigned to the study cohort for analysis. This made it difficult to model allergen exposure along with measures of airway inflammation or spirometry in this study.

Cross-sectional studies are at risk of exposure misclassification as they may fail to capture individuals moving from a high to low exposure environment, or retiring from the industry altogether (142). Similarly, it may be difficult to accurately account for historical exposures in a cross-sectional study. Since both duration and intensity of exposure to laboratory animal allergens are known to be important in the development of LAA, misidentifying historical exposures is important when considering current risk of disease (208). However, years of employment has previously been shown to be a marker of risk for LAA and its use has been reported elsewhere (218, 219).

The main aim of SPIRAL was to determine whether work in IVC-only facilities and associated exposure reduction has led to a reduction in laboratory animal allergy. Individuals with previous exposures to laboratory animal allergens in other facilities were excluded from the primary analysis in SPIRAL. Many researchers will spend time in multiple facilities during their doctoral and post-doctoral training; excluding those with previous animal exposures could have led to exclusion of some older workers with higher historical exposures, thus attenuating any exposure-response observed (207). This is reflected in the findings of the current study where we compared workers with more rather than less than three years' exposure and found that workers with more than three years' exposure had increased airway inflammation and poorer age-adjusted spirometry.

The present analysis of a subset of participants from SPIRAL may have introduced selection bias. It is possible that the limited 12-month sampling frame led to sampling differences in workplace

exposures. Indeed, the prevalence of workers employed in IVC-only facilities reported in the current study was lower than across the whole SPIRAL cohort, at 14% versus 28%. However, rates of work-related respiratory symptoms, specific sensitisation, and LAA were broadly similar in the current study compared with SPIRAL, suggesting this subset is representative of the whole sample (215). It was not feasible to extend lung function and FE_{NO} testing to the larger group of SPIRAL participants, but future examination of measures of airway inflammation and lung function in laboratory animal workers, particularly those with lower exposures, will help to understand how useful these tools are in detecting LAA or OA in these populations.

The majority of participants in the current study were scientists rather than laboratory technicians. Scientists have a more limited role in animal husbandry and culling and therefore exposures among this group may be lower than in other laboratory animal workers (207). Furthermore, the gender, smoking habits, and educational status of scientists differed significantly from technicians in our study, with scientists being more likely to be women, less likely to smoke, and having higher educational attainment. These factors may have influenced FE_{NO} measurements, which are known to be modified by gender and smoking (47).

The current study was powered to detect significance in odds ratios at a mean level of exposure of 1.26 $\mu g/m^3$. However, exposures in SPIRAL were lower than this which may have underpowered the current results. However, exposure measurements were made on a convenience basis and did necessarily include high-exposure tasks, for example cleaning IVC filters or cages (209). Data was sparse for some of the outcomes of interest, for example for specific sensitisation or current asthma. This lack of data reflects findings from the whole SPIRAL cohort where formal diagnoses of LAA were rare. However, despite difficulties in comparing groups due to small numbers, the overall reduction in prevalence of LAA, relative to previous studies, is likely to reflect reductions in exposure in animal units (215).

5.4.2 Work-related symptoms, allergy and sensitisation are related to atopy but not to duration of allergen exposure

Prevalence of work-related symptoms in the current study was low, with only 3% of the study population reporting any WRRS. Furthermore, only 3% of the current study population reported a current diagnosis of asthma, and only 5% self-reported LAA. Previous studies of laboratory animal workers have demonstrated a higher prevalence of work-related symptoms and asthma. Cullinan *et al.* reported work-related symptoms in 29% of their cohort (218). In their study of 225 laboratory animal workers, Kruize *et al.* reported rates of work-related rhinitis in 14% and work-related asthma in 8% (220). This suggests the introduction of IVCs and subsequent reduction in exposure to laboratory animal allergen has led to a reduction in WRRS and asthma prevalence.

No significant relationships between increasing years' exposure to laboratory animal allergens and upper, lower, or work-related respiratory symptoms were identified in the current study. Work-related nasal and ocular symptoms were more common among those with over three years' exposure to laboratory animals, whereas work-related respiratory symptoms tended to be less common, although neither finding reached statistical significance. In addition, linear regression models demonstrated no clear association between years' exposure to laboratory animals and respiratory symptomrs or asthma. Since most cases of LAA occur within the first three years' exposure, a higher prevalence of work-related symptoms or asthma may be expected in those with shorter exposures. Workers with less than three years' exposure were more likely to have worked in mixed facilities and had spent a shorter time in their current job. Individuals who develop symptoms or sensitisation early after exposure to laboratory animal allergens may leave the workplace, leading to a healthy worker survivor effect among the current study population (146). A healthy worker hire effect may also pre-select workers with allergic tendencies or asthma out of employment (197). Furthermore, other factors are thought to be important in the development of LAA. Stable moderate exposures and variable high-level exposures are associated with distinct immunological responses in

laboratory animal workers, with evidence to support an attenuation of LAA at higher exposures (221). This may explain the absence of an association between years' exposure and either respiratory or work-related respiratory symptoms observed in the present study.
Similarly, asthma and LAA were not associated with exposure in logistic regression models. The current study population had spent an average of 5.22 years exposed to laboratory animals. Since symptoms of LAA most commonly occur within the first three years of exposure, the relative absence of LAA among our study population could be explained by a survivor effect. Improvements in working conditions and a reduction in exposure to laboratory allergen have been described over the last decade, and a recent cohort study has demonstrated declining rates of LAA over the last 15 years (222). Detecting significant changes in disease prevalence across small exposure thresholds is challenging and larger studies of exposed populations are needed to more accurately characterise exposure thresholds that cause clinically apparent disease.

Atopy was significantly associated with upper airway symptoms, respiratory symptoms and sensitisation among laboratory animal workers. Atopic workers were more likely to report both work-related and non-work-related nasal and ocular symptoms compared to non-atopic workers. Similarly, atopic workers were significantly more likely to be sensitised to mouse or rat than non-atopics (OR 7.79, 95% CI 2.26 – 26.83). This finding reflects previously reported evidence that atopy is a significant risk factor for nasal, ocular, respiratory and work-related symptoms in laboratory animal workers (9, 216) Atopy was associated with around a three-fold risk increase for ocular, nasal, or respiratory symptoms in controlled analyses. However, significant interactions between atopy and years' exposure were not identified, findings that have been supported elsewhere (216). Atopic workers with HMW exposures have been reported to be at increased risk of OA compared to non-atopic workers (9). However, individual risk of developing LAA varies significantly and these findings do not support workforce selection on the basis of atopic status (207). When considering

risk reduction for LAA emphasis should be placed on modifiable risk factors, the most important of which is allergen exposure.

5.4.3 Specific sensitisation is a key risk factor for symptoms and LAA

The current findings suggest specific sensitisation to mouse epithelium is strongly associated with nasal, ocular, and respiratory symptoms, as well as LAA. Risk of work-related nasal, ocular, and respiratory symptoms was significantly higher in sensitised workers. Similarly, risk for LAA was higher among sensitised workers, although confidence intervals were wide. Previous studies have demonstrated a strong relationship between specific sensitisation and both work-related symptoms and the development of LAA (216, 217). However, there was no clear association between specific sensitisation and asthma in the current study. No sensitised workers self-reported a current diagnosis of asthma despite a number of sensitised individuals reporting work-related symptoms who were also taking asthma inhalers. The pressures of undertaking research in which animal models are a requisite part may make it impossible for PhD students or scientists to report symptoms. Although the study was confidential, there may have been reporting bias in the current study. Individuals may under-report symptoms if they do not recognise that they are work-related or because they are concerned about symptoms negatively influencing their job (15). This demonstrates the importance of testing other than symptom questionnaires in the screening for LAA.

5.4.4 Airway inflammation is related to longer duration of exposure in laboratory animal workers

The current study suggests longer exposure is associated with increasing airway inflammation even after considering known confounders such as atopy, smoking, and sex. GMR FE_{NO} was 63% higher in workers with more versus less than three years' exposure to laboratory animals (GMR FE_{NO} 1.63ppb, 95% CI 1.21 - 2.10, $p<0.01$). These findings suggest that increasing duration of exposure to

laboratory animal allergens is associated with increasing airway inflammation. A number of studies have looked at FE_{NO} in laboratory animal workers. Adisesh and colleagues reported a mean exhaled NO of 6.08 (95% CI 4.58–8.07) ppb in asymptomatic laboratory animal workers versus 12.53 (6.50–24.14) ppb in those with early LAA (workers who were sensitised and with work-related symptoms) (44). This reflects the current findings, where the risk of FE_{NO} >40ppb was higher among sensitised workers. Hewitt and colleagues reported GM FE_{NO} of 17.7ppb in the whole study population and significantly higher FE_{NO} in two sensitised workers (213). Overall, FE_{NO} in the current study was similar at 18.15 (GSD 2.35) ppb, with GM FE_{NO} in sensitised workers 20.52 (GSD 2.84) ppb. This suggests FE_{NO} levels have not changed significantly over a decade, in spite of significant improvements in exposure and an apparent reduction in symptoms and asthma prevalence.

We found no difference in FE_{NO} among participants with work-related symptoms. Furthermore, set analysis showed little overlap between airway inflammation, lung function, or respiratory symptoms. This contrasts to data reported previously in this thesis, where there was more overlap between respiratory symptoms and airway inflammation in wood and foundry workers. This is surprising given the relationship between HMW exposures (such as laboratory animal allergens) and eosinophilic airway inflammation is better established (40). A study comparing airway inflammation in apprentice bakers and hairdressers reported higher levels of physician-diagnosed asthma in bakers (with HMW exposures) with high FE_{NO} compared with hairdressers (124). The same study reported little overlap between airway inflammation and airflow obstruction. Evaluating airway inflammation from both LMW and HMW exposure environments should help to understand the determinants of FE_{NO} in these settings, as well as further explore its use as a screening tool in workplace environments.

Cohort evidence has suggested a significant increase in FE_{NO} in sensitised laboratory animal workers after six and 12 months exposure, with the difference disappearing at 24 months (219). Palmberg *et*

al. reported similar overall levels of exhaled NO in their longitudinal study of newly exposed workers, though reported a significant difference in exhaled NO only among sensitised workers at six and 12, but not 24, months. GM FE_{NO} among those with more than three years' exposure was significantly higher than those with fewer than three years' exposure (22.18 versus 13.83 ppb, $p<0.01$). Although the current study was unable to prospectively evaluate FE_{NO} in the current study, longitudinal studies would be useful to enhance understanding of how FE_{NO} changes over time in laboratory animal workers.

Interestingly, no association between smoking or atopy and FE_{NO} was found among laboratory animal workers. This is surprising as multiple studies have demonstrated both to be important modifiers of FE_{NO}, with airway inflammation increasing in atopic individuals and decreasing in current smokers (46). Rates of current smoking were low in the current study, at only 13%. In contrast, prevalence of atopy was high, with 42% of workers having at least one positive SPT to common aeroallergens. The association between HMW exposures (such as laboratory animals) and Th-2 predominant inflammation in OA is better established than for LMW exposures (9). Atopy may therefore have been expected to play a more significant role in modifying airway inflammation in laboratory animal workers. The absence of such associations may reflect the low overall exposures within the study and support the continued use of IVCs in the prevention of LAA.

5.4.5 Lung function and increasing exposure to laboratory animals

Individuals with longer exposure to laboratory animals had poorer absolute and percent predicted FEV_1, even after controlling for age, sex, and smoking habits. Overall percent predicted FEV_1 was normal, reflecting the young age of the study population. However, a lower FEV_1 in those with longer tenure may reflect excess FEV_1 decline in a population with prolonged or heavier exposure to laboratory animal allergens (219). Excess FEV_1 decline is a feature of OA with ongoing exposures, and

its identification is a key part of health surveillance in order to recognise disease early and reduce or remove exposures appropriately (11).

In addition, increasing years of exposure was associated with poorer percent predicted and absolute FVC. Although we excluded spirometry that failed to meet acceptability criteria, the FVC is more susceptible to technical variation than the FEV_1, which may have contributed to the observed reduction in both FEV_1 and FVC (133). While the main exposures of interest in the current study were animal allergens (primarily Mus-m-1), laboratory workers may also be exposed to other agents harmful to the respiratory tract, either as agents required for animal husbandry (for example cleaning agents or antibiotics) or as part of the research they are undertaking (223). Such agents could attenuate FVC as well as FEV_1, leading to a reduction in both parameters, as observed in the present study.

There was little association between low FEV_1 and either respiratory symptoms or airway inflammation in the current study. Only 2% of the current study population had an FEV_1/FVC LLN, likely due to a number of factors including young age (mean age 29.21 years), low smoking prevalence, and good physical health (mean BMI 24.28 kg/m^2). However, it might be expected those with airway obstruction would have respiratory symptoms. Asymptomatic airflow obstruction can occur in healthy populations and relates to increased exercise-induced dyspnoea and dynamic hyperinflation (224) Such groups require closer follow-up, as airflow obstruction, irrespective of cause, is associated with impaired future health and functional status (225).

5.4.6 Summary

The current study supports a relationship between increasing years' exposure to laboratory animal allergens and increasing airway inflammation, reduced FEV_1 and FVC, but not with an increase in respiratory symptoms or LAA. The finding of lower exposures and disease prevalence is likely to

reflect an overall reduction in LAA associated with reduced exposures secondary to the introduction of IVC-only facilities. Further evidence from SPIRAL will improve understanding of the way in which exposure variation and duration influences the development of LAA in light of changes in laboratory practise. Larger studies are needed to explore the role of FE_{NO} populations of laboratory animal workers, particularly in light of exposure reductions.

6 Fractional exhaled nitric oxide in allergen-exposed workers

6.1 Introduction

6.1.1 Fractional exhaled nitric oxide in the diagnosis of occupational asthma

Fractional exhaled nitric oxide (FE_{NO}) is a simple, non-invasive measure of eosinophilic airway inflammation used in the diagnosis of asthma (4). In OA, rises in FE_{NO} have been reported following positive SIC to both HMW and LMW agents: levels peak at between 24 and 48 hours and show correlation with rises in sputum eosinophilia (182, 183, 226). Increases in FE_{NO} have been reported following positive SIC to latex (227), flour (228), animal dander (229), and isocyanates (230). Challenge studies have suggested such increases are more marked among individuals with low/normal baseline FE_{NO} and a late bronchial response (183). Recent guidance has recommended FE_{NO} as an aid to identifying positive SICs for OA where sputum cytology is unavailable (155). However, evidence from challenge studies remains conflicting, with some studies reporting an increase in FE_{NO} only following HMW exposures (126).

6.1.2 Fractional exhaled nitric oxide in screening for OA

In Britain, health surveillance is mandatory for workers at risk of OA (29). Respiratory questionnaires, spirometry, and where appropriate, specific sensitisation are commonly used tools for evaluating asthma in those at risk (35). However, sensitivity and specificity of both questionnaires and spirometry is low, particularly in early disease (9). Early diagnosis is crucial in OA in order to prevent disease progression and developing more accurate health surveillance programmes is important to improve disease outcomes, prevent job losses, and reduce the economic burden of OA (28). Since FE_{NO} is inexpensive, easy to perform, portable, and increasingly available, it is an attractive potential tool for health surveillance in OA. In their study of bakers and hairdressers, Bohadana *et al.* demonstrated little overlap between apprentices with high FE_{NO} or airway obstruction, suggesting

the two tests provide distinct information on OA risk in such groups (124). In a follow-up study, Tossa *et al.* demonstrated increases in FE_{NO} were significantly associated with incident BHR, with a two-fold increase in risk of developing BHR per log ppb increase in FE_{NO} (41).

In screening studies, FE_{NO} has been associated with increased asthma incidence in spice factory workers (231), bakery workers (42), laboratory animal workers (44), lifeguards (232), and aluminium workers (233). FE_{NO} has also been used to monitor efficacy of exposure reduction, with sustained reductions demonstrated among farmers following educational interventions to reduce workplace exposures (113). However, relatively few studies have looked at relationships between quantitative measures of exposure and FE_{NO} (43, 45, 234). Evidence from other exposure environments is lacking, particularly with reference to known modifiers such as gender, height, cigarette smoking, atopy, age, and ICS use. No studies have explored the modifying effects of atopy or smoking in woodworkers, foundry workers, or laboratory animal workers.

First, this study explores the determinants of FE_{NO} in a population of workers exposed to either HMW or LMW allergens, in particular the degree of influence of smoking and atopy. Second, it evaluates relationships between FE_{NO} and other indicators of OA including current asthma symptoms, work-related respiratory symptoms, and obstructive spirometry. Finally, it aims to understand the degree to which allergen exposure determines FE_{NO}, using quantitative allergen exposures.

6.2 Methods

6.2.1 Study population

In brief, foundry, wood, and laboratory animal workers were recruited over a period of four years (as described in detail in sections 3.2, 4.2, and 5.2). Foundry and wood workers were recruited as part of

HSE's SRP. A subset of workers participating in the SPIRAL study comprised the laboratory animal worker group. Workers in all studies were included if they were currently working in an exposed environment and were over 16. Individuals who were entirely office-based were excluded.

6.2.2 Questionnaire

All workers underwent a validated questionnaire as described in sections 3.2, 4.2, and 5.2 (Appendices B, C and D). Questionnaires detailed demographic information, smoking history, and allergic status; job history; nasal, ocular and respiratory symptoms, and their work-relatedness; and current or past respiratory diagnoses.

6.2.3 Fractional exhaled nitric oxide and spirometry

FE_{NO} and spirometry measurements are described in detail in chapters 3.2, 4.2, and 5.2. In brief, FE_{NO} was performed before spirometry wherever possible, and following a minimum of 24 hours' exposure in the workplace, according to ATS/ERS standards (46). Subsequently, spirometry was performed sitting and without a nose clip, according to ATS/ERS standards (133).

6.2.4 Atopy and sensitisation assessment

Consenting woodworkers underwent serum IgE analysis for TIgE and SIgE to hard and soft wood as described in section 3.2.6. Foundry workers reported current allergy symptoms (hayfever, rhinitis, or asthma) as described in section 4.2.8. SPRIAL participants underwent SPTs for common aeroallergens (house dust mite, grass, cat, dog) as well as specific SPTs for mouse epithelium as described in section 5.2.3. In wood and laboratory animal studies, total IgE and SPT positivity respectively were associated with a significantly increased likelihood of an allergy diagnosis.

6.2.5 Definitions

Work-related nasal symptoms (WRNS), WROS, and WRRS were those that worsened at work or improved away from work or on holiday. Additionally, WRS were defined as those workers with

either WRNS or WRRS. CAS, ever, and current asthma were defined using ECRHS criteria, as described in section 3.2.9.

Workers were considered atopic if they had either: at least one positive SPT to a common aeroallergen (laboratory animal workers), a TIgE above 100 kU/L (wood workers) or reported allergic symptoms such as hayfever and rhinitis within the last 12 months (foundry workers). Current and ever smokers were identified by smoking history and pack years.

A FE_{NO} of at least 40ppb was considered high, and LLN criteria were used to define abnormal spirometry, as described in section 3.2.9.

6.2.6 Data analysis

Only workers with technically acceptable FE_{NO} data and who had complete atopy and smoking information were included in the final analysis. Workers who reported a current respiratory tract infection were excluded (46). FE_{NO} data were not normally distributed so were log transformed and then back-transformed into units of measurement, presented as GM and GSD for descriptive data, and GMR in linear regression models, as previously described in chapter 3.2.10 (45, 139).

Demographic data were compared with univariate analyses and presented as means ± SD or proportions for categorical variables. Univariate analyses were conducted to explore relationships between FE_{NO} and known determinants including atopy, current smoking, sex, age, ICS use, height, and asthma diagnosis. Relationships between key determinants were further explored using adjusted linear and logistic regression analyses. Four groups of workers were created based on atopic and smoking status: atopic smokers, atopic non-smokers, non-atopic smokers, and non-atopic non-smokers (45).

Multiple linear regression was used to examine associations between FE_{NO} and WRS, CAS, current asthma, abnormal spirometry, and HMW or LMW allergen exposure. Multiple logistic regression was used to explore relationships between FE_{NO} above 40ppb with WRS, CAS, current asthma, abnormal spirometry, and HMW or LMW allergen exposure.

In order to explore relationships between FE_{NO} and commonly used health surveillance tools, FE_{NO} was combined with other diagnostic features of asthma to create composite asthma indices and evaluated against other features of OA (100). Composite asthma indices included: CAS and FE_{NO} above 40ppb; CAS and FEV_1 < LLN; and CAS and FEV_1/FVC < LLN. Logistic regression models were constructed to explore their association with other features of OA such as WROS, WRNS, WRRS, self-reported asthma, and exposure to LMW or HMW allergens. All models were controlled for age, sex, atopy, height, and current smoking. Degree of agreement between airway inflammation (FE_{NO} >40ppb) and obstructive spirometry (FEV_1/FVC <LLN) was assessed in symptomatic individuals using intersecting Venn diagrams, as previously described in section 3.2.10.

In order to explore the effect of exposure on FE_{NO}, data were pooled and stratified by exposure to either HMW or LMW allergens. Laboratory animal workers formed the HMW exposure group and foundry and wood workers the LMW exposure group. To evaluate the effect of quantitative exposure measures on FE_{NO}, a 'higher' versus 'lower' exposure group was created. Exposure assessment differed between HMW and LMW groups. SPIRAL used a three-year exposure threshold to determine the primary analysis cohort, whereas in the wood and foundry studies analyses were stratified by quantified allergen exposures. Thus, analysis of FE_{NO} data in 'higher' versus 'lower' exposure environments was restricted to LMW-exposed workers only. Woodworkers in the highest quartile of exposure (section 3.3.1), or foundry workers in the highest tertile of exposure (section 4.3.3), formed the 'higher' exposure group. The remaining participants formed the 'lower' exposure category.

All statistical analyses were conducted using SPSS statistical software, v23 (141).

6.3 Results

6.3.1 Study population

The baseline study group included 773 participants: 351 foundry workers; 269 wood workers; and 153 laboratory animal workers (Figure 19). Thirteen workers refused FE_{NO} measurement or were unavailable during assessment. A further 100 (13%) were excluded as they were unable to perform a reproducible result or reported symptoms of a current respiratory tract infection. Smoking or atopy data was incomplete in five workers. The final study population comprised 655 workers.

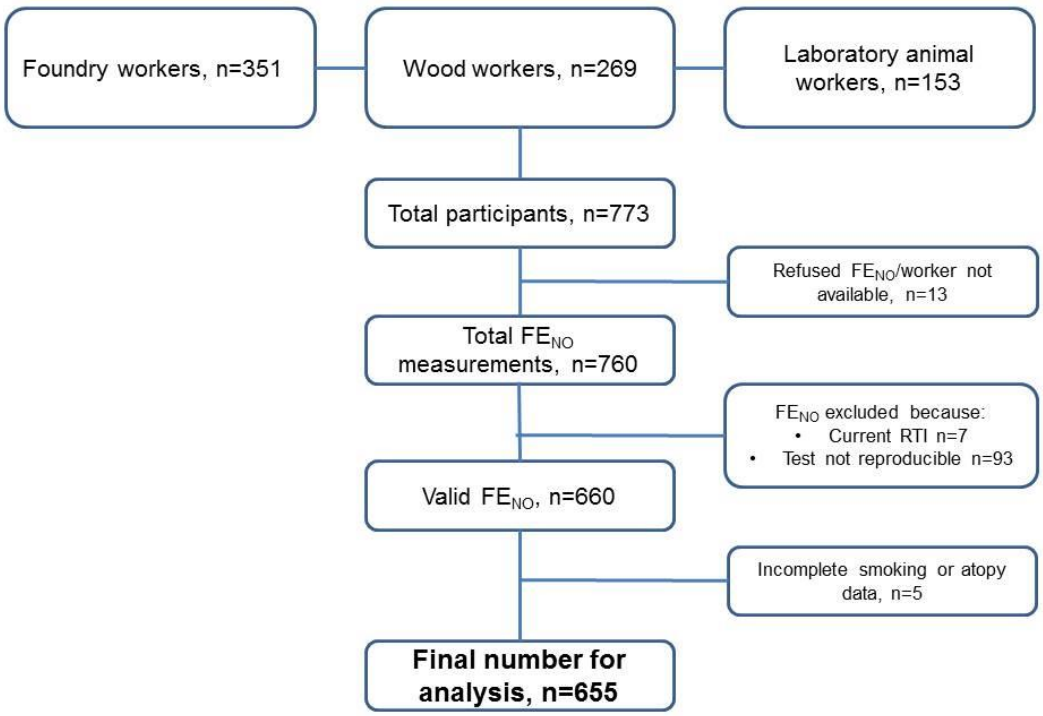

Figure 19: Flow diagram showing participants included from foundry, wood, and laboratory animal studies.

RTI = respiratory tract infection

Subjects excluded from analysis were more likely to be LMW exposed, were older than the study population (43.98 versus 38.96 years, $p < 0.01$), and were significantly more likely to be male (93%

versus 86%, p <0.05). There were no significant differences in smoking status, atopy, clinical symptoms, or asthma among the excluded versus the included groups.

6.3.2 Study demographics

The population demographics are shown in Table 33. Average worker age was 38.96 (SD 12.53) years. The majority (n=565, 86%) were male, with a mean 7.43 (7.80) years spent working in their current job. Overall, symptoms were common, with rates of nasal, eye and respiratory symptoms varying between 22 – 40%, rates of work-related symptoms ranging between 13-16%, and CAS reported by 33%. One-third of workers were atopic, with one-sixth self-reporting ever asthma and one-tenth fulfilling criteria for current asthma. GM FE_{NO} was 17.80 (GSD 2.48). Forty percent of the population had a FE_{NO} >25 ppb and 20% had a FE_{NO} >40ppb.

Rates of smoking and atopy varied significantly between the three groups. Laboratory animal workers were less likely to smoke: rates of current smoking among laboratory animal workers were 13% versus 24% and 30% in wood and foundry workers respectively (p for trend <0.01). However, atopy was commoner among laboratory animal workers compared to wood and foundry workers (41% versus 25% and 36% respectively, p for trend <0.01). Laboratory animal workers differed from wood and foundry workers being younger in age, more likely to be female, and having a lower BMI and height (Table 30). Laboratory animal workers were also less likely to use RPE at work and had spent a shorter time employed in their current job. Laboratory animal workers were less likely to have work-related symptoms or CAS and were less likely to have current asthma. Rates of asthma were very low in laboratory animal workers at 1%.

There was no difference between AM or GM FE_{NO} between the three groups. However, more foundry workers had a FE_{NO} above 40ppb (25% versus 18% and 13% in wood and laboratory animal workers respectively, p for trend <0.05). Similarly, foundry workers were more likely to have a FE_{NO}

above 25 ppb. There was no difference in percent predicted FEV_1, FVC, or PEF between the three groups.

Table 33: Key demographic, exposure and health characteristics of the study population by occupation

	Woodworkers (n=225)	Foundry workers (n=295)	Laboratory animal workers (n=135)	Total (n=655)
		Demographics		
Age, years (SD)	41.23 (12.23)	41.91 (12.23)	28.75 (7.47)**	38.96 (12.53)
Sex, m (%)	218 (97)	294 (99)	53 (40)**	565 (86)
Current smoker, y (%)	54 (24)	89 (30)	18 (13)**	161 (25)
BMI, height m^2/weight in kg (SD)	26.88 (4.88)	28.25 (4.49)	23.83 (4.27)**	26.91 (4.87)
Height, cm (SD)	176.17 (8.26)	176.13 (6.47)	168.48 (8.89)**	174.64 (8.21)
		Exposure		
Currently uses RPE, y (%)	172 (76)	216 (73)	64 (47)**	452 (69)
Time exposed in current job, years (SD)	7.53 (6.87)	8.41 (9.23)	5.13 (4.82)*	7.43 (7.80)
		Health		
Any WRUAS, n (%)	56 (25)	73 (25)	27 (20)	156 (24)
WRRS, n (%)	23 (10)	60 (20)	5 (4)**	88 (13)
Any WRS, n (%)	52 (23)	86 (29)	25 (19)*	163 (25)
CAS, n (%)	108 (48)	134 (45)	21 (16)**	263 (40)
Atopy, n (%)	57 (25)**	107 (36)	55 (41)	219 (33)
Inhaled steroid use, n (%)	19 (8)	22 (8)	4 (3)	45 (7)
Current asthma, n (%)	31 (14)	34 (12)	1 (1)**	66 (10)
AM FE_{NO}, ppb (SD)	27.18 (27.80)	29.33 (27.38)	25.85 (28.02)	27.87 (27.65)
GM FE_{NO}, ppb (GSD)	18.38 (2.47)	20.14 (2.55)	18.03 (2.32)	19.08 (2.48)
FE_{NO} >25 ppb, n (%)	80 (36)	135 (46)*	47 (35)	262 (40)
FE_{NO} > 40 ppb, n (%)	41 (18)	73 (25)*	18 (13)	132 (20)
Percent predicted FEV_1 (SD)	100.17 (13.03)	98.27 (13.47)	100.42 (10.96)	99.35 (12.88)
Percent predicted FVC (SD)	102.98 (14.58)	103.38 (12.22)	104.31 (10.81)	103.42 (12.84)
Percent predicted PEF (SD)	108.49 (16.79)	107.86 (18.29)	108.87 (15.35)	108.28 (17.20)

*p <0.05, **p <0.01

RPE = respiratory protective equipment. WUAS = work-related upper airway symptom (nasal or ocular symptoms). WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms. CAS = current asthma symptoms defined by wheezing, nocturnal chest tightness, breathlessness on exertion, at rest, or at night, or asthma medication use within the last 12 months as per ECRHS. Atopy defined by total IgE >100kU/L or symptoms within 12 months among LMW workers and positive SPTs to common aeroallergens in HMW workers. Current asthma was defined using ECRHS criteria of CAS plus a current or ever asthma diagnosis.

6.3.3 Determinants of FE_{NO} in the study population

Univariate relationships between FE_{NO} and known modifiers including sex, atopy, smoking, height, ICS use, and current asthma are shown in Table 34. Sex, atopy, and current smoking were significantly associated with both GM FE_{NO} and FE_{NO} >40ppb. FE_{NO} was higher among men, atopics, and non-smokers. Height was positively correlated with FE_{NO}. FE_{NO} was higher among current asthmatics compared with non-asthmatics. ICS use was associated with a significantly increased GM FE_{NO} and risk of FE_{NO} >40ppb, with a similar magnitude of effect to a diagnosis of asthma. Contingency table analysis revealed ICS prescription to be highly correlated to a current asthma diagnosis: 85% of individuals taking asthma inhalers fulfilled ECRHS criteria for a current asthma diagnosis, whereas 95% of those not taking an inhaler failed to meet diagnostic criteria ($p<0.01$). There was no significant difference in FE_{NO} between workers with and without WRRS.

Table 34: Relationships between key modifiers including sex, age, atopy, smoking, height, inhaled corticosteroid use, and current asthma with geometric mean FE_{NO} and FE_{NO} >40ppb.

	GM FE_{NO}, ppb (GSD) (n=655)	FE_{NO} >40ppb, n (%) (n=655)
	Sex	
Male (n=565)	19.66 (2.51)	125 (22)
Female (n=90)	15.81 (2.25)	7 (8)
P value	<0.05	<0.01
	Age, years	
	Pearson's' r = 0.045	
P value	0.255	
	Atopy	
Yes (n=219)	23.08 (2.66)	62 (28)
No (n=436)	17.33 (2.35)	70 (16)
P value	<0.01	<0.01
	Current smoker	
Yes (n=161)	11.98 (2.28)	17 (10)
No (n=494)	22.20 (2.29)	115 (23)
P value	<0.01	<0.01
	Height	
	Pearson's' *r* = 0.10	
P value	<0.01	
	ICS use	
Yes (n=45)	28.51 (2.48)	15 (48)
No (n=610)	18.69 (2.47)	117 (19)
P value	0.01	<0.01
	Current asthma	
Yes (n=66)	24.53 (2.38)	24 (36)
No (n=589)	18.55 (2.49)	108 (18)
P value	<0.05	<0.01

Adjusted linear and logistic regression analyses were performed to further explore the determinants of FE_{NO} (Table 35). In linear models, smoking had the most influence on FE_{NO} with current smoking reducing GMR FE_{NO} by almost half (GMR FE_{NO} 0.53, 95% CI 0.45 -0.62). Atopy increased GMR FE_{NO} by approximately a quarter (1.28, 1.11 - 1.47). Non-significant associations were observed for sex. In logistic models, current smoking, sex, and atopy were associated with a FE_{NO} >40ppb. Among current smokers, risk of significant airway inflammation was less than half that of non-smokers (OR 0.35, 95% CI 0.20 – 0.61). Risk was significantly lower in female versus male workers (0.40, 0.16 – 0.96) whereas for atopic workers (1.87, 1.26 – 2.82) it was significantly increased. Relationships for height were not significant in adjusted models. Adjusted R^2 for both models was 0.12.

Table 35: Adjusted multiple linear and logistic relationships between GMR FE_{NO} and FE_{NO} >40ppb and sex, atopy, and current smoking.

	GMR FE_{NO}, ppb	FE_{NO} >40ppb	*P* value
Female sex (female =1)	0.83 (0.66 - 1.04)	0.40 (0.16 – 0.96)	GMR FE_{NO} p=0.11 FE_{NO} >40 ppb = <0.05
Atopic (atopic =1)	1.28 (1.11 - 1.47)	1.85 (1.22 – 2.79)	GMR FE_{NO} p<0.01 FE_{NO} >40 ppb p<0.01
Current smoker (yes =1)	0.53 (0.45 -0.62)	0.35 (0.20 – 0.62)	GMR FE_{NO} p<0.01 FE_{NO} >40 ppb p<0.01
R^2	0.12	0.12	

6.3.4 Modifiers of airway inflammation stratified by atopy and smoking

Multiple linear and logistic regression models were constructed to explore associations between airway inflammation and clinical parameters including CAS, WRS, WRRS, current asthma, and spirometry across the study population (Table 36). Since rates of specific sensitisation were very low among workers exposed to LMW allergens (<1% in this study), no further analyses considering specific sensitisation were performed. As ICS use was closely related to current asthma diagnosis and was not significant in models, it was not included as an obligate confounder. All models were controlled for sex, age, smoking, height, and atopy.

Eosinophilic airway inflammation was associated with a diagnosis of current asthma among the whole study population (OR for FE_{NO} >40ppb 2.12, 95% CI 1.19 – 3.78). The association for GMR FE_{NO} was also significant (GMR FE_{NO} 1.26, 95% CI 1.01 – 1.58). No significant relationships were observed between FE_{NO} and any of CAS, WRS, WROS or WRRS. There were no significant associations observed between either GMR FE_{NO} or FE_{NO} >40ppb and percent predicted FEV_1, FVC, or PEF.

Table 36: Linear and logistic associations between FE_{NO} and respiratory symptoms, asthma, and spirometry across the study population. Models were adjusted for sex, atopy, and smoking.

	GMR FE_{NO}	FE_{NO} >40ppb	*P value*
CAS (yes=1)	1.02 (0.89 - 1.17)	1.12 (0.74 – 1.68)	GMR FE_{NO} p=0.78 FE_{NO} >40ppb = 0.60
WROS or WRNS (yes=1)	0.97 (0.84 - 1.14)	0.68 (0.42 – 1.11)	GMR FE_{NO} p=0.75 FE_{NO} >40ppb p=0.12
WRRS (yes=1)	1.12 (0.93 - 1.36)	1.09 (0.62 – 1.91)	GMR FE_{NO} p=0.24 FE_{NO} >40ppb p=0.78
WRS (yes=1)	1.02 (0.88 - 1.19)	0.77 (0.48 – 1.24)	GMR FE_{NO} p=0.79 FE_{NO} >40ppb p=0.28
Current asthma (yes=1)	1.26 (1.01 – 1.58)	2.12 (1.19 – 3.78)	GMR FE_{NO} p=0.04 FE_{NO} >40 ppb p=0.01
%FEV_1	1.00 (0.99 - 1.01)	0.99 (0.97 – 1.01)	GMR FE_{NO} p=0.59 FE_{NO} >40ppb p=0.45
%FVC	1.00 (0.99 - 1.01)	1.00 (0.99 – 1.02)	GMR FE_{NO} p=0.88 FE_{NO} >40ppb p =0.98
%PEF	1.00 (0.99 - 1.01)	1.00 (0.99 – 1.01)	GMR FE_{NO} p = 0.73 FE_{NO} >40ppb p=0.89

WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms. FEV_1 = forced expiratory volume in 1 second. FVC = forced vital capacity. PEF = peak expiratory flow.

To further investigate associations between FE_{NO} and features of asthma, data were stratified by atopy and smoking (Figure 20, Tables 37-40). Relationships were analysed for: atopic smokers (n=49, Table 37); atopic non-smokers (n=170, Table 38); non-atopic smokers (n=112, Table 39); and non-atopic non-smokers (n=324, Table 40).

FE_{NO} was most strongly associated with current asthma among non-smoking groups. Among atopic non-smokers, a current asthma diagnosis was associated with an increased risk of FE_{NO} >40ppb (OR for FE_{NO} >40ppb 2.70, 1.17 – 6.26) and a higher GMR FE_{NO} (1.37, 0.94 – 1.99). Current asthma also increased the likelihood of high FE_{NO} among non-atopic non-smokers: OR for FE_{NO} >40ppb among non-atopic non-smokers was 2.64 (0.92 – 7.63, Table 40).

Among smokers, WRS rather than asthma were significantly associated with airway inflammation (Table 39). In non-atopic smokers, WRRS were significantly associated with both an increase in GMR

FE_{NO} (1.94, 1.24 – 3.04) and risk of FE_{NO} >40ppb (OR 4.44, 1.16 – 16.99). Similar associations were also seen for WRS and GMR FE_{NO} in the same group. In atopic smokers, risk of FE_{NO} >40ppb tended to be higher in those with WRRS (Table 37).

Table 37: Associations between FE_{NO} and respiratory symptoms, work-related symptoms and current asthma in 49 atopic smokers. Models were controlled for sex.

	Atopic smokers (n=49)		
	GMR FE_{NO}	***FE_{NO} >40ppb***	***P value***
CAS (yes=1)	0.71 (0.40 – 1.28)	0.41 (0.08 – 2.14)	GMR FE_{NO} =0.25 FE_{NO} >40ppb = 0.29
WROS or WRNS (yes=1)	1.21 (0.63 – 2.32)	2.44 (0.45 – 13.26)	GMR FE_{NO} =0.57 FE_{NO} >40ppb = 0.30
WRRS (yes=1)	1.08 (0.47 – 2.5)	2.32 (0.35 – 15.43)	GMR FE_{NO} p=0.85 FE_{NO} >40ppb = 0.38
WRS (yes=1)	0.72 (0.38 – 1.39)	1.11 (1.82 – 6.78)	GMR FE_{NO} p=0.33 FE_{NO} >40ppb = 0.91
Current asthma (yes=1)	0.85 (0.41 – 1.78)	0.46 (0.05 – 4.39)	GMR FE_{NO} p=0.66 FE_{NO} >40ppb = 0.50
WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms.			

Table 38: Associations between FE_{NO} and respiratory symptoms, work-related symptoms and current asthma in 170 atopic non-smokers. Models were controlled for sex.

	Atopic non-smokers (n=170)		
	GMR FE_{NO}	***FE_{NO} >40ppb***	***P value***
CAS (yes=1)	1.07 (0.81 – 1.41)	0.97 (0.50 – 1.90)	GMR FE_{NO} = 0.65 FE_{NO} >40ppb = 0.94
WROS or WRNS (yes=1)	0.75 (0.56 – 1.01)	0.49 (0.23 – 1.06)	GMR FE_{NO} =0.06 FE_{NO} >40ppb = 0.07
WRRS (yes=1)	0.85 (0.57 -1.26)	0.47 (0.16 – 1.36)	GMR FE_{NO} = 0.41 FE_{NO} >40ppb =0.16
WRS (yes=1)	0.85 (0.63 – 1.15)	0.60 (0.27 – 1.30)	GMR FE_{NO} = 0.29 FE_{NO} >40ppb= 0.19
Current asthma (yes=1)	1.37 (0.94 – 1.99)	2.70 (1.17 – 6.26)	GMR FE_{NO} = 0.10 FE_{NO} >40ppb = 0.02
WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms.			

Table 39: Associations between FE_{NO} and respiratory symptoms, work-related symptoms and current asthma in 112 non-atopic smokers. Models were controlled for sex.

	Non-atopic smokers (n=112)		
	GMR FE_{NO}	***FE_{NO} >40ppb***	***P value***
CAS (yes=1)	0.96 (0.65 – 1.41)	1.91 (0.46 – 7.79)	GMR FE_{NO} = 0.83 FE_{NO} >40ppb = 0.37
WROS or WRNS (yes=1)	1.30 (0.84 – 2.01)	0.73 (0.15 – 3.67)	GMR FE_{NO} =0.24 FE_{NO} >40ppb = 0.73
WRRS (yes=1)	1.94 (1.24 – 3.04)	4.44 (1.16 – 16.99)	GMR FE_{NO} <0.01 FE_{NO} >40ppb =0.03
WRS (yes=1)	1.67 (1.11 – 2.51)	2.63 (0.71 – 9.81)	GMR FE_{NO} = 0.01 FE_{NO} >40ppb= 0.15
Current asthma (yes=1)	1.50 (0.82 – 2.72)	0.88 (0.10 – 7.61)	GMR FE_{NO} = 0.19 FE_{NO} >40ppb = 0.91
WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms.			

Table 40: Associations between FE_{NO} and respiratory symptoms, work-related symptoms and current asthma in 324 non-atopic non-smokers. Models were controlled for sex.

	Non-atopic non-smokers (n=324)		
	GMR FE_{NO}	***FE_{NO} >40ppb***	***P value***
CAS (yes=1)	1.10 (0.92 – 1.32)	1.33 (0.73 – 2.41)	GMR FE_{NO} = 0.28 FE_{NO} >40ppb = 0.35
WROS or WRNS (yes=1)	1.01 (0.83 – 1.24)	0.75 (0.36 – 1.58)	GMR FE_{NO} =0.89 FE_{NO} >40ppb = 0.73
WRRS (yes=1)	1.02 (0.78 – 1.33)	1.09 (0.45 – 2.64)	GMR FE_{NO} =0.87 FE_{NO} >40ppb =0.86
WRS (yes=1)	1.02 (0.84 – 1.24)	0.67 (0.32 – 1.41)	GMR FE_{NO} = 0.86 FE_{NO} >40ppb= 0.30
Current asthma (yes=1)	1.19 (0.82 – 1.73)	2.64 (0.92 – 7.63)	GMR FE_{NO} = 0.36 FE_{NO} >40ppb = 0.07
WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms.			

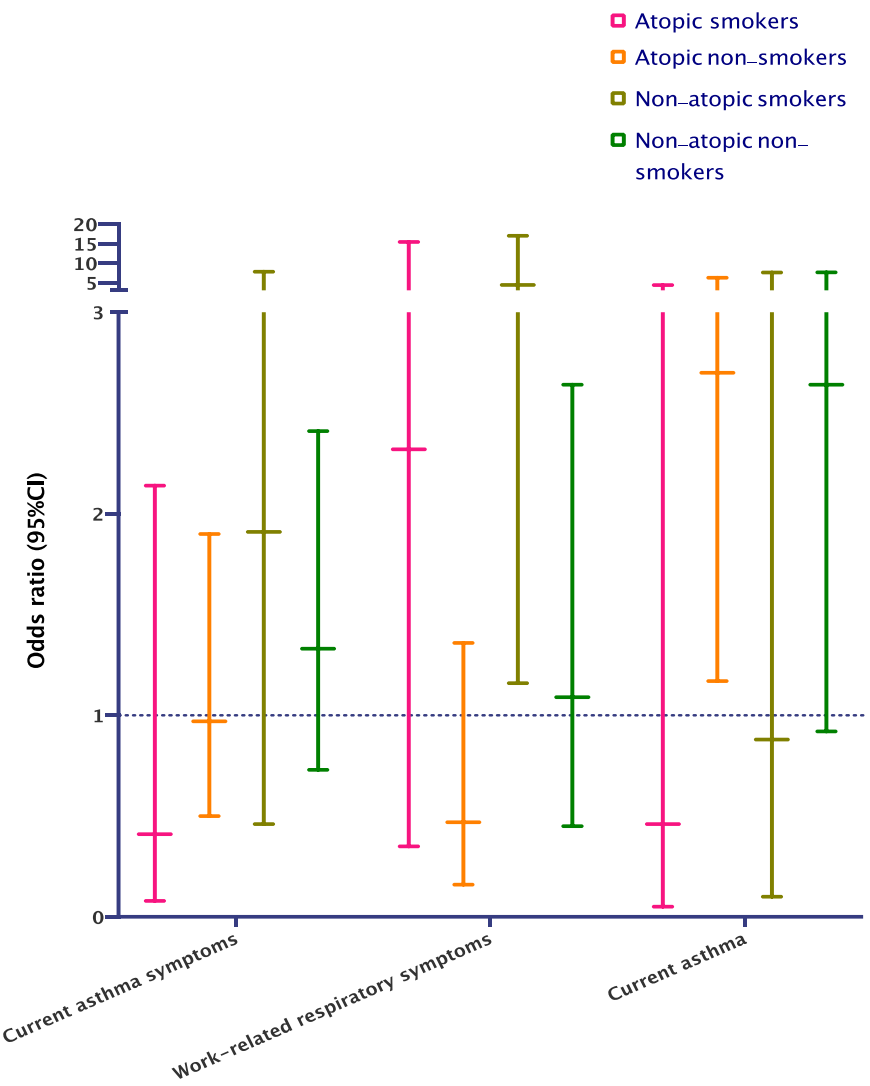

Figure 20: Box and whisker plot showing odds ratios (95% confidence intervals) for FE_{NO} >40ppb in workers with current asthma symptoms, work-related respiratory symptoms, and current asthma, stratified by atopy and smoking.

6.3.5 Association of airway inflammation with grouped asthma variables

Table 41 shows the prevalence of work-related symptoms and asthma split by asthma indices - CAS plus airway inflammation (FE_{NO} >40ppb) or CAS plus abnormal spirometry (FEV_1<LLN, or FEV_1/FVC<LLN). Nasal or ocular symptoms, WRS, and WRRS were more common among workers with CAS and FE_{NO}>40ppb compared to those without. Asthma was significantly more prevalent in workers with either CAS and an FEV_1<LLN and CAS and FEV_1/FVC <LLN versus those without. Asthma prevalence was 42% in those with CAS and FE_{NO} >40ppb versus 32% in those with CAS and FEV_1 <LLN

and 35% in those with CAS and FEV_1/FVC <LLN. The majority of workers in all three groups were exposed to LMW allergens.

Table 41: Prevalence of work-related symptoms, respiratory symptoms, asthma, and exposure type in workers with a combination of current asthma symptoms and either airway inflammation (FE_{NO} >40ppb), an FEV_1 <LLN, or FEV_1/FVC <LLN.

	CAS+FE_{NO} >40ppb, n=57	CAS+FEV_1 <LLN, n=19	CAS+FEV_1/FVC <LLN, n=26
WROS or WRNS, n (%)	17 (30)	4 (21)	5 (19)
WRS, n (%)	25 (44)	3 (16)	5 (19)
WRRS, n (%)	19 (33)	2 (11)	4 (15)
Ever asthma, n (%)	24 (42)	6 (32)	9 (35)
Current asthma, n (%)	24 (42)	6 (32)	9 (35)
LMW exposed, n (%)	55 (97)	18 (95)	24 (92)

WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms

Associations between the three composite asthma variables and work-related symptoms, asthma diagnosis, and exposure were explored using multiple logistic regression (Table 42). All models were adjusted for current smoking, sex and atopy. Workers with CAS and FE_{NO} >40ppb were at increased risk of WRS (OR 2.54, 95% CI 1.44 – 4.48) or WRRS (3.75. 2.01 – 7.00). Similarly, workers with CAS and high FE_{NO} were at increased risk for both ever (4.44, 2.43 – 8.12) and current (8.28, 4.33 – 15.86) asthma. Workers with CAS and FE_{NO} >40ppb were significantly more likely to be LMW exposed (5.71, 1.14 – 28.61). The presence of CAS and FEV_1 <LLN was associated with a significantly increased risk of current asthma (3.34, 1.17 – 9.56). CAS and an FEV_1/FVC <LLN was significantly associated with an increased risk of both ever (3.16, 1.3 – 7.67) and current (4.48, 1.80 – 11.16) asthma.

Table 42: Odds ratios for work-related symptoms and asthma across groups of workers with current asthma symptoms plus either FE_{NO}>40ppb, FEV_1<LLN or FEV_1/FVC<LLN. All models controlled for sex, atopy and smoking. Data presented as odds ratios and associated 95% confidence intervals in parentheses.

	CAS and FE_{NO} >40ppb, n=57	*P* value	CAS and FEV_1 <LLN, n=19	*P* value	CAS and FEV_1/FVC <LLN, n=26	*P* value
WROS or WRNS (yes=1)	1.25 (0.68 – 2.31)	0.47	0.75 (0.24 – 2.31)	0.62	0.69 (0.25 – 1.87)	0.47
WRS (yes=1)	2.54 (1.44 – 4.48)	<0.01	0.51 (0.15 – 1.79)	0.30	0.64 (0.24 – 1.75)	0.38
WRRS (yes=1)	3.75 (2.01 – 7.00)	<0.01	0.66 (0.15 – 2.96)	0.59	0.97 (0.32 – 2.93)	0.95
Ever asthma (yes=1)	4.44 (2.43 – 8.12)	<0.01	2.24 (0.80 – 6.28)	0.12	3.16 (1.30 – 7.67)	0.01
Current asthma (yes=1)	8.28 (4.33 – 15.86)	<0.01	3.34 (1.17 – 9.56)	0.02	4.48 (1.8 – 11.16)	<0.01
LMW exposed (yes=1)^	5.71 (1.14 – 28.61)	0.03	6.32 (0.43 – 92.89)	0.18	4.77 (0.50 – 45.27)	0.17

^ = versus HMW exposed workers. WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms

The degree of agreement between airway inflammation and obstructive spirometry among workers with CAS was assessed using contingency analysis and presented using an intersecting Venn diagram (Figure 21). Two-hundred-and-sixty-three (40%) workers had CAS, 132 workers (20%) had high FE_{NO} and 35 (5%) had an FEV_1/FVC <LLN.

Among participants with CAS there was little overlap between those with airway inflammation and those with obstructive spirometry. Fifty-two (15%) had airway inflammation, whereas only 21 (6%) had obstructive spirometry. Only five (1%) symptomatic workers had both a high FE_{NO} and obstructive spirometry. There was no significant difference in prevalence of airway inflammation between workers with and without CAS (Chi-square for difference 0.10, p=0.75).

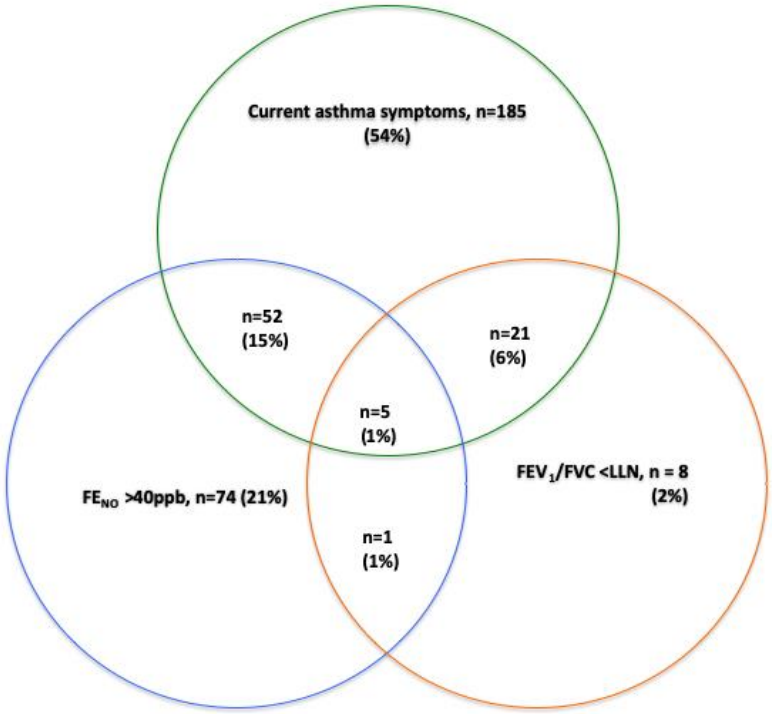

Figure 21: Venn diagram showing contingency table analysis of workers with current asthma symptoms and either FE_{NO} >40ppb or FEV_1/FVC <LLN.

Relationships between WRRS, airway inflammation and obstructive spirometry were explored using contingency analyses (Figure 22). A similar lack of overlap was found between airway inflammation and obstructive spirometry in workers with WRRS: only 1 symptomatic worker (1%) had both airway inflammation and obstructive spirometry. Airway inflammation was found in 18 (8%) of those with WRRS, compared to obstructive spirometry in only 3 (1%) of those with WRRS.

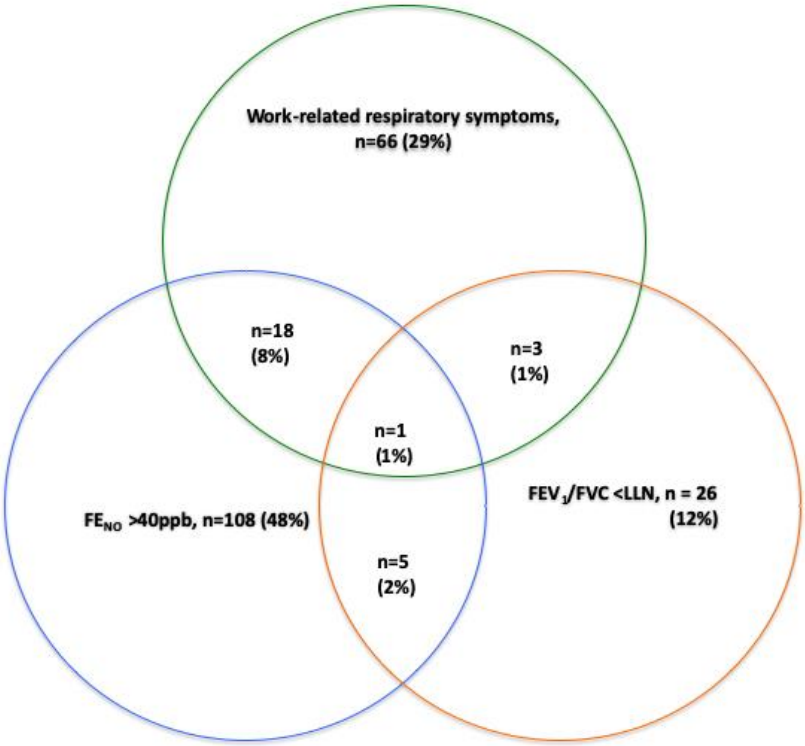

Figure 22: Intersecting Venn diagram showing relationships between WRRS, airway inflammation and obstructive spirometry.

Most workers with CAS and airway inflammation did not have a diagnosis of asthma (Figure 23). Among the 52 symptomatic workers with airway inflammation, 24 (46%) self-reported an asthma diagnosis whereas the remaining 28 (54%) did not. Among 21 symptomatic workers with obstructive spirometry, nine (43%) self-reported asthma whereas the remaining 12 (57%) did not. Among five workers with CAS, airway inflammation and obstructive spirometry, two (40%) had no current asthma diagnosis.

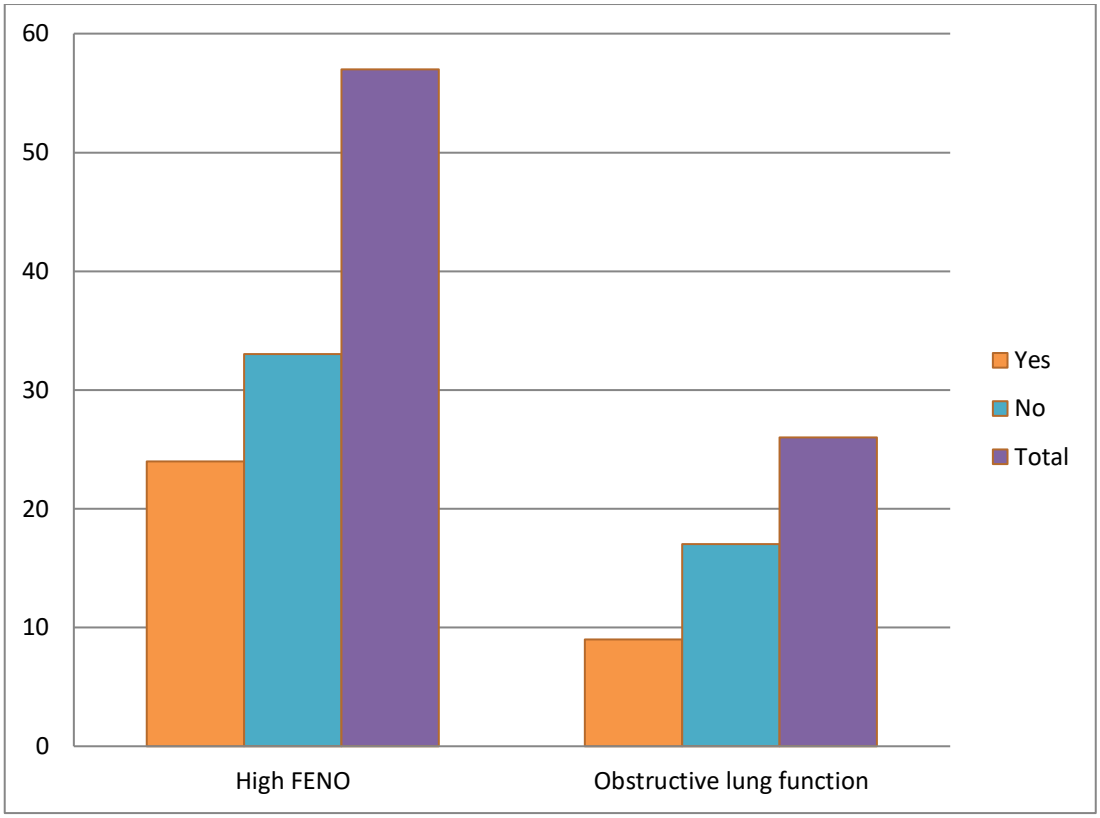

Figure 23: Prevalence of self-reported asthma diagnosis among workers with current asthma symptoms and either airway inflammation or obstructive spirometry. Fifty-two workers had a high FE_{NO}, 21 workers had obstructive lung function, and 5 workers had both.

6.3.6 Effects of exposure on FE_{NO}

In order to further characterise the effect of exposure on airway inflammation, relationships between key modifiers of FE_{NO} (atopy, smoking, sex and current asthma) were stratified by exposure to either HMW or LMW allergens (Table 43). Overall, associations between key modifiers remained for LMW exposures but were less strong for HMW exposures. Current asthma was significantly associated with both GMR FE_{NO} and FE_{NO} above 40ppb in LMW but not HMW-exposed workers. Smoking was strongly associated with both GMR FE_{NO} and FE_{NO} >40ppb in LMW but not HMW-exposed workers, although analyses for FE_{NO} >40ppb were limited among HMW-exposed workers due to low smoking rates. Female sex was associated with significantly lower GMR FE_{NO} in HMW-exposed workers, and a similar trend was observed for LMW-exposed workers although the

relationship was not significant. Atopy was significantly associated with increased GMR FE_{NO} and risk of FE_{NO} >40ppb among workers exposed to LMW but not HMW agents.

Table 43: Relationships between smoking, sex, atopy and asthma with airway inflammation, stratified by exposure to HMW or LMW allergens.

		GMR FE_{NO}	FE_{NO} >40ppb	*P value*
HMW, n=135	**Female sex, yes**	0.74 (0.56 - 1.01)	0.26 (0.09 – 0.76)	GMR FE_{NO} p=0.06 FE_{NO} >40pp, p=0.01
	Atopic, yes	1.08 (0.80 - 1.50)	0.79 (0.28 – 2.28)	GMR FE_{NO} P=0.61 FE_{NO} >40pp p=0.79
	Current smoker, yes	0.98 (0.64 – 1.50)	0.89 (0.18 – 4.47)	GMR FE_{NO} p=0.94 FE_{NO} >40ppb = 0.67
	Current asthma, yes	0.90 (0.17 – 4.93)	0.56 (0.01 – 1.02)	GMR FE_{NO} p=0.91 FE_{NO} >40pp p=1.0
LMW, n=520	**Female sex, yes**	0.58 (0.31 - 1.04)	0.42 (0.05 – 3.53)	GMR FE_{NO} p=0.07 FE_{NO} >40 ppb, p=0.42
	Atopic, yes	1.35 (1.15 - 1.58)	2.45 (1.59 – 3.79)	GMR FE_{NO} p <0.01 FE_{NO} >40 ppb, p <0.01
	Current smoker, yes	0.49 (0.41 - 0.57)	0.34 (0.19 – 0.61)	GMR $FE_{NO\,p}$ <0.01 FE_{NO} >40 ppb, p <0.01
	Current asthma, yes	1.27 (1.01 – 1.59)	1.88 (1.04 - 3.39)	GMR FE_{NO} p=0.04 FE_{NO} >40pp p= 0.04

Associations between FE_{NO} and work-related symptoms, CAS, and asthma were examined by exposure group and stratified by atopy and smoking. Analyses were limited among HMW workers due to small numbers of smokers and the low prevalence of asthma, therefore they were stratified by LMW exposure only (Table 44). Among LMW-exposed workers, observations were comparable to the whole population analyses. The strongest associations were seen for non-smokers, where current asthma was associated with a risk of FE_{NO} >40ppb in both atopic non-smokers (OR 2.82, 1.18 – 6.77) and non-atopic non-smokers (2.60, 0.90 – 7.53). Among smokers, significant associations between current asthma and FE_{NO} were not found. In contrast, smoking groups with work-related symptoms tended to be at increased risk of airway inflammation, although relationships were not significant (OR for WRRS in non-atopic smokers 3.47, 0.85 – 14.22). No significant relationships were

observed among atopic smokers between any of the key independent symptom or asthma variables and FE_{NO} >40ppb.

Table 44: Odds ratios for FE_{NO} >40ppb in workers exposed to LMW allergens, stratified by atopy and smoking

	Atopic smoker (n=38)	*P* value	Atopic non-smoker (n=126)	*P* value	Non-atopic smoker (n=105)	*P* value	Non-atopic non-smoker (n=251)	*P* value
CAS	0.26 (0.04 – 1.66)	0.16	1.07 (0.52 – 1.20)	0.85	1.56 (0.37 – 6.59)	0.55	1.36 (0.72 – 2.55)	0.34
WROS or WRNS	1.77 (0.27 – 11.86)	0.55	0.43 (0.19 – 1.00)	0.05	0.38 (0.05 – 3.15)	0.37	0.82 (0.38 – 1.78)	0.62
WRRS	1.08 (0.10 – 11.32)	0.95	0.42 (0.14 – 1.23)	0.11	3.47 (0.85 – 14.22)	0.08	1.10 (0.45 – 2.70)	0.84
WRS	0.51 (0.05-5.00)	0.56	0.51 (0.22 – 1.19)	0.12	2.15 (0.54 – 8.65)	0.28	0.72 (0.33 – 1.55)	0.40
Current asthma	0.51 (0.05 – 5.00)	0.56	2.82 (1.18 – 6.77)	0.02	0.97 (0.11 – 8.47)	0.98	2.60 (0.90 – 7.53)	0.08

WROS = work-related ocular symptoms. WRNS = work-related nasal symptoms. CAS = current asthma symptoms. WRRS = work-related respiratory symptoms. WRS = work-related nasal or respiratory symptoms

Finally, evidence for a dose-response effect for exposure was sought using stratified analysis. No quantified exposure measurements were available for HMW-exposed workers, so analyses were limited to the 520 LMW-exposed workers (Table 45). One hundred and fourteen workers were higher exposed – either in the highest tertile of foundry exposure or highest quartile of wood dust exposure. GM FE_{NO} was significantly lower in the higher exposed at 16.75 versus 20.83 ppb (p=0.01). FE_{NO} >40ppb was less prevalent in the higher exposed. In adjusted linear regression models, the higher exposed had a lower GMR FE_{NO} (GMR FE_{NO} 0.84, 95% CI 0.72 – 0.98) versus the lower exposed (Table 46). In logistic regression models, risk of FE_{NO} >40ppb was lower in the higher exposed (OR 0.61, 95% CI 0.37 – 0.99).

Table 45: Univariate analyses of FE_{NO} among LMW-exposed workers, by higher or lower exposure.

	Higher exposed (n=114)	Lower exposed (n=406)	*P* value
AM FE_{NO}, ppb (SD)	24.69 (26.14)	30.28 (28.10)	0.03
GM FE_{NO}, ppb (SD)	16.75 (2.53)	20.83 (2.49)	0.01
FE_{NO} >40ppb, n (%)	28 (16)	86 (25)	0.02
The higher exposed group included woodworkers in the highest exposure quartile and foundry workers in the highest exposure tertile.			

Table 46: Linear and logistic regression models for airway inflammation among 520 LMW-exposed workers, stratified by higher versus lower exposure. 114 workers were higher exposed, and 406 were lower exposed.

	GMR FE_{NO}	FE_{NO} >40ppb	*P* value
Higher exposed (lower exposed = 1)	0.84 (0.72 – 0.98)	0.61 (0.37 – 0.99)	GMR FE_{NO} p=0.03 FE_{NO} >40 ppb = 0.05
Models were adjusted for age, sex, smoking, atopy, height, asthma, and RPE use.			

6.4 Discussion

This study is the first of its kind investigating airway inflammation, alongside qualitative exposure measurements, in a large group of wood, foundry, and laboratory animal workers. After controlling for key confounders, both GMR FE_{NO} and FE_{NO} >40ppb were significantly associated with current asthma in the overall population, but in stratified analyses associations remained significant only for non-smokers. Associations between markers of OA, such as WRRS or an asthma diagnosis, were stronger among symptomatic workers with eosinophilic airway inflammation (FE_{NO} >40ppb) versus symptomatic workers with obstructive spirometry. There was little overlap between these groups, and over 50% of symptomatic workers with either airway inflammation or airflow obstruction had no asthma diagnosis. This finding suggests measuring eosinophilic airway inflammation provides additional information to spirometry in symptomatic workers and supports a possible role for FE_{NO} in screening for OA.

6.4.1 Limitations

Both smoking and atopy significantly modified eosinophilic airway inflammation in the current study. Therefore, differences in the definition of atopy and smoking may have influenced associations between FE_{NO} and other outcomes of interest. The majority of participants in the current study had a diagnosis of atopy based on either total IgE or skin prick testing (235). Symptoms were used to determine atopy in foundry workers who underwent neither blood nor skin tests, as in other cross-sectional studies examining relationships between occupational exposures and OA (236). Data from each occupational group was collected according to pre-agreed study designs and approved by an ethics committee, and therefore it was impossible to alter this retrospectively. Relying on individuals' ability to recall atopic symptoms may have introduced reporting bias into the current study (30). Although the prevalence of atopy differed among the three occupational groups included, rates were comparable to those reported in a 2010 systematic review of the UK population, where prevalence in the five included studies ranged from 19 to 54% (151). This suggests a large under- or overestimation of atopy was not present. Furthermore, data previously presented in this thesis (sections 3.3.1 and 5.3.1) supports a strong relationship between atopic symptoms and either a total IgE >100 kU/l or positive SPTs. Since symptom data is often the only information available for practitioners conducting health surveillance, use of such data may more accurately reflect 'real-world' experiences, and therefore investigating relationships between atopic symptoms and OA is relevant to daily practice.

Furthermore, definitions of smoking may have influenced FE_{NO} in the current study. Laboratory animal workers reported sporadic smoking habits, some identifying as current but irregular smokers. The use of current rather than ever smoking in models may have underestimated the effect of smoking in the population, particularly in spirometry models where historic smoking is more relevant to the development of airflow obstruction. However, current smoking is known to influence

FE_{NO} more strongly than ever smoking (46), and its use in models exploring eosinophilic airway inflammation is likely to be more appropriate.

Significant demographic differences existed between the three occupational groups. Laboratory animal workers were approximately 10 years younger than wood or foundry workers, were approximately 10cm shorter, around 50% were female compared to very few female wood or foundry workers, and only 13% were current smokers. This made comparisons between workers exposed to either HMW or LMW allergens very difficult due to the significant influence of such demographic factors on airway inflammation. The demographics of the foundry and wood industry are typical of those seen in British manufacturing and heavy industry: manual workforces have historically been men, with higher smoking rates and greater retention of staff compared with other occupations (237). In contrast, the HMW sample in the current study was drawn from academic institutions where the majority of the participants (81%) were academics undertaking doctoral or post-doctoral work. Since these sample differences are likely to represent differences at a population level rather than sampling bias within the study, matching on a case basis would have been impossible. In future, measuring FE_{NO} in HMW-exposed workers with more comparable demographics to the wood or foundry industry (for example in plant bakery workers) could be useful to examine differences in airway inflammation between HMW and LMW-exposed groups (238).

This study was limited in evaluating an exposure-response across the whole study population. Methodological differences in exposure assessment among laboratory animal workers made it impossible to compare exposure-response relationships across the whole study group. Thus, such analyses were restricted to LMW-exposed workers. In the current study, mean exposures among wood and foundry workers were lower than the current WEL set by HSE (24). Aligning exposures from different work environments is difficult: identifying clinically relevant exposure thresholds is often impossible in OA, even for single allergens (9). Furthermore, exposure assessment does not

account for individual responses to workplace exposures, or for exposure-reduction measures such as use of exhaust ventilation or RPE. Finally, exposure misclassification may occur in cross-sectional studies, which cannot accurately capture past exposures and therefore may underestimate historic exposures if health and safety practices have improved or individuals with allergy or OA have left the workforce due to ill-health. Longitudinal studies with quantitative exposure measures are needed to better quantify exposure-response relationships with airway inflammation in workplaces.

FE_{NO} release is a marker of Th-2 predominant airway inflammation, in which the eosinophil plays a key immunomodulatory role. Th-2 airway inflammation is present in the majority of adults with asthma to environmental allergens (5). The majority of OA is associated with Th-2 inflammation manifested by a spectrum of specific IgE positivity, sputum eosinophilia, and other biomarker increases (10). However, a smaller but significant proportion of OA is non-eosinophilic, with some studies reporting prevalence as high as 40% in LMW-exposed populations (19). Neutrophilic or paucigranulocytic airway inflammation is not associated with an increased FE_{NO}, and symptomatic workers with airflow obstruction may have non-eosinophilic asthma (125, 239). However, prevalence of obstructive airways disease among symptomatic workers in the current study was low. It was not feasible to perform sputum cytology or BAL on participants in the current study, who were recruited and surveyed at their places of work. More detailed analysis of symptomatic workers using additional markers of airway inflammation, infection, or bronchoconstriction, would be interesting in understanding the mechanisms for OA in different exposure environments.

Since the majority of the present study population were exposed to LMW agents (namely isocyanates, formaldehyde, and wood dust), it might be expected that non-eosinophilic airway inflammation would be more likely. However, the present findings do not support this. Foundry and wood workers had a higher GM FE_{NO} and a higher proportion had a FE_{NO} >40ppb compared to laboratory animal workers. In fact, overall GM FE_{NO} levels were higher than have been previously

reported in workers with non-eosinophilic OA. Anees *et al.* reported GM FE_{NO} of 5.1 (interquartile range 3.7 – 6.6) ppb in workers with OA and sputum eosinophils <2%, and 10.4 (5.6–17.4) ppb in workers with OA and sputum eosinophilia (240). In contrast, GM FE_{NO} in the current study was higher at 19.08 (SD 2.48) ppb. Follow-up studies are vital in understanding whether these high levels of baseline airway inflammation translate to future respiratory disease.

A strength of this research is its workplace setting. Undertaking research in a workplace provides the most realistic environment in which to explore associations between OA and workplace exposures. However, quantifying exposures in such research is problematic. For example, though the LMW group were primarily exposed to wood dust, isocyanates, and formaldehyde, workers may have been exposed to a number of other potential irritants or allergens including foundry dust, welding fume, solvents, or glues. Under such circumstances identifying a causative agent for airway inflammation may be challenging. In contrast, FE_{NO} may be useful as a biomarker of exposure as well as disease and be useful as an objective tool to assess exposure and improve workplace controls. Cohort and control studies are needed to assess how exposures are related to FE_{NO}. Such research would be extremely valuable in considering the expansion of the role of FE_{NO} as a biomarker of exposure as well as of airway inflammation.

6.4.2 FE_{NO} in the diagnosis of occupational asthma

Sex, atopy, smoking, height, and inhaler use were key determinants of FE_{NO} in the current study. Even after controlling for confounders, the current study found atopic workers were 80% more likely to have a high FE_{NO} than non-atopic workers, current smokers were 65% less likely to have a high FE_{NO} than never smokers, and men were 60% more likely to have a high FE_{NO} than women. Smoking exerted a greater effect than atopy in linear regression models. Current smoking was associated with a 47% lower FE_{NO} (GMR 0.53, 95% CI 0.45 -0.61) where atopy was associated with a 28% increase in FE_{NO} (GMR 1.28, 1.11 - 1.47). The modifying effects of sex, atopy and smoking on FE_{NO} are well

documented in occupational and non-occupational studies (41, 45-47). Surprisingly, inhaler use was associated with an increased, rather than decreased, FE_{NO} in the current study. FE_{NO} is used to identify steroid responsiveness in some asthmatic populations and to monitor ICS use (241). The current study did not examine inhaler compliance in detail and therefore did not assess whether participants were using their inhaler correctly. Since inhaler use was strongly correlated with asthma in the current study, the positive association between inhaler use and FE_{NO} may have reflected an asthma diagnosis where some individuals were not taking treatment. FE_{NO} may therefore provide objective evidence of treatment compliance and exacerbation risk in occupational populations, which is important in the ongoing management of asthma particularly where populations may still be exposed to the causative agent.

The current study supports a relationship between high FE_{NO} and current asthma (based on self-reported disease and CAS or inhaler use), suggesting that FE_{NO} is related to asthma in an occupational setting. As such, FE_{NO} could be a useful tool in diagnosing new-onset asthma in workplaces. In particular, FE_{NO} may be useful in occupational environments where individuals with asthma may have normal spirometry and other tests for asthma such as bronchodilator reversibility, bronchial provocation testing or sputum cytology are not available. Since early recognition of disease and removal or reduction of exposure is key in OA, the addition of FE_{NO} to questionnaires and spirometry could highlight populations at higher risk for asthma and prompt more regular surveillance or earlier referral to occupational lung disease specialists. This has great potential for improving outcomes in OA, where delays in diagnosis are common and associated with poorer prognosis (11).

However, when analyses were stratified by smoking and atopy the associations between FE_{NO} and current asthma remained only in non-smoking groups. Few occupational studies have reported associations between asthma and FE_{NO} by atopic and smoking status. The findings here reflect those

of Jonaid *et al.* who reported an association between isocyanate exposure and airway inflammation only in non-smoking atopic workers (45). Smit *et al.* described stronger associations between wheeze and FE_{NO} in non-smoking endotoxin-exposed workers (242). Studies in non-occupational cohorts have suggested using reference values for atopic and smoking populations in order to identify those with significant changes in FE_{NO} (47), but this has yet to be adapted in wider clinical practice where a cut-off of 40ppb is more commonly used (4). Interpreting FE_{NO} in workers requires consideration of their smoking and atopic status, and longitudinal studies are needed to identify clinically relevant changes in FE_{NO} in occupational settings that could be useful in the diagnosis of OA.

In contrast, WRRS were significantly associated with FE_{NO} in non-atopic smokers. Among atopic non-smokers with WRRS, risk of high FE_{NO} was four times higher than in those without WRRS (OR 4.44, 1.16 – 16.99). In their analysis of spice mill workers, Van der Walt *et al.* reported a five-fold increased risk of FE_{NO} >50ppb among workers with work-related lower respiratory tract symptoms in adjusted regression analyses (43). Smoking is a risk factor for asthma in some occupational studies and associations between work-related symptoms and airway inflammation in smoking populations may be a sign of early disease (9). Alternatively, airway inflammation in symptomatic smoking populations may be suggestive of early COPD (243). Early recognition of symptoms in smokers may help encourage smoking cessation and would be advantageous for early diagnosis of airway disease, leading to improved health outcomes for the working population (35).

6.4.3 Relationships between allergen exposure and FE_{NO}

Comparing FE_{NO} in HMW versus LMW exposure environments was not the main aim of the current study. However, these findings suggest that FE_{NO} is associated with asthma in LMW-exposed groups even after accounting for smoking and atopy. Although analyses were limited for HMW-exposed workers, in LMW-exposed workers GMR FE_{NO} was 27% (95% CI 1.01 – 1.59) higher in those with

current asthma, and risk of FE_{NO} >40ppb was 1.88 (1.04 - 3.39) higher. LMW-exposed workers were five times more likely to have CAS and FE_{NO} >40ppb versus HMW-exposed workers (OR for CAS and FE_{NO} >40ppb 5.71, 95% CI 1.14 – 28.61). In contrast, significant relationships between current asthma and either GMR FE_{NO} or FE_{NO} >40ppb were not observed for HMW-exposed workers. Previous studies in wood and isocyanate-exposed workers have suggested the role of FE_{NO} may be limited (72). In studies of SIC for OA, FE_{NO} appears to relate more strongly to HMW rather than LMW exposures (40). Indeed, evidence has suggested some workers with OA due to LMW exposures do not manifest an increase in FE_{NO} following a positive challenge. Lemiere *et. al.* suggested such differences could be used to phenotype OA, with LMW-exposed patients having no FE_{NO} increases, lower rates of atopy, increased nonspecific airway hyperresponsiveness, and higher rates of late asthmatic reaction (126). The same study suggested phenotypic differences may be partly related to differing underlying mechanisms for OA secondary to HMW or LMW exposures. The applicability of this in a workplace population, where the majority of individuals do not have asthma, is not established. However, these findings suggest that LMW exposure is associated with an increased risk of high FE_{NO} and cohort studies are needed to establish how FE_{NO} relates to exposure over time, and what proportion of individuals with a high FE_{NO} will go on to develop clinically relevant disease.

Unusually, atopy was found to be more strongly associated with a high FE_{NO} among workers exposed to LMW rather than HMW agents. This contrasts with studies where HMW exposure has been more strongly associated with high FE_{NO}, particularly among atopic workers (42). This finding may reflect overrepresentation of LMW workers among the study population, or differing definitions for atopy between HMW and LMW workers. Atopy has been shown to be associated with high FE_{NO} following positive SIC in some workers with LMW OA (244). Since atopy is a constitutional risk factor for FE_{NO} and therefore unmodifiable, emphasis should be placed on managing modifiable risks such as exposure and, as part of a health surveillance programme, smoking.

Analyses comparing quantitative exposure assessment with airway inflammation were limited to LMW-exposed groups. However, no evidence for an exposure-response effect was found. In fact, workers in the higher exposure group had both lower GMR FE_{NO} and a lower risk of FE_{NO} >40ppb compared with workers in the lower exposure groups. Few studies have evaluated risk of airway inflammation using quantitative exposure measurements, with inconsistent findings. Van der Walt *et. al.* reported significant 24-hour increases in FE_{NO} in spice workers with medium, but not high, particulate exposure (43). Increasing isocyanate exposure has been associated with rises in FE_{NO} in atopic non-smokers (45). Nasal NO has been shown to increase with rising ozone exposures in bleachery workers (234). The absence of a clear exposure-response effect in the current study may be due to a number of reasons: FE_{NO} measurements were made at a single time point, but rises are known to peak 24 to 48 hours from initial exposure (155); exposures in wood and foundry workers were lower than the current UK WEL, and may not have been sufficient to produce a clinically relevant rise in FE_{NO} (42); and the absence of such an exposure-response may be further evidence of a healthy worker effect (146).

6.4.4 FE_{NO} as a health surveillance tool

The current study suggests that FE_{NO} may have a role in identifying a different population of workers at risk for asthma when compared with spirometry. Among workers with symptoms of asthma, 15% had evidence of significant airway inflammation whereas only 6% had airflow obstruction. The degree of overlap between airway inflammation and airflow obstruction was very low at only 1%. The lack of overlap between airway inflammation and airway obstruction reflects previous findings. In their study of apprentice hairdressers and bakers, Bohadana *et. al.* found little overlap between FE_{NO} and obstructive spirometry (124). Additionally, cohort studies have demonstrated sustained decreases in FE_{NO} in farmers with confirmed OA but spirometry within the normal range following a programme of exposure reduction (113). The presence of airway inflammation in symptomatic workers may represent the early signs of clinically apparent asthma (43). FE_{NO} may therefore be

useful in identifying symptomatic workers before they develop airflow obstruction. This is vital in improving outcomes in health surveillance, since established airflow obstruction is associated with poorer long-term outcomes in OA (14).

Workers with CAS and airway inflammation were at significantly increased risk of WRS, WRRS, and asthma. In contrast, only asthma was associated with CAS and abnormal spirometry in analyses, either using FEV_1 <LLN alone or using FEV_1/FVC <LLN criteria. Work-related nasal or respiratory symptoms are used as key referral criteria in health surveillance programmes aimed at identifying respiratory diseases that are caused, or exacerbated, by work (11). Furthermore, in workplaces where 'low-level' health surveillance is used and spirometry not performed, work-related symptoms may be the only evidence to support an occupational component where respiratory symptoms exist (11). Underreporting of symptoms during health surveillance programmes has been demonstrated, with workers having perceived or real fears about divulging work-related symptoms if they believe it may threaten their employment (16, 30). Work-related symptoms related better to FE_{NO} and CAS than to airway obstruction and CAS in the current study. The addition of FE_{NO} to a programme of health surveillance could help to identify workers who are unwilling or unable to report symptoms in order to prompt more active surveillance, evaluation of their work area, or referral to specialist services. This could present an important opportunity to improve outcomes in OA among allergen-exposed workers.

In the current study less than half the symptomatic workers with either airway inflammation or airflow obstruction had ever been diagnosed with asthma. Although some symptomatic workers may have had a falsely positive FE_{NO} (4), or irreversible airways disease such as COPD (13), at least some of these patients could have undiagnosed asthma. Longitudinal studies could provide more information on the risk of developing asthma in those with symptoms and airway inflammation.

6.4.5 Summary

This large cross-sectional analysis of FE_{NO} in wood, foundry, and laboratory animal exposed workers shows FE_{NO} is strongly associated with self-reported asthma, particularly in non-smokers. Key determinants of FE_{NO} include atopy, smoking, and sex, and these must be taken into consideration when interpreting FE_{NO} in occupational studies. There was little association between FE_{NO} and spirometry in the current study and, in symptomatic workers, FE_{NO} was more strongly related to features of asthma including work-related symptoms, current asthma symptoms, and a self-reported asthma diagnosis. This suggests that FE_{NO} provides additional information than spirometry alone in allergen-exposed populations at risk of OA. Further studies are needed to understand how the type and level of allergen exposure influences FE_{NO} in occupational studies, and to describe relevant threshold increases for FE_{NO} that would allow its use as a health surveillance tool.

7 Conclusions

This thesis represents a large cross-sectional study of over 700 workers exposed to occupational allergens in the wood, foundry, and laboratory sectors, and captures information on the prevalence and associations of respiratory symptoms, asthma, airway inflammation and lung function with quantitative allergen exposure. For wood and foundry workers, this represents the first British data on asthma prevalence and spirometry for over two decades. Exposures within the wood, foundry, and laboratory industries have changed significantly over the last 20 years. This research therefore provides important evidence to guide understanding of the current and future risk of asthma in these environments.

The population of workers in which fractional exhaled nitric oxide has been measured is among the largest in published occupational studies. This is the most recent research studying FE_{NO} in wood, foundry, and laboratory animal workers. Very few studies have considered the role of FE_{NO} in allergen-exposed wood and foundry workers, despite persistently high prevalence of OA. Approached from a health surveillance perspective, this research demonstrates consistent associations between a FE_{NO} >40ppb (the threshold used to identify those at high risk of asthma) and self-reported asthma in exposed symptomatic workers. In contrast, associations between obstructive spirometry and self-reported asthma were weaker, and little evidence for overlap between airway inflammation and airflow obstruction in symptomatic wood, foundry, and laboratory animal exposed workers was found. Since most health surveillance programmes for OA rely on symptoms and spirometry to identify workers at risk, any workers with normal spirometry may be missed. Using FE_{NO} as part of a health surveillance programme may help to identify workers earlier in their disease process, before they develop obstructive spirometry, and therefore could improve outcomes in OA.

7.1 Exposure-response relationships in wood, foundry, and laboratory animal workers.

Risk for respiratory symptoms and asthma was not clearly related to increasing exposure to wood dust, foundry (isocyanates or formaldehyde) or laboratory animal allergens. In fact, both work-related symptoms and self-reported asthma tended to be less common among higher exposed groups. There are a number of possible explanations for these observations. Firstly, the healthy worker effect is well described in occupational studies with cross-sectional studies and asthma studies particularly liable to its influence (146). This is likely to account for the absence of disease among higher exposed workers, particularly in the less sterile working environments of the wood and foundry industries. Secondly, exposures in all three industries were lower than those previously reported, and significantly lower than the current UK WELs (24). This exposure reduction may also explain the observed reduction in symptoms. This is an important finding, as a fundamental principle of health and safety legislation and enforcement is identifying and eliminating workplace exposures order to reduce disease risk as low as reasonably practicable (23). Though the observed reduction could be explained by selection bias of workplace with lower exposures, it also suggests such legislation is having some effect. Thirdly, the current work compared higher to lower exposed groups, where, although asthma risk was assumed to be lower, it may not have been as low as in non-exposed populations. Across the population of wood and foundry workers, respiratory symptoms were highly prevalent. This may have limited the effect estimation across exposure groups, masking any exposure-response effect (245). Finally, altered dynamics in the exposure-response relationship may lead to an attenuated response at higher exposures. This phenomenon has been previously described for asthma among laboratory animal workers, and forms part of the rationale for using immunotherapy for allergic diseases (246, 247).

Similarly, relationships between exposure and abnormal lung function were not consistently seen among the populations studied here. Only in laboratory animal workers was increasing years'

exposure related to poorer lung function: quantitative exposures in wood and foundry workers were associated with no change or paradoxical increases in FEV_1, FVC, and PEF. The absence of an exposure-response effect for lung function in a cross-sectional study is not newly described. Workers with poorer baseline lung function are less likely to remain in a job with respiratory irritant or allergen exposure, particularly where there is a risk of OA (146). In addition, spirometry is a relatively insensitive tool when evaluating exposure-response. In healthy individuals, airway dynamics vary over the course of the day and variation in asthmatics may significantly exceed that of normal airways. Spirometry is not recommended as a tool in assessing day-to-day workplace exposures: its use is mainly in longitudinally identifying excess lung function decline (9). As historical allergen exposures reduce with improved health and safety measures, the sensitivity and specificity of spirometry may reduce also. Developing a role for other objective measures of airway disease, such as FE_{NO}, may therefore be beneficial.

7.2 Developing the role of fractional exhaled nitric oxide as a screening tool for OA

Screening for OA entails a number of difficulties. Most health surveillance relies on information from questionnaires plus spirometry and/or sensitisation testing. Symptom questionnaires lack sensitivity and specificity and rely on self-report. Workers may underreport symptoms for a variety of reasons, meaning they are missed at health surveillance (16, 30). Spirometry is the primary objective test used to monitor lung function in populations at risk of OA. It too has issues with sensitivity and specificity and there is a lack of consensus as to a significant FEV_1 decline that may highlight an individual needing enhanced health surveillance or referral to specialist care (35). Furthermore, obstructive lung function (defined with either percent predicted or LLN values) is a late sign in OA, and its use as a prompt for case identification may lead to poorer disease outcomes due to late exposure remediation, delayed referral, and/or delayed treatment of affected individuals (14, 248). Specific sensitisation is not available or not useful in many workers at risk of OA, particularly those

with LMW exposures (35). Since outcomes for OA in the UK remain suboptimal (28), there is considerable scope to improve efforts to prevent, understand, and screen for the condition. However, the use of FE_{NO} as a potential tool for monitoring or diagnosing OA remains controversial. There is a paucity of evidence to support the use of FE_{NO} in workplace settings and its use in the diagnosis of OA in SIC is debated (40). This study presents a significant contribution to understanding how FE_{NO} may be useful in such settings and supports the hypothesis that FE_{NO} is a useful screening tool for OA.

Evidence presented in this thesis suggests FE_{NO} has three potential uses in occupational settings (Figure 24). Firstly, there is evidence that there is scope for its use as a diagnostic tool: in the whole-population analysis, both GMR FE_{NO} and FE_{NO} >40ppb were strongly related to a diagnosis of asthma, particularly in symptomatic workers. Secondly, there is indication it may be of value as a screening tool: in the whole population analysis, the presence of symptoms and airway inflammation was strongly related to work-related symptoms, commonly used for identifying asthma in the workplace. Furthermore, there was little overlap between airway inflammation and airflow obstruction in all three occupational groups studied, suggesting FE_{NO} identifies a group of workers not recognised by spirometry alone. Thirdly, it may be possible to use as an exposure assessment tool: in the current research isocyanate exposure was associated with small but significant increases in GMR FE_{NO}, although in the limited overall analysis a clear exposure-response relationship was not found. These areas require further exploration in order to fully understand the potential benefits FE_{NO} may bring to the management of OA.

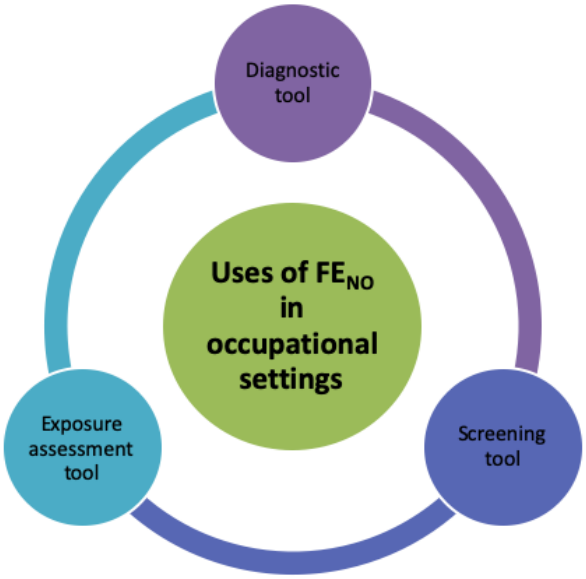

Figure 24: Schematic showing potential uses for FE_{NO} in occupational settings

7.2.1 Fractional exhaled nitric oxide as a diagnostic tool for OA

An asthma diagnosis depends on the presence of asthma symptoms such as wheeze, chest tightness, cough, or breathlessness. No single investigation used for asthma is 100% diagnostic. As such, all investigations are supportive rather than conclusive of a diagnosis, and are most sensitive or specific in the presence of asthma symptoms (4). This study found a FE_{NO} >40ppb was related to a self-reported asthma diagnosis, particularly among non-smokers. Significantly, this association was stronger in the presence of asthma symptoms. These workers may have undiagnosed asthma. A recent study of FE_{NO} and spirometry in children in primary care has suggested that a symptoms-based approach may not be adequate in managing individuals with asthma (249). Recent guidelines from the Global Initiative for Asthma suggest high FE_{NO} is a risk for future asthma exacerbation (3). There is an imperative to diagnose asthma earlier in workers at risk of OA, and FE_{NO} may be helpful in this effort.

Atopy, smoking, and sex were key modifiers of FE_{NO} in the current study. Among LMW-exposed workers atopy and smoking played a significant role in modifying FE_{NO}: among the overall population

associations existed between a self-reported asthma diagnosis and an increasing GMR FE_{NO} or risk of FE_{NO} >40ppb in non-smoking groups, whereas FE_{NO} was associated with an increased risk of WRRS in non-atopic smokers. In the overall analysis, smoking had a greater magnitude of effect than atopy on GMR FE_{NO}, whereas the converse was true for FE_{NO} >40ppb. This finding emphasises the importance of considering determinants of FE_{NO} in its interpretation. Both atopy and smoking are well-documented to alter eosinophilic airway inflammation in both healthy and asthmatic populations, and some authors have suggested using modified reference ranges to take this into account (47). However, international guidance on FE_{NO} suggests it is not practicable to use reference ranges in its interpretation due to known multiple confounders and the significant overlap in 'normal' FE_{NO} ranges between healthy and asthmatic populations (46). Clinically relevant cut-points such as a cut off >40ppb, signifying significant airway inflammation and possible steroid responsiveness, are recommended in the UK (4). Like other physiological tests for asthma, FE_{NO} must be interpreted in an individual context, considering the presence of current respiratory symptoms and/or a history of respiratory disease.

FE_{NO} remains most strongly associated with Th-2 high airway disease. Th-2 high asthma is associated with eosinophilic airway inflammation. FE_{NO} correlates to sputum eosinophilia in both non-occupational and occupational studies, whilst showing little relationship with neutrophilic airway inflammation (125, 250). Not all OA is associated with eosinophilic airway inflammation. Approximately 40% may be associated with neutrophilic airway inflammation, and neutrophilic airway inflammation in more commonly reported following LMW exposures (19).

Interestingly, though the majority of workers were exposed to LMW allergens in the current research, a significant proportion had a FE_{NO} >40ppb, a level strongly suggestive of eosinophilic airway inflammation (46). In fact, LMW-exposed workers were significantly more likely to have a FE_{NO} >40ppb than HMW-exposed workers. Other methods assessing airway inflammation such as

sputum cell counts, cytokine analysis, or BAL cytology, were not practical in this research where data collection was performed in geographically disparate locations and often some distance from laboratories where samples needed to be processed. Therefore, corroborating FE_{NO} with other markers of eosinophilic airway inflammation was not possible, and it was also not possible to rule out non-eosinophilic airway inflammation in symptomatic workers with a normal FE_{NO}. Interpreting FE_{NO} in conjunction with clinical symptoms remains key in identifying workers with likely asthma, irrespective of the underlying immunopathological mechanism, and the absence of a high FE_{NO} in a symptomatic worker with allergen exposure should not deter further investigation.

7.2.2 Fractional exhaled nitric oxide as a screening tool for OA

This research has consistently shown little overlap between FE_{NO} and airflow obstruction in allergen-exposed workers, suggesting a potential additive role for FE_{NO} in screening programmes for OA (Figure 25). Furthermore, effect sizes for predicting work-related symptoms or asthma diagnosis in symptomatic workers were greater for the combination of CAS and FE_{NO} >40ppb than for CAS and airflow obstruction using either FEV_1<LLN or FEV_1/FVC <LLN. Previous studies have reported similar results, with minimal overlap between spirometry and FE_{NO} observed in apprentice bakers and hairdressers (124). One explanation for this is that FE_{NO} identifies workers with airway disease earlier than spirometry. This is an attractive prospect for screening programmes, where the aim is to identify workers as early as possible and to reduce disease-associated morbidity and impact on work.

Measures of airway inflammation may also be helpful in monitoring efficacy of health surveillance programmes. A sustained reduction in FE_{NO} and asthma symptoms has been demonstrated in farmers with OA and normal spirometry, following a programme of exposure reduction (251). Thus, FE_{NO} may be useful in identifying workers with OA in the presence of normal lung function and could

be used to monitor interventions aimed at improving disease outcomes and preventing disease progression.

Figure 25: Schematic showing how FE_{NO} may complement existing health surveillance programmes. There is overlap in the information provided by FE_{NO}, symptoms and spirometry.

Additionally, spirometry and FE_{NO} provide different information on airway status. The former is a measure of airflow obstruction, indirectly related to airway inflammation, bronchial hyperresponsiveness, and airway remodelling (5). The latter is a direct measure of airway inflammation mediated largely through IL-13 and closely related to Th-2 inflammation that often predominates in asthma (125). As such the two tests provide complementary, but different, information on the likelihood of an asthma diagnosis, as well pointing towards the potential

underlying pathophysiology. For FE_{NO} in OA, this difference has important implications when considering the type of exposures and potential mechanisms for disease. Finally, airflow obstruction affects airway dynamics, and may therefore limit the measurable fraction of nitric oxide in exhaled breath. One study of the National Health and Nutrition Examination Survey cohort found COPD to be associated with higher dust exposures and an increased risk of a FE_{NO} below 50ppb, with COPD risks at FE_{NO} >50ppb reduced (252).

Extending FE_{NO} testing into workplaces would require a significant change in current practice for OA surveillance. The introduction of additional tests to health surveillance programmes may help to improve their efficacy, but efforts to increase the provision of health surveillance for OA in the UK are vital in order to best capture those populations at risk and will not be accomplished by the introduction of a new test such as FE_{NO} alone (35). However, improvement in existing screening programmes for OA could make a significant difference to workers at risk. It is therefore vital to provide evidence to support the role of FE_{NO} in such settings, to inform regulators, policy makers, and employers.

7.2.3 Fractional exhaled nitric oxide a measure of exposure to occupational allergens

Allergen exposure is considered a key modifiable factor for asthma (5). In OA, workplace exposures provide the main opportunity to reduce the risk of disease, and much legislative and organisational energy has been focused on achieving this over the last 50 years (9). As such, OA research has often considered relationships between exposure and disease markers. One strength of this research is the quantitative exposure assessment in the wood and foundry workers, allowing accurate evaluation of relationships between exposure and markers of asthma in these groups. To date, only a small number of studies have examined exposure-response between occupational asthmagens and FE_{NO}. Data from this research therefore provides novel information on the relationships between quantitative allergen exposure and airway inflammation.

This research found a positive association between GMR FE_{NO} and isocyanate exposure, and between GMR FE_{NO} and increasing years' exposure to laboratory animal allergens. However, no clear exposure-response relationship was seen between FE_{NO} and wood dust exposure, and when a cut-point of FE_{NO} >40ppb was used clear exposure-response effects were not observed in any of the individual occupational groups studied. In fact, in the overall analysis, LMW-exposed workers in the higher exposure group had a lower risk of high FE_{NO} compared to workers in the lower exposure category. Workers with medium, but not high, exposure to spice dust have previously been found to be at increased risk of FE_{NO} >50ppb (43). Cement workers with increasing years exposure have been found to have higher FE_{NO} than age-matched controls (253). The current research suggests that although increasing exposure to isocyanates may be associated with small increases in GMR FE_{NO}, evidence for increasing allergen exposure causing clinically significant airway inflammation is lacking. Longitudinal studies examining the impact of allergen exposure on long-term risk of airway inflammation are important in clarifying whether FE_{NO} is useful in monitoring workplace exposures.

A strength of this thesis was its workplace design, allowing assessment of airway inflammation in a 'real-world' exposure environment. However, controlling exposures in such studies is difficult. All three exposure populations studied may have had additional allergen or irritant exposures not accounted for here. Potential co-exposures included solvents or paints in wood workers; foundry particulate, silica dust, or heat in foundry workers; and cleaning agents, animal bedding, or research chemicals in laboratory animal workers. Co-exposure to such agents may have played a role in observed increases in airway inflammation. Even with more accurate quantification of exposure, the cross-sectional design of this research precluded the inference of causation. Although FE_{NO} is most consistently associated with Th-2 type inflammation, its exact cellular source remains unknown, and its use is being increasingly explored in other respiratory diseases including eosinophilic bronchitis, COPD, cystic fibrosis, and pulmonary infections (243). In future, FE_{NO} may be useful in understanding

the occupational contributions of exposure to respiratory conditions other than asthma and could be a potentially useful tool in monitoring workplace exposures as well as risk of individual respiratory conditions.

7.3 Using FE_{NO} in workplace studies: suggestions for future research

There are a number of areas in which future research is needed in order to better understand the role of FE_{NO} in allergen-exposed workers. Firstly, understanding reproducibility and validity of FE_{NO} in the workplace is crucial in exploring its use as a diagnostic or screening tool. Evidence reported here demonstrates that FE_{NO} can be performed in different workplaces and is at least as reproducible as spirometry. In 760 workers who underwent FE_{NO} testing, 93 (12%) were unable to perform a valid test compared with 97 (13%) unable to perform valid spirometry. Only one worker reported an issue with the test as they had recently had dental surgery so were unable to perform either FE_{NO} or spirometry measurements. Validation studies of FE_{NO} in workplaces, showing within- and between-worker reproducibility are needed to clarify reproducibility and acceptability criteria.

Secondly, consideration of environmental exhaled nitric oxide is an important potential confounder in occupational studies. This was eliminated in the current research by inhaling through the scrubber before the test is performed, and devices are calibrated to high levels of ambient NO >200ppb (254). However, the impact of higher levels of ambient nitric oxide on FE_{NO} is not known. Environmental conditions will vary hugely between workplaces, and ambient nitric oxide levels may be very high particularly where there is release of gases and fumes (243). The performance of FE_{NO} devices should be evaluated at different levels of ambient NO to ensure that its use is appropriate across the spectrum of occupational environments.

Thirdly, using FE_{NO} in a health surveillance programme would require the identification of clinically significant increases in airway inflammation. The ATS/ERS guidelines suggest an increase of more

than 20% in individuals with a FE_{NO} >50ppb or more than 10ppb in individuals with a FE_{NO} <50ppb is clinically significant, but acknowledge that evidence to support this assertion is weak (46). A number of papers have considered the sensitivity and specificity of threshold increases in FE_{NO} to predict OA following SIC, but few studies have considered threshold increases in FE_{NO} in exposed populations at risk for OA. In their study of apprentice bakers and hairdressers, Florentin and colleagues reported the combination of FE_{NO} greater than 8.5 ppb and a positive clinical questionnaire to have a 79% sensitivity and 80.5% specificity for diagnosing OA (255). Van der Walt *et. al.* found medium exposure to spice particulate led to rises in FE_{NO} of >12% when measured 24 hours from initial exposure (43). In their longitudinal study of bakers and hairdressers, Tossa and colleagues reported FE_{NO} increases of 20% in nonatopic and 16% in atopic workers with incident BHR (41). Due to its cross-sectional design this research was unable to identify thresholds at which FE_{NO} was able to predict WRRS, CAS or asthma. Follow-up of the current study populations will enable better understanding of threshold increases in FE_{NO} at which WRRS, CAS or asthma may be predicted, and provide further information for those considering using FE_{NO} to diagnose or monitor asthma in allergen-exposed workers.

Finally, understanding how FE_{NO} relates to the timing and intensity of allergen exposure is important for it to be useful in monitoring asthma and allergen exposure in workers. SIC studies have demonstrated that FE_{NO} peaks 24 to 48 hours from exposure and levels reduces thereafter (182). FE_{NO} may therefore be more representative of short- to medium-term exposure, and more research is needed to understand how these exposures relate to long-term disease risk of respiratory disease. There is a paucity of workplace studies considering the effect of differing timing or magnitude of exposure on FE_{NO}, for example whether intermittent high-dose exposure exerts a greater effect than chronic low-dose exposure. Occupational cohort studies of FE_{NO} in different exposure environments should be conducted to help improve understanding of how exposures relate to FE_{NO} rises, and whether these are sustained or short-lived. Randomised controlled trials from more general asthma

populations, such as those being conducted to determine whether FE_{NO} is useful for monitoring steroid responsiveness, will also be useful in better understanding how FE_{NO} changes over time.

In summary, this thesis presents new evidence on respiratory symptoms, asthma, and lung function in British wood, foundry, and laboratory animal workers. Novel understanding of the use of FE_{NO} in occupational environments, including its association with symptoms, lung function, and allergen exposure, suggests airway inflammation may be a useful additional tool for diagnosing and screening OA in workplaces, but evidence for an exposure-response effect is lacking. Defining the role of FE_{NO} in occupational environments may provide information not just on OA risk, but also on disease phenotype and airway responses to exposure, and is an exciting prospect for further study.

Appendices

Appendix A: Additional tables and checklist for systematic review in woodworkers

1. *Checklist – studies assessing risk factors in systematic review of British woodworkers*

This set of criteria should be used for assessing studies aimed at identifying the extent to which characteristics or behaviour of a person, an environmental exposure or the characteristics of a disease alter the outcome.

DESCRIPTIVE INFORMATION ABOUT THE STUDY	NOTES	DESCRIPTION
Study identification	Include author, title, reference and year of publication (if available) and the study time frame	
What is the study type?	Cohorts, case control (C-CS)	
What risk factors are considered?		
What outcomes are considered?	e.g. disease, surgical rates, death	
What other factors affect the outcome?	Include potential confounders, demographic characteristics	
What are the characteristics of the population and study setting?	Personal characteristics e.g. sex; disease characteristics of the population.	

	Study setting e.g. rural, urban, hospital inpatient or outpatient, general practice, community.	
EXAMINATION CRITERIA FOR THE STUDY	**COMMENTS**	**CODE OPTIONS** **A, B1, B2, C, ? n/a**
Are the study participants well defined in terms of time, place and person?		
What percentage of individuals or clusters refused to participate?		
Are outcomes measured in a standard, valid and reliable way?		
Are risk factors and outcomes measured independently (blind) of each other?		
Are all important risk factors included in the analysis?		
What percentage of individuals or clusters recruited into the study are not included in the analysis?		
OVERALL ASSESSMENT OF THE STUDY	**COMMENTS**	**CODE OPTIONS** **A, B1, B2, C**

How well (code A, B1, B2, C) was the study done to minimise bias? If code B1, B2, or C, what is the likely direction in which bias might affect the results?		
Include other comments concerning areas for further research, applicability of evidence to target population, importance of study to policy development.		

2. *List of all papers included in the systematic review by publication date.*

Author, date	Study type, Country	Industry, Control population.	No of workers exposed/ controls	Wood type(s) used[a]	Arithmetic Mean exposures (mg/m^3 unless otherwise stated)[b]	Study outcomes [c]	SIGN and MERGE rating	Summary findings
Jacobsen *et al.*, 2013.	Longitudinal. Denmark	Furniture manufacture. Non-exposed controls from same Geographical area	813/136	Softwood	Median ID = 0.96	LF	2+ A	No significant difference in longitudinal lung function between workers and controls
Mohan *et al.*, 2013.	Case control India	Carpentry. Non-exposed controls from other industries.	150/150	-	-	LF	2- C	PEFR in workers with longer tenure significantly lower than those with shorter tenure.
Schlünssen *et al.*, 2011	Longitudinal. Denmark	Furniture manufacture. As per Jacobsen 2013.	1506/195	Softwood, wood composites, less hardwood	ID = 0.55	SZ	2++ A	No significant relationship between IgE to beech or pine and symptoms or asthma. Significantly more IgE positivity in high exposure group.
Boskabady *et al.*, 2010.	Case control. Iran	Carpentry. Non-carpenters from the same residential area.	66/66	-	-	S; LF	2- B2	S: No significant differences between respiratory symptoms in exposed vs controls. LF: Significantly reduced FEV_1, FVC, PEF, and mid expiratory flow measurements between carpenters and controls.
Campo *et al.*, 2010	Cross sectional. Spain	Carpentry apprentices.	101	Alder, cedar, cherry, European beech, iroko, walnut, olive, obeche, pine, oak, sapele	-	S; SZ	2- B1	S: 53% of workers reported respiratory symptoms; 40% reported rhinitis; 18% reported asthma symptoms. SZ: 49% of study group atopic. 7% had skin prick test positivity to wood species.
Gomez *et al.*, 2010.	Cross sectional. Colombia.	Carpentry.	177	Hard and softwood	WD = 1.6 – 61.4	S	2- C	Ocular symptoms most frequent, followed by work-related cough, then shortness of breath.

Pérez-Ríos *et al.*, 2010	Meta-analysis.	Wood dust exposed	19 studies		-	A	1- A	Pooled RR 1.5 for asthma in woodworkers vs gen population
Thetkathuek *et al.*, 2010	Cross sectional. Thailand.	Furniture manufacture. Unexposed office workers	685/?	Rubber tree	TD = 4.08	LF	2+ B1	FVC and FEV_1/FVC negatively correlated with increasing dust exposure
Baran *et al.*, 2009.	Cross sectional. Poland	Furniture manufacture.	70	Softwood	TD = 0.49 – 18.2	LF	2- B2	Lower LF values in longer exposed – no data given
Jacobsen *et al.*, 2009	Longitudinal. Denmark.	Furniture manufacture. As per Jacobsen 2013.	2032/474	Softwood	Baseline ID = 0.9; follow up ID = 0.6	S; A	2+ A	S: significantly increased OR for cough (3.8) and chronic bronchitis (6.0) in female workers in highest vs lowest exposure categories. A: OR for ever asthma (3.4) and current asthma (6.9) in female workers vs controls. For female workers with no baseline symptoms OR for asthma symptoms = 11.3
Osman *et al.*, 2009	Case control. Turkey	Furniture manufacture. Furniture sale workers.	328/328	MDF	TD = 2.04	LF	2- B1	LF among furniture workers significantly worse than controls across all measured parameters.
Sripaiboonkij *et al.*, 2009	Case control. Thailand.	Furniture manufacture. Non-exposed office workers	103/94	Rubber tree	ID = 0.02 - 2.93	S; A; LF	2+ A	S: No significant difference in adjusted OR for different exposure levels A: Exposed vs office workers OR for ever asthma = 6.1. OR for ever asthma in low exposure vs control category = 8.41. No effect across other exposure categories. LF: Incremental FEV_1 and FVC loss significantly more in factory workers vs controls and in highest vs lowest exposed groups.
Glindmeyer *et al.*, 2008.	Longitudinal. USA	Sawmill; plywood manufacture; cabinet and furniture. Between industry controls.	1164	>70% hardwood in cabinet and furniture facilities.	ID = 1.45; RD = 0.18	S; LF	2++ A	S: Across industry groups, self-reported upper respiratory symptoms ranged from 45 – 53%, lower respiratory symptoms from 25-39%, ever asthma from 8.3 –

				>90% softwood in sawmill and plywood				13%, pneumonia from 9.4 – 29%, and COPD from 2 – 4%. LF: Negative effect for respirable residual particulate matter in milling industry. Positive effect for respirable wood solids in sawmill/planing. No effect on LF in furniture/cabinetry and plywood.
Heikkilä *et al.*, 2008.	Longitudinal. Finland	Wet and dry wood industries; boat building and repair; construction. All employed Finns aged 20 – 59.	158,000	Hard and softwoods, particleboard, MDF.	TD = 0.02 – 1.5	A	2++ A	RR 1.5 for both male and female woodworkers vs general population. No increase in risk described with increasing exposures.
Jacobsen *et al.*, 2008	Longitudinal. Denmark	Furniture manufacture. As per Jacobsen 2013.	1819/415	Softwood	Baseline ID = 0.94; follow up ID = 0.6	LF	2+ A	Negative effect on LF for female woodworkers who smoked. Dose response relationship observed among female workers.
Meo, 2006	Case control. Pakistan	Small scale wood industry. Shopkeepers and salesmen	46/46	Teak	-	LF	2- C	Significantly reduced FEV_1 and FVC in woodworkers vs controls.
Arbak *et al.*, 2004.	Case control. Turkey	Furniture decoration students. Students from the same school.	64/62	Fir	-	S	2- B2	Ocular symptoms and rhinitis significantly more common in woodworkers vs controls.
Meo, 2004	Case control. Pakistan	Small scale wood industry. Shopkeepers and salesmen	46/46	Teak	-	LF	2- C	Significantly reduced PEFR in group with ≥8 years exposure vs 4-8 years and ≤4 years.
Priha *et al.*, 2004.	Case control. Finland	Furniture manufacture. Office workers at FIOSH	55/15	MDF	ID (gm) = 1.2 - 1.3; FORM (gm) = 0.1 – 0.17ppm	S	2+ B2	Nasal symptoms more common in MDF workers vs woodworkers and controls.
Schlünssen *et al.*, 2004a	Case control. Denmark	Furniture manufacture. As per Jacobsen 2013.	302/71	Softwood, wood composites, less hardwood	ID (gm) = 0.96	A	2++ A	A: Increased OR for current asthma in highest vs lowest exposure categories (3.3 vs 2.1). Strong association between atopic workers and asthma indices.

Ricciardi *et al.*, 2003	Case control. Italy	- Exposed symptomatic vs exposed asymptomatic vs asthmatic workers	9/10/10	Iroko	-	SZ	2-C	Negative specific IgE to iroko amongst all patients with iroko-induced occupational asthma.
Skovsted *et al.*, 2003	Longitudinal; nested case control. Denmark	Furniture manufacture. As per Jacobsen 2013.	339/142	Softwood, wood composites, less hardwood	-	SZ	2+ B1	55% of participants atopic. No significant difference between pine IgE positivity between atopics vs non-atopics.
Milanowski, 2002.	Case control. Poland	Furniture manufacture. Office workers	48/41	Fibreboard, chipboard	-	S; LF	2-C	S: 79% of workers reported work-related symptoms. 805 of board-processers reported cough. LF: Significant cross-shift reduction in FEV_1 and FVC in under 30s.
Rongo *et al.*, 2002.	Case control. Tanzania	SSWI. Non-exposed office workers	546/565	Variety of African hardwoods, podo, cypress, pine.	ID (gm) = 3.86	S	2+ A	Increased OR for work-related cough, sputum, SOB, wheeze. OR in lower exposure group vs higher exposure group: WHZ = 4.6 (1.9 – 11) vs 1.9 (0.5 – 7.3); SOB = 3.2 (1.6 – 6.3) vs 2 (0.7 – 5.4).
Schlünssen *et al.*, 2002a	Case control. Denmark	Furniture manufacture. As per Jacobsen 2013.	161/91	Softwood, wood composites, less hardwood	ID = 1.17	S	2+ A	VAS assessed nasal obstruction increased with exposures
Schlünssen *et al.*, 2002b	Case control. Denmark	Furniture manufacture. As per Jacobsen 2013.	2033/475	Softwood, wood composites, less hardwood	ID = 1.17	S; A; LF	2+ A	S: OR for cough (2.7), wheeze (2.5), and chronic bronchitis (6.84) higher in women with 2-8 years industry service compared to controls. A: Self-reported and physician diagnosed asthma more common in atopic workers and female workers in medium and high exposure groups. LF: No significant difference observed when stratified by exposure.
Skórska *et al.*, 2002.	Case control. Poland	Furniture manufacture, board processing. Office workers	48/32	Wood composites	-	SZ	2-C	High rates of early and late positivity to fungal species in exposed workers.

Bohadana *et al.*, 2000	Case control. France	Furniture manufacture. 13 males of similar socioeconomic status, 200 historical controls.	114/213	Beech and oak	ID = 4.08 – 12.74	S; LF	2+ A	S: No significant difference in symptoms observed between exposure groups. LF: Excess FEV_1 and FVC observed across exposure groups compared to controls. BHR increased significantly with exposures.
Alwis *et al.*, 1999.	Case control. Australia.	Sawmill, wood chipping and joinery. Maintenance workers from same work sites	195/34	Mostly eucalyptus. Other Australian hardwoods, MDF and WRC also in use.	ID = 0.5 – 15.33; RD = 0.03 – 1.01; TF = 3.34 – 74.06 x 103 cfu/m3; BACT = 2.86 – 25.72 103 cfu/m3; IEND = 0.74 – 21.08 ng/ m3; REND = 0.13 – 2.03 ng/ m3; IBDG = 0.33 -4.78 ng/ m3; RBDG = 0.11 – 0.85 ng/ m3.	S	2- B1	Significantly more cough, sputum, chronic bronchitis and runny nose in joiners vs sawmill/chipmill workers and controls. Exposed to dust associated with increased bronchitis in joiners. Very strong association with ear irritation and bronchitis in joiners exposed to BDG.
Mandryk *et al.*, 1999	Cross sectional. Australia.	Sawmill Kitchen and cabinet manufacture Maintenance workers from same work sites	197/30	As per Alwis *et al.*, 1999.	ID = 0.83 – 15.33; RD = 0.16 – 1.01; END = 0.74 – 21.08 ng/ m3; BDG = 0.33 – 4.63 ng/ m3	S; LF	2+ A	S: Nasal and respiratory symptoms more common in exposed vs control populations. LF: Cross-shift lung function significantly worse in joiners vs controls.
Lipscomb *et al.*, 1998	Cohort with nested case control. USA	Carpentry. Matched by age and sex from pool of identified carpenters of Western Washington	10628	-	-	S; A	2+ B1	S: Exposed control populations reported high rates of cough (30%), bronchitis (32%), wheeze (40%), chest tightness (34%), and nasal congestion (38%). A: No significant difference in asthma outcomes between cases and controls.
Talini *et al.*, 1998	Case control. Italy	Furniture manufacture. Furniture assembly workers.	195/63	Pine, beech.	-	S; LF.	2+ B1	S: Sputum production more common in woodworkers vs spray painters. LF: Normal FEV_1 and FVC observed between workers and controls.
Eriksson *et al.*, 1997	Cross sectional. Sweden.	Joinery. NA	38	Pine	gmWD = 0.3 – 0.9; gmTERP = 19 – 123 mg/m^3	S; LF.	2- B2	No significant difference in symptoms over work shift. Significantly lower than predicted lung function in workers even when smokers removed from analysis. No change in cross shift lung function.

Åhman *et al.*, 1996.	Case control. Sweden	Industrial arts teachers. School employees.	39/32	Pine	-	S	2-B1	Nasal congestion reported in 35% of workers.
Åhman *et al.*, 1995	Case control. Sweden.	Industrial arts teachers. School employees	130/112	Pine	-	S	2-B2	Respiratory symptoms more common in shops where poorer control reported.
Herbert *et al.*, 1995.	Case control. Canada	Board manufacture. Oil field and gas workers from same geographical area.	127/165	OSB	RD = 0.05 – 0.5; FORM = < 0.05 ppm	S; LF	2-B2	S: Nocturnal shortness of breath, wheeze and shortness of breath more common in workers vs controls. LF: Significant cross-shift lung function loss observed in cases vs referents.
Herbert *et al.*, 1994.	Cross sectional. Canada	Board manufacture. Oil field and gas workers from same geographical area.	99	OSB	TD = 0.27; FORM = 0.27ppm	S; LF	2-B2	S: OR above 5 observed in nocturnal SOB, wheeze/chest tightness and asthma symptoms between exposed workers and controls. LF: Significant cross-shift FVC change observed in workers vs controls.
Norrish *et al.*, 1992	Case control. New Zealand.	Furniture manufacture and joinery. Office workers.	44/38	Rimu, MDF, tawa, kauri, Californian redwood.	Median ID = 3.6; median FORM = 0.06	S; A	2-B2	S: 77% of workers experienced sneezing, 32% of exposed experienced work-related cough, 25% experienced work-related wheeze. A: 12% of exposed workers diagnosed with asthma
Pisaniello *et al.*, 1992.	Case control. Australia.	Furniture manufacture. Hospital maintenance staff.	134/298	Softwood, MDF, particleboard	Wood machinists ID = 3.2; cabinet makers ID = 5.2	S	2+B1	Hardwood dust exposure associated with significantly increased OR for nasal symptoms.
Shamssain, 1992.	Case control. South Africa.	Furniture manufacture. Bottling firm workers.	145/152	MDF, pine	TD = 3.82	S; LF	2+B2	S: Cough, sputum, SOB, wheeze and winter nasal symptoms significantly more common amongst exposed workers vs controls. LF: $FEV_{1,}$ FVC, FEV_1/FVC, FEF and PEF significantly lower in furniture workers vs controls. Significantly higher percentage of exposed workers with longer tenure had an FEV_1/FVC of <70%.
Holmström *et al.*, 1991	Case control. Sweden.	Furniture manufacture. Office workers (civil servants).	45/36	MDF, particle board, hardwoods	MDF group: WD = 0.8 – 3.8; FORM = 0.17 – 0.48.	S; LF	2-C	S: Nasal and ocular symptoms significantly more common in MDF groups vs referents.

					Wood dust group: WD = 0.7 – 1; FORM -= 0.08 – 0.17			LF: FEV_1 and FVC significantly worse in MDF exposed and wood dust exposed relative to controls.
Pisaniello *et al.*, 1991.	Case control. Australia	Furniture manufacture. Hospital maintenance staff.	193/46	Softwood, MDF, particleboard	ID = 3.7	S	2-B2	No significant difference in symptoms observed between exposure groups.
Malaka *et al.*, 1990	Case control. Indonesia.	Plywood manufacture. Non-exposed workers from same company.	93/93	Plywood	RD = 0.6; TD = 5.78; FORM = 0.5 – 2.36ppm	S; LF	2-B2	S: Significantly raised OR for asthma, work absence due to chronic cough, phlegm, episodic cough/phlegm and chronic bronchitis between exposed workers and controls. LF: No effect observed in baseline lung function measurements or between exposure groups.
Alexandersson *et al.*, 1989.	Longitudinal. Sweden	Kitchen cabinet manufacture. Non-exposed referents.	47/20	Chipboard	TD = 0.1; RD = 0.1; FORM = 0.5	LF	2-B2	Excess longitudinal decline observed in smoking woodworkers. Significant improvement in FEV_1, FVC, and FEV_1/FVC observed in non-smokers following exposure removal for 4 weeks.
Beretić-Stahuljak *et al.*, 1988	Cross sectional. Croatia.	Wood processing. NA	398	Softwood	TD = 0.8 – 40	LF	2-B2	LF: Small but significant decrements in FEV_1. FVC and FEV_1FVC associated with exposure.
Goldsmith *et al.*, 1988	Case control. USA	Furniture manufacture. Yardmen, shippers, office workers.	78/16	Hardwoods	-	S; LF	2+B1	S: Work-related sneezing and ocular symptoms significantly more common in exposed workers vs controls. LF: Cross-shift FEV_1, PEFR, and FEF values significantly worse in exposed workers.
Holmström *et al.*, 1988	Case control. Sweden.	Furniture manufacture. Office workers (civil servants).	100/106	-	WD = 1-2; FORM = 0.2 – 0.3	S; LF	2-C	S: Workers in formaldehyde exposed group experienced more nasal, ocular and lower respiratory symptoms. Work related symptoms similar between formaldehyde and wood dust exposed groups. LF: FEV_1 and FVC significantly lower in wood/formaldehyde vs control group.
Carosso *et al.*, 1987	Case control. Italy.	Furniture manufacture.	90/53	Variety of mostly European hard (e.g. iroko, oak) and soft (e.g.	-	LF; SZ	2-B2	LF: FEV_1. FVC, FEV_1FVC, TLCO and KCO significantly lower in exposed symptomatic vs exposed asymptomatic workers.

		Unexposed laboratory population.		Douglas fir, poplar) wood				SZ: Low frequencies of skin prick positivity to wood species. Low specific IgG positivity in exposed vs non-exposed group.
Tan *et al.*, 1987.	Cross sectional. Hong Kong.	Furniture manufacture. NA	30	Rosewood	TD = 4.43	S; LF	2-C	S: 50% or more workers reported nasal/throat symptoms or sputum. LF: Loss of FEV_1/FVC in linear regression with cumulative exposure. No effect for cross-shift change.
Holness *et al.*, 1985	Case control. Canada.	Carpentry/Joinery. Hospital staff.	50/50	-	TD = 1.83; RD = 0.29; FORM = 0.08	S; LF	2-B1	S: More than 30% of workers reported either cough, sputum, or rhinitis vs controls. 5% reported asthma symptoms. LF: Negative for cross-shift change in FEV_1 and FVC; negative for longitudinal change in FEV_1 and FEF75.
Wilhelmsson *et al.*, 1984 (a)	Cross sectional. Sweden.	Furniture manufacture. Non- or slightly exposed controls.	484/192	Hardwood	-	S; LF	2-C	S: Nasal symptoms significantly more frequent in heavily vs slightly exposed group. LF: Significantly lower FEV_1 and FVC in exposed workers.
Wilhelmsson *et al.*, 1984 (b)	Case control	Furniture manufacture. Exposed asymptomatic workers.	23/14	Hardwood	TD = 0.32 – 4.03	SZ	2-B2	Significantly higher levels of skin prick positivity to specific woods in symptomatic vs asymptomatic workers. Low overall positivity to specific woods.
Al-Zuhair *et al.*, 1981	Cross sectional. UK.	Furniture and cabinet manufacture. NA	333	Softwood, hardwood, chipboard.	TD = 0.69 – 4.93; RD = 0.03 – 0.66	LF	2-B1	Negative effect observed for cross-shift LF change between high and low exposure groups.
Whitehead, 1981	Cross sectional. USA.	Furniture manufacture, sawmills, plywood manufacture. Within study controls.	1157	Hardwood, pine	-	LF	2+ B1	Lower FEV_1/FVC ratios observed within high vs low exposure group.

[a] Wood types where stated: MDF = medium density fibreboard; OSB = oriented strand board; WRC = western red cedar

[b] Where exposures were measured in the study, they are expressed as a single value or range depending on the data provided: ID = inhalable dust, RD = respirable dust, TD = total dust, WD = wood dust where fraction size has not been specified, FORM = formaldehyde, END = endotoxin, BGD = (1,3)-β-D-glucan, TF = total fungi, BACT = bacteria, GNBACT = gram negative bacteria, TERP = terpenes, gm = geometric mean, ppm = parts per million.

[c] Where identified as primary study outcome measures: S = symptoms, A = asthma, LF = lung function, SZ = sensitisation

Appendix B: Supporting documents used in the study 'Assessment of respiratory ill-health in British woodworkers'

1. *Health Questionnaire: Assessment of respiratory ill health in the woodworking population*

PERSONAL DETAILS							
Forename(s)		Surname					
DOB		N.I Number					
Company ID		Personal ID					
Home address			GP Details				
Contact Tel.							
Shift pattern	Daytime hours (e.g. 9-5)		Nighttime hours		Other		
Gender		Height in cm		Weight in kg		BMI	

OCCUPATIONAL HISTORY					
Current occupation		Current work area			
Time worked in this work area	Years	Months	Time worked in this industry exposed vapours, gases, dusts or fumes	Years	Months

Please list your job history, with your current job first (if more room is needed, use continuation sheet) *If you have performed different tasks at your current worksite, please consider each a separate job and answer below*	
Dates/duration (Provide a range e.g. 2000-2010 or time in months/years)	Type of work and material used (e.g. sawing, cutting, planing, sanding, staining/finishing, using MDF, hard or soft woods or mix)
CURRENT JOB	CURRENT JOB

No.	RESPIRATORY PROTECTIVE EQUIPMENT						Yes	No
1	Do you wear any respiratory protection equipment (RPE) during your shift? *If no, go to question 4*							
2	How many hours a day do you wear it?						Number of hours	
3	If yes, which type do you usually wear from the below list? *See picture guide. Next, go to question 5.*						Yes	No
	Disposable half mask respirators							
	Simple cloth or fabric face mask							
	Valved disposable half mask respirators							
	Disposable welding respirators							
	Half mask filter respirators							
	Full-face respirators							
	Power air purifying							
	Air fed helmet							
	Self-contained breathing apparatus							
4	If no, why do you not wear RPE?							
	Not available	Not required	Uncomfortable	Unaware of RPE	Choose not to	Other		
No.	MEDICAL HISTORY							

5	Have you ever had to change or leave your job because it affected your breathing? *If no, go to question 7*					Yes	No
6	If yes, what was the job, and when were you employed in that job? *Please state here*			Job		Dates (duration)	
7	Have you ever worked in a job, which exposed you to vapours, gas, dust or fumes (apart from those mentioned previously)? *If no, go to question 11*					Yes	No
8	If yes, what was the job? *Please state here.*						
9	What was the exposure? *For example; wood dust, spray paints, resin*						
10	How long were you exposed?			Years		Months	
11	Have you ever had any of these medical problems diagnosed by a doctor?					sYes	No
	Asthma						
	Rhinitis						
	Eczema or dermatitis						
	COPD (Chronic bronchitis, chronic pulmonary disease, or emphysema)						
	Conjunctivitis/other allergies						
	Nasopharyngeal cancer						
	Other respiratory problems; if yes, please state						
No.	ASTHMA & ATOPY						
12	Do you currently have a diagnosis of asthma made by a doctor? If no go straight to question 24					Yes	No
13	How long have you been diagnosed with asthma?			Years		Months	
14	Have you had an attack of asthma in the last 12 months? *If no, go to question 16*					Yes	No
15	How many asthma attacks have you had in the last 12 months?						
16	Have you been woken up by an attack of asthma in the last 12 months?					Yes	No
17	During the past four weeks, how much has your asthma interfered with your usual activities (e.g. at work, exercising, at home)? *Please chose from the options below*						
	1 – all of the time	2 – most of the time	3 – some of the time	4 – a little of the time	5 - never		
18	Are you currently taking any medicines, including inhalers, aerosols or tablets for asthma? *If no, go to question 23*					Yes	No
19	If yes, are you taking regular inhaled steroids *Please refer to picture guide*			Name (if known)		Dose (if known)	
20	Are you taking a rescue inhaler or nebuliser? *Please refer to picture guide. If no, go to question 23*			Name (if known)		Dose (if known)	
21	During the past four weeks, how often have you used your rescue inhaler or medicine?						
	3 or more times a day	Once or twice a day	Two or three times a week	Once a week or less	Not at all		
22	Compared with work days, how much do you use your inhaler on days off?						
	Same		More		Less		✓ *as applicable*
23	Is your asthma the same, better or worse on days off?						
	Same		Better		Worse		✓ *as applicable*
24	Have you ever suffered from eczema, hay fever, or other allergies?						
25	Do you have a family history of asthma or allergy?						
No.	SMOKING					Yes	No
26	Do you smoke? *If yes go to question 28*						
27	Have you ever smoked as much as 1 cigarette per day, or 1 cigar per week, or 1oz tobacco a month for as long as a year? If no go to question 30						
28	How many cigarettes (or equivalent e.g. roll ups) do you (did you) smoke per day? *Please provide a number (e.g. 10) or a range (e.g. 5-10)*						
29	For how many years?			Years		Months	
No.	COUGH					Yes	No

30	Have you been woken by an attack of coughing at any time in the last 12 months?											
31	Do you usually cough first thing in the morning in winter?											
32	Do you usually cough at other times of the day or night in winter? *If no, go straight to question 38*											
33	*Do you usually bring up any phlegm from the chest when you cough?*											
34	Is this cough the same, better or worse on days off?											
	Same		Better		Worse		✓ *as applicable*					
35	Is this cough the same, better or worse on holiday?											
	Same		Better		Worse		✓ *as applicable*					
36	Do you cough like this on most days for as much as three months out of a year?										Yes	No
37	How long have you had this cough?					Years			Months			
No.	CHEST TIGHTNESS										Yes	No
38	Have you been woken up with a feeling of tightness in your chest at any time in the last 12 months?											
39	Does your chest ever become tight or breathing become difficult? *If no, go straight to question 45*											
40	Do you get this chest tightness only with colds? *If yes, go straight to question 45*											
41	On the days when you have chest tightness, is it worse at any particular time of the day or night? *If yes, please tick box(es) below*											
	Morning		Afternoon		Evening		Night		At Work			
42	Is the chest tightness the same, better or worse on days off?											
	Same		Better		Worse		✓ *as applicable*					
43	Is this chest tightness the same, better or worse on holiday?											
	Same		Better		Worse		✓ *as applicable*					
44	How long have you had this chest tightness?					Years			Months			
No.	WHEEZE										Yes	No
45	Have you had wheezing or whistling in your chest at any time in the last 12 months?											
46	Have you been woken up with a wheezing of whistling in your chest in your chest at any time in the last 12 months?											
47	Does your chest ever sound wheezy or whistle? If no, go straight to question 56											
48	Do you get this only with colds? *If yes, go straight to question 56*											
49	Have you been at all breathless when the wheezing noise was present?											
50	Is (was) your breathing absolutely normal in between attacks of wheezing?											
51	On the days when you wheeze, is it worse at any particular time of the day or night? *If yes, please tick box(es) below*											
52	Morning		Afternoon		Evening		Night		At Work			
53	Is the wheezing the same, better or worse on days off?											
	Same		Better		Worse		✓ *as applicable*					
54	Is this wheezing the same, better or worse on holiday?											
	Same		Better		Worse		✓ *as applicable*					
55	How long have you had this wheeze?					Years			Months			
No.	SHORTNESS OF BREATH										Yes	No
56	Have you been woken by an attack of shortness of breath at any time in the last 12 months?											
57	Are you disabled from walking by a problem other than heart or lung disease*?* *If yes, go straight to question 66*											
58	At worst are you troubled by shortness of breath or breathing when either hurrying on level ground or walking up a slight hill*? If no, go straight to question 66*											
59	At worst do you get short of breath walking with other people of your own age on level ground*? If no, go straight to question 62*											
60	At worst do you have to stop for breath when walking at your own pace on level ground*? If no, go straight to question 62*											
61	At worst do you become short of breath while resting*?*											
62	Is the shortness of breath worse at any particular time of the day or night*? If yes, please tick box(es) below*											
	Morning		Afternoon		Evening		Night		At Work			
63	Is the shortness of breath the same, better or worse on days off?											
	Same		Better		Worse		✓ *as applicable*					
64	Is the shortness of breath the same, better or worse on holiday?											
	Same		Better		Worse		✓ *as applicable*					
65	How long have you had this shortness of breath?					Years			Months			

No.	EYE IRRITATION	Yes	No
66	Do you suffer from eye irritation such as pricking, itching, burning, dryness, watering, soreness, or stinging? *If no, go straight to question 72*		
67	Is the eye irritation worse at any particular time of the day or night? *If yes, please tick box(es) below*		

Morning		Afternoon		Evening		Night		At Work	

68	Is the eye irritation the same, better or worse on days off?						
	Same		Better		Worse		✓ *as applicable*
69	Is the eye irritation the same, better or worse on holiday?						
	Same		Better		Worse		✓ *as applicable*

70	Is the eye irritation worse during any season of the year?								
	Spring		Summer		Autumn		Winter		✓ *as applicable*

71	How long have you had this eye irritation?	Years	Months

No.	NASAL IRRITATION	Yes	No
72	Apart from when you have a cold do you suffer from nasal irritation such as pricking, itching, burning, sneezing, or a runny, dry or blocked nose? *If no, go straight to question 77*		
73	Is the nasal irritation worse at any particular time of the day or night? *If yes, please tick box(es) below*		

Morning		Afternoon		Evening		Night		At Work	

74	Is the nasal irritation the same, better or worse on days off?						
	Same		Better		Worse		✓ *as applicable*
75	Is the nasal irritation the same, better or worse on holiday?						
	Same		Better		Worse		✓ *as applicable*

76	How long have you had this nasal irritation?	Years	Months

No.	PARTICIPATION IN HYGIENE STUDY	Yes	No
77	Have you taken part in the recent HSE hygiene study? *If no, go straight to question 79*		
78	If yes, did you have personal monitoring?		

No.	HEALTH SURVEILLANCE	Yes	No
79	Are you currently under a health surveillance programme at your workplace?		
80	If yes, have you had any of the following; *please tick box(es) below*		

Questionnaire		Spirometry		Other (please state)

eNO TEST RESULTS			
Date completed		By whom	
Time work commenced		Time test recorded	
Time last:	Eaten	Smoked	Used steroid inhaler

Attempt number	Result
1st attempt	
2nd attempt	
3rd attempt	
eNO test comment	

QUALITY CONTROL FOR eNO	Yes	No
Has the ATS recommendation been fulfilled?		
Is the equipment log up to date with verification and calibration records?		

LUNG FUNCTION TEST RESULTS					
Date completed		By whom			
FEV_1 (litres)	FVC (litres)	PEF (litres/min)	Percentage predicted FEV_1	Percentage predicted FVC	Percentage predicted PEF
Lung function test comments					

QUALITY CONTROL FOR LUNG FUNCTION	Yes	No
Has the ATS criteria been fulfilled?		
Is the spirometer log up to date with verification and calibration records?		

2. Consent form for woodwork study

CONSENT FORM

Study Number: PH06113

Participant Identification Number for this trial:

Title of Project: Assessment of respiratory ill health in the woodworking population of Britain

Name of Researcher:

1. I confirm that I have read and understand the information sheet for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily. ☐

2. I understand that my participation is voluntary and that I am free to withdraw at any time without giving any reason, without my medical care or legal rights being affected. ☐

3. I agree to any prior workplace health surveillance being reviewed for the purposes of the study. ☐

4. I understand that my results will be looked at by the study team, at the Health & Safety Laboratory. ☐

5. I understand that anonymised results from my workplace may be included in a report to HSE and my employer. ☐

6. I agree to being contacted, along with my GP, in the event of any important abnormality being detected from my tests. ☐

7. I understand that in the event of any other important abnormality being found, this information will only be passed onto my GP with my consent ☐

8. I agree to take part in the above study. ☐

……	……	……
Name of Participant	Date	Signature
……..	……	……..
Name of Person taking consent	Date	Signature

When completed: 1 for participant & 1 for research file

3. Participant information sheet for woodwork study

Assessment of respiratory ill health in the woodworking population of Britain

INFORMATION FOR VOLUNTEERS

Your workplace has agreed to take part in a Health and Safety Executive (HSE) research project looking into safe levels of exposure to dusts, vapours and fumes in the woodworking industry (a hygiene study). Alongside this study there will be health study that you may be asked to be involved in. This leaflet provides more information on the health study.

What is the purpose of this study?

This study is looking at the health of workers who work in the woodworking industry. This is because we know that wood dust and some of the processes of treating wood can cause breathing problems. We are interested in your general health and the health of your lungs.

What will be involved if I agree to take part in this study?

If you agree to take part, we will visit your worksite during your normal working hours, with the agreement of your employers. We will ask some simple questions about your work history, any breathing problems you might have, and your health at work. We would also like to look at any old breathing tests you may have had done at work and to review any of your past health information your work may have.

We will ask you to carry out 2 simple breathing tests. These are quick and painless test, similar to blowing out candles on a birthday cake. The tests are done at GP surgeries and hospitals around the UK, and you may have had one in the past. One measures breathing function, and the other is a simple test for asthma.

We will also ask you to provide a small blood sample that will be taken by the study doctor. This is done in the same way as your normal GP does blood tests and may cause some minor discomfort. The test is to help us see who is at risk of developing problems from working with wood. If you do not want to give a blood sample, we would still like you to take part as the other information you can give is also very useful.

Do I have to take part?

No, the study is voluntary. You can decline to join the study, and you can withdraw from the study at any time without giving any reason.

What are the possible benefits of taking part?

If your breathing function is reduced, it may be helpful for you and your family doctor (or GP) to know. If your blood tests show any signs of allergy then it would be helpful to contact you and your GP about it. We will only contact your GP if you agree to it.

Your workplace will get feedback following participation in the study about the health of the workforce. Your personal information will not be shared with anyone outside of the study team.

What are the possible risks of taking part?

It is possible that you might develop a bruise from the blood tests.

What if there is a problem?

In the unlikely event that something does go wrong and you are harmed during the study due to someone's negligence, you may have grounds for compensation against HSE or the NHS but you may have to pay your own legal costs.

Will my taking part be confidential?

Yes. No one other than the study team will know about this unless you choose to inform them. All information will be treated with strict medical confidentiality. If we do think your information should be shared with your GP, this will only be done with your agreement.

What will happen to the results of the research study?

The results of the study will be published in a report for the HSE and in scientific journals. Your own results will not be identifiable from any of these publications.

Who is organizing and funding the research?

The study is being organized by the Centre for Workplace Health. The study is being funded by the Health and Safety Executive.

Who has looked at the study to make sure it is safe?

This study has been reviewed and approved by an NHS Ethics committee.

Contact for further information?

If you have any concerns or questions about this study, you can contact the staff at the Centre for Workplace Health who would be happy to help you:

cwh@hsl.gsi.gov.uk
ruth.wiggans@hsl.gsi.gov.uk
Dr Ruth Wiggans: 01298218646

4. Employer information for woodworking study

Assessment of respiratory ill health in the woodworking population of Britain

INFORMATION FOR EMPLOYERS

Your workplace has agreed to participate in a Health and Safety Executive (HSE) research project looking into safe levels of exposure to dusts, fumes and vapours in the woodworking industry (a hygiene study). Alongside this study there will be a health study that we would like your employees to participate in. This leaflet provides more information about the health study.

What is the purpose of this study?

This study is part of a larger HSE study looking at improving the health of workers exposed to a range of substances in the workplace. Over the past two years, this study has successfully collected data from over 300 foundry workers, and 300 workers exposed to brick and stone dust. We know that wood workers can also develop breathing problems, and would therefore like to include them in the study. We are interested in measuring their health, and linking this to current and historical levels of exposure.

What will be involved if my company agrees to take part in this study?

We will visit your worksite during normal working hours at an agreed time. We will spend a maximum of 30 minutes with individual employees who have agreed to take part in the study. We will ask questions about their general health, work history, and current breathing problems, as well as other things that might affect breathing like smoking. We will perform simple breathing tests and take a small blood sample. The blood sample will be used to look for signs of allergies, and will not be used for any other purpose.

Does my company have to take part?

No, the study is voluntary. You can decline to join the study, and you can withdraw from the study at any time without giving any reason. Your taking part is very important however, as measuring health and hygiene data in your company offers a unique opportunity to better understand the effects of wood dust exposure.

What are the possible benefits of taking part?

The results from your workplace will be fed back to you alongside results from other sites for comparison. These results do not contain any personal information that would allow individual people to be identified and your own company results will not be identifiable to anyone else. The information provided may assist your company in making improvements in the workplace that help protect the health of your workforce in the future.

If any workers taking part are found to have breathing problems, or a reduction in breathing function, they will be advised to consult their GP. We will not do this without that individual's permission. Finding such problems early is generally good for the future health of these workers.

What will happen to the results of the research study?

The results of the study will be published in a report for the HSE and in scientific journals. Each company will also be provided with a copy of the report.

Who is organizing and funding the research?

The study is being organized by the Centre for Workplace Health. The study is being funded by the Health and Safety Executive.

Who has looked at the study to make sure it is safe?

This study has been reviewed and approved by an NHS Ethics committee.

Contact for further information?

If you have any concerns or questions about this study, you can contact the staff at the Centre for Workplace Health who would be happy to help you:

Email: ruth.wiggans@hsl.gsi.gov.uk or cwh@hsl.gsi.gov.uk

Dr Ruth Wiggans: 0129821818646

Appendix C: Supporting materials for study 'health impact assessment and surveillance for long-latency diseases: foundry exposed workers'

1. *Foundry Respiratory Questionnaire*

PERSONAL DETAILS					
Forename(s)		Surname			
DOB		N.I Number			
Company ID number		Personal ID number			
Home address			Gender	Male / Female	
			Hours worked		
Height in cm		Weight in kg		BMI	

OCCUPATIONAL HISTORY					
Current occupation			Current work area		
Time worked in this work area	Years	Months	Time worked in this industry exposed vapours, gases, dusts or fumes	Years	Months
Please list any personal protective equipment used:					

Please list job history (if more room is needed, use continuation sheet in appendix 3)				
Dates (e.g. 2000-2001)	Type of work (e.g. furnace work ferrous metals, furnace work non-ferrous metals, moulding, core making, pattern making, casting, knockout, fettling, finishing, supervisor, other)	Did you wear RPE?		If yes what % of day did you wear RPE?
		Yes	No	

No.	RESPIRATORY PROTECTIVE EQUIPMENT		Yes	No
1	Do you wear any respiratory protection equipment (RPE) during your shift? *If no, go to question 2, if yes go to question 3*			
2	If no, why do you not wear RPE?			
3	If yes, how many hours a day do you wear it?		Number of hours	
4	If yes, which type do you usually wear from the below list?		Yes	No
	Disposable half mask respirators			
	Simple cloth or fabric face mask			
	Valved disposable half mask respirators			
	Disposable welding respirators			
	Half mask filter respirators			
	Full-face respirators			
	Power air purifying			
	Air fed helmet			
	Self-contained breathing apparatus			
	MEDICAL HISTORY			
No.		HEALTHY	UNHEALTHY	
5	How would you assess your recent health condition? *Please tick as appropriate*			
6	Have you ever had to change or leave your job because it affected your breathing?		Yes	No
7	If yes, what was the job? *Please state here*			
8	Have you ever worked in a job, which exposed you to vapours, gas, dust or fumes (apart from those mentioned previously)?			

9	If yes, what was the job? *Please state here*			
10	Have you ever had any of these medical problems diagnosed by a doctor?	Date	Yes	No
	COPD (Chronic bronchitis, chronic pulmonary disease, or emphysema)			
	Silicosis			
	Pleurisy			
	Tuberculosis			
	Lung cancer			
	Other respiratory problems; if yes, please state			
	Kidney problems			
	Arthritis or connective tissue problems; if yes, please state			

No.	ASTHMA & ATOPY	Yes	No
11	Have you ever had asthma? If no go straight to question 16		
12	Have you had an attack of asthma in the last 12 months?		
13	Are you currently taking any medicines, including inhalers, aerosols or tablets for asthma?		
14	If yes, are you taking regular inhaled steroids?		
15	Other medicines are taken for asthma; if yes, please state		
16	Have you ever suffered from eczema, hay fever, or other allergies?		
17	Do you have a family history of asthma or allergy?		

No.	SMOKING	Yes	No
18	Do you smoke? If yes go straight to question 20		
19	Have you ever smoked as much as 1 cigarette per day, or 1 cigar per week, or 1oz tobacco a month for as long as a year? If no go straight to question 22		
20	How many cigarettes (or equivalent e.g. roll ups) do you (did you) smoke per day?		
21	For how many years?	Years	Months

No.	COUGH	Yes	No
22	Do you usually cough first thing in the morning in winter?		
23	Do you usually cough at other times of the day or night in winter? If no, go straight to question 30		
24	*Do you usually bring up any phlegm from the chest when you cough?*		
25	Is this cough the same, better or worse on days off?		
26	Is this cough the same, better or worse on holiday?		

Same		Better		Worse		✓ *as applicable*	

27	Do you cough like this on most days for as much as three months out of a year?		
28	Have you been woken by an attack of coughing at any time in the last 12 months?		
29	How long have you had this cough?	Years	Months

No.	CHEST TIGHTNESS	Yes	No
30	Does your chest ever become tight or breathing become difficult? *If no, go straight to question 37*		
31	Do you get this chest tightness only with colds? If yes go straight to question 37		
32	On the days when you have chest tightness, is it worse at any particular time of the day or night? *If yes, please tick box(es) below*		

Morning		Afternoon		Evening		Night		At Work		

33	Have you been woken up with a feeling of tightness in your chest at anytime in the last 12 months?		
34	Is the chest tightness the same, better or worse on days off?		
35	Is this chest tightness the same, better or worse on holiday?		

Same		Better		Worse		✓ *as applicable*

36	How long have you had this chest tightness?	Years	Months

No.	WHEEZE	Yes	No
37	Have you had wheezing or whistling in your chest at any time in the last 12 months?		
38	Does your chest ever sound wheezy or whistle? If no, go straight to question 46		
39	Do you get this only with colds? *If yes, go straight to question 46*		
40	Have you been at all breathless when the wheezing noise was present?		
41	Is (was) your breathing absolutely normal in-between attacks of wheezing?		

42	On the days when you wheeze, is it worse at any particular time of the day or night? *If yes, please tick box(es) below*												
	Morning		Afternoon		Evening		Night		At Work				
43	Is the wheezing the same, better or worse on days off?												
	Same		Better		Worse								✓ *as applicable*
44	Is this wheezing the same, better or worse on holiday?												
	Same		Better		Worse								✓ *as applicable*
45	How long have you had this wheeze?						Years		Months				
No.	SHORTNESS OF BREATH											Yes	No
46	Are you disabled from walking by a problem other than heart or lung disease*?* *If yes, go straight to question 56*												
47	At worst are you troubled by shortness of breath or breathing when either hurrying on level ground or walking up as slight hill*?* *If no, go straight to question 56*												
48	At worst do you get short of breath walking with other people of your own age on level ground*?* *If no, go straight to question 56*												
49	At worst do you have to stop for breath when walking at your own pace on level ground*?* *If no, go straight to question 56*												
50	At worst do you become short of breath while resting*? If no, go straight to question 56*												
51	Is the shortness of breath worse at any particular time of the day or night*? If yes, please tick box(es) below*												
	Morning		Afternoon		Evening		Night		At Work				
52	Have you been woken by an attack of shortness of breath at any time in the last 12 months?												
53	Is the shortness of breath the same, better or worse on days off?												
	Same		Better		Worse								✓ *as applicable*
54	Is the shortness of breath the same, better or worse on holiday?												
	Same		Better		Worse								✓ *as applicable*
55	How long have you had this shortness of breath?						Years		Months				
No.	EYE IRRITATION											Yes	No
56	Do you suffer from eye irritation such as pricking, itching, burning, dryness, watering, soreness, or stinging? *If no, go straight to question 62*												
57	Is the eye irritation worse at any particular time of the day or night? *If yes, please tick box(es) below*												
	Morning		Afternoon		Evening		Night		At Work				
58	Is the eye irritation the same, better or worse on days off?												
	Same		Better		Worse								✓ *as applicable*
59	Is the eye irritation the same, better or worse on holiday?												
	Same		Better		Worse								✓ *as applicable*
60	Is the eye irritation worse during any season of the year?												
	Spring		Summer		Autumn		Winter						✓ *as applicable*
61	How long have you had this eye irritation?						Years		Months				
No.	NASAL IRRITATION											Yes	No
62	Apart from when you have a cold do you suffer from nasal irritation such as pricking, itching, burning, sneezing, or a runny, dry or blocked nose? *If no, go straight to question 67*												
63	Is the nasal irritation worse at any particular time of the day or night? *If yes, please tick box(es) below*												
	Morning		Afternoon		Evening		Night		At Work				
64	Is the nasal irritation the same, better or worse on days off?												
	Same		Better		Worse								✓ *as applicable*
65	Is the nasal irritation the same, better or worse on holiday?												
	Same		Better		Worse								✓ *as applicable*
66	How long have you had this eye irritation?						Years		Months				

No.	WELDING AND METAL WORKING FUME	Yes	No

67	Do you currently work with metals e.g. molten metals, welding, burning? *If no, go straight to question 72*		
68	During the last 12 months did you experience any of the following symptoms within the first few hours of starting work?		
	Metallic taste		
	Fever		
	Feelings of flu		
	General malaise		
	Chills		
	Loss of appetite		
	Nausea		
	Abdominal cramps		
	Fatigue		
	Yawning		
	Difficulty concentrating		
69	If yes to any one; how long does it take after you start being exposed for this to happen in hours?	Number of hours	
70	What base metal were you working with when you got this problem? *Please state below*		
71	How many times have you had the problem?		

No.	PARTICIPATION IN HYGIENE STUDY	Yes	No
72	Have you taken part in the recent HSE hygiene study? *If no, go straight to question 75*		
73	If yes, did you have personal monitoring?		
74	If yes, did you have a urine test?		
No.	HEALTH SURVEILLANCE	Yes	No
75	Are you currently under a health surveillance programme at your workplace?		
76	If yes, have you had any of the following; *please tick box(es) below*		

Questionnaire		Spirometry		Chest x-ray		Other (please state)

eNO TEST RESULTS			
Date completed		By whom	
Attempt number		Result	
1st attempt			
2nd attempt			
3rd attempt			
eNO test comments			
As per the American Thoracic Society (ATS) recommendations for exhaled nitric oxide, three satisfactory manoeuvres should be recorded. It should take place prior to measurement of lung function.			

QUALITY CONTROL FOR eNO	Yes	No
Has the ATS recommendation been fulfilled?		
Is the equipment log up to date with verification and calibration records?		

LUNG FUNCTION TEST RESULTS					
Date completed		By whom			
FEV_1 (litres)	FVC (litres)	PEF (litres/min)	Percentage predicted FEV_1	Percentage predicted FVC	Percentage predicted PEF
Lung function test comments					

As per the American Thoracic Society (ATS) quality criteria for spirometry, three satisfactory manoeuvres should be recorded with the best two FEV_1 and FVC being within 150mls. The largest FEV_1 and the FVC should be recorded above, even if they come from separate blows.		
QUALITY CONTROL FOR LUNG FUNCTION	Yes	No
Has the ATS criteria been fulfilled?		
Is the spirometer log up to date with verification and calibration records?		

2. Foundry participant information sheet

Health Impact Assessment and Surveillance for Long-latency Diseases: Foundry Exposed Workers.

INFORMATION FOR VOLUNTEERS

You are invited to take part in a research study. Before you decide if you want to take part, please read this information sheet, which will tell you why this study is happening. Please talk to others about this if you need to, and take time to decide if you want to take part. Taking part is voluntary, however, the more people who do take part the more useful the findings will be.

What is the purpose of the study?

This study is looking at the health of workers who work in foundries and are involved in any work where fumes, dusts and gases may be breathed in. This is because exposures in foundries can cause breathing problems, and we are therefore interested in your lung health. This study will also look at breathing test results to decide how best to monitor workers who are currently breathing in dusts, fumes and gases whilst working in a foundry.

Your worksite has been selected as it is already participating in a separate study looking at the levels of fumes, gases and dust in foundries and this presents a unique opportunity to use a variety of methods to measure the health of workers and how this is affected by substances in the workplace.

What will be involved if I agree to take part in this study?

If you agree to take part, we will carry out a simple questionnaire asking about your work history, any breathing problems and other things that might affect your breathing such as smoking. We would also ask you to let us see any old measures of your lung function that your work may have and previous health questionnaires.

We will ask you to carry out a simple breathing test. This is a common test that is carried out a lot in hospitals and by GPs. It is a simple and painless test to measure the size of your lungs and your breathing capacity using a small portable machine. We will use another simple blowing test to measure any inflammation in your airways.

We will also ask you to provide a small sample of urine. This is to measure a substance that might help us to better understand the health effects of foundry exposures. This sample will only be used for this study and will not be used for any other purpose such as testing for drug or alcohol levels.

This study will run for three years, and we would like to come back and see you a second time to repeat the questionnaire, breathing tests and to collect another urine sample. This will be towards then end of the three-year study period.

Do I have to take part?

No, the study is totally voluntary. You can refuse to join the study, and you may withdraw from the study at any time without giving any reason.

What are the possible benefits of taking part?

The breathing tests will measure the size of your lungs and if there is any inflammation. If these tests do show any reduction in your breathing or show inflammation, it would be helpful for you and your family doctor (or GP), to know. We will only contact your GP with your agreement.

The overall findings of this research will also help many other workers in foundries, as it will help us understand the best way to protect their health at work.

What if there is a problem?

If you have a concern about any aspect of this study, you should ask to speak to Dr Chris Barber who will answer your questions [01298 218169].

In the unlikely event that something does go wrong and you are harmed during the research and this is due to someone's negligence, then you may have grounds for a legal action for compensation against HSE or the NHS but you may have to pay your legal costs.

Will my taking part be confidential?

Yes. No one else, other than the study team will know about this. All information will be treated with strict medical confidentiality. If we think information should be sent to your GP, we will only do this if you agree.

What will happen to the results of the research study?

The results of the study will be published in a report for the Health & Safety Executive (HSE) and in scientific journals. Your own results will not be able to be identifiable from any of these publications.

Who is organising and funding the research?

The Health and Safety Executive (HSE) has funded this research.

Who has reviewed the study?

This study has been reviewed and approved by an NHS Research Ethics Committee.

Contact for further information?

If you require further information about any aspect of this study, general information about research, advice on participation or what you should do if you are in any way unhappy with the study please contact Dr Chris Barber, 01298 218169, david.fishwick@hsl.gov.uk, HSL, Harpur Hill, Buxton, SK17 9JN.

3. Foundry consent form

TITLE OF PROJECT: LONG-LATENCY DISEASES IN BRITISH FOUNDRY WORKERS

CENTRE NUMBER:

STUDY NUMBER:

PATIENT IDENTIFICATION NUMBER FOR THIS TRIAL:

NAME OF RESEARCHER: _____

1. I CONFIRM THAT I HAVE READ AND UNDERSTAND THE INFORMATION SHEET DATED 01/02/2012(VERSION 2) FOR THE ABOVE STUDY. I HAVE HAD THE OPPORTUNITY TO CONSIDER THE INFORMATION, ASK QUESTIONS AND HAVE HAD THESE ANSWERED SATISFACTORILY. ☐

2. I UNDERSTAND THAT MY PARTICIPATION IS VOLUNTARY AND THAT I AM FREE TO WITHDRAW AT ANY TIME WITHOUT GIVING ANY REASON, WITHOUT MY MEDICAL CARE OR LEGAL RIGHTS BEING AFFECTED. ☐

3. I AGREE TO ANY PRIOR WORKPLACE BREATHING TESTS BEING REVIEWED FOR THE PURPOSES OF THE STUDY. ☐

4. I UNDERSTAND THAT MY RESULTS WILL BE LOOKED AT BY THE STUDY TEAM, AT THE HEALTH & SAFETY LABORATORY. ☐

5. I UNDERSTAND THAT ANONYMISED RESULTS FROM MY WORKPLACE MAY BE INCLUDED IN A REPORT TO HSE AND MY EMPLOYER. ☐

6. I AGREE TO BEING CONTACTED, ALONG WITH MY GP, IN THE EVENT OF ANY IMPORTANT ABNORMALITY BEING DETECTED FROM MY URINE TESTS. ☐

7. I UNDERSTAND THAT IN THE EVENT OF ANY OTHER IMPORTANT ABNORMALITY BEING FOUND, THIS INFORMATION WILL ONLY BE PASSED ONTO MY GP WITH MY CONSENT ☐

8. I AGREE TO TAKE PART IN THE ABOVE STUDY. ☐

NAME OF PARTICIPANT DATE SIGNATURE

NAME OF PERSON TAKING CONSENT DATE SIGNATURE

One copy to be kept by the researcher; one copy to be given to the patient.

Appendix D: Supporting materials for SPIRAL study of laboratory animal workers

(reproduced with kind permission of Dr Johanna Feary)

1. SPIRAL Questionnaire

Q1 Please enter your unique identifier code
Q2 Please enter your gender: Male (1) Female (2)
Q3 Have you been given a Participant Information Sheet for this study?
Yes (1) No (0)
Q4 Please contact a member of the SPIRAL team if you have not been given a Participant Information Sheet
Q5 Have you signed the consent form?
Yes (1) No (0)
Q6 Please contact a member of the SPIRAL team if you have not signed a consent form
Q7 Have you been given the contact telephone number and email address to get your results if you would like them?
Yes (1) No (0)
Q8 Please contact a member of the SPIRAL team to get the contact details so you can get your results if you would like them

Q9 THE FOLLOWING QUESTIONS ARE ABOUT YOUR CURRENT POST (JOB)

Q10 What year did you start in your current post? (yyyy)
Q11 Which of the following best describes your current post?
Animal technician (1) Scientist/research student/research assistant (2)
Department manager/administrator/secretary (3) Maintenance/cleaning/stores/security/estates (4)
Other (please enter details) (5) _____
Q12 Which of the following species are housed within the animal facilities where you work? (tick as many as applicable)
Mice (1) Rats (2)
Q13 In your current post, is your work:
More or less the same every week (1) Variable from week to week (2)

Q14 Do you work full-time or part-time in your current post?
Full time (1) Part time (2)
Q15 If you work part-time, how many hours a week do you work?
Q16 In the last 5 years, what is the longest single period of time (in months) you have spent without any contact with mice?
_____ months (2)
Q17 In the last 12 months, how many complete months have you spent without going into animal facilities?
0 (0) 1 (1) 2 (2) 3 (3) 4 (4) 5 (5) 6 (6)
7 (7) 8 (8) 9 (9) 10 (10) 11 (11) 12 (12)
Q18 In the last 12 months, how many complete months have you spent without contact with mice? (handling mice, going into a room where others handle mice, or handling their cages)
0 (0) 1 (1) 2 (2) 3 (3) 4 (4) 5 (5) 6 (6)
7 (7) 8 (8) 9 (9) 10 (10) 11 (11) 12 (12)

Q19 The NEXT SET OF QUESTIONS ARE ABOUT different TASKS YOU HAVE DONE AT WORK IN THE LAST 12 MONTHS. Some of them may be quite difficult to answer but please just try and give the best answer you can. it is important that we get a good idea about your level of exposure to mice at work.

Q20 TASK 1: GOING INTO THE ANIMAL FACILITIES

Q21 In the last 12 months: during those periods when you went into the animal facilities, which of the following best describes how often you did this? (i.e. went into the animal facilities)
Every day (1) At least once a week but not every day (2) At least once a month but every week (3)
Less than once a month (4) I did not go into the animal facilities(s) (5)
If I did not go into the anima... Is Selected, Then Skip to End of Block
Q22 In the last 12 months: on the days you went into the animal facilities, how much time (approximately) did you spend there on a typical day?
_____ Hrs (1)
Q23 In the last 12 months, what personal protective equipment did you use when you went into the animal facilities?

None (1)					

Gloves (2) Overshoes or special shoes (3) Lab coat/overall (4) Scrubs (5) Face mask (6) Powered helmet (7) Safety glasses or visor (8) Hair net or mob cap (9)					

Q24 In the past 12 months, are the mice in the animal facilities which you go into housed in IVCs or open cages?
IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)
I don't know (88)

Q25 TASK 2: DOSING/INJECTING MICE

Q26 In the last 12 months, during those periods when you had contact with mice, which of the following best describes how often you dosed/injected mice?
Every day (1) At least once a week but not every day (2)
At least once a month but not every week (3) Less than once a month (4)
I did not dose mice (5)
If I do not dose mice Is Selected, Then Skip to End of Block

Q27 In the last 12 months, on the days you dosed/injected mice, how much time on a typical day did you spend doing this?
_____ Hrs (1) _____ Mins (2)

Q28 In the last 12 months, on the days when you dosed/injected mice, how many mice did you dose/inject on a typical day?
1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q29 In the last 12 months, which type of personal protective equipment did you use when you dosed/injected mice?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1) Gloves (2) Overshoes or special shoes (3) Lab coat/overall (4) Scrubs (5) Face mask (6) Powered helmet (7) Safety glasses or visor (8) Hair net or mob cap (9)					

Q30 In the last 12 months, when you dosed/injected mice, do you do this on an open bench or under a hood?
Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q31 In the last 12 months, are the mice who you dosed/injected housed in IVCs or open cages?
IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q32 TASK 3: SHAVING MICE

Q33 In the last 12 months, during those periods when you had contact with mice, which of the following best describes how often you shaved mice?
Every day (1) At least once a week but not every day (2)
At least once a month but not every week (3) Less than once a month (4) I did not shave mice (5)
If I do not shave mice Is Selected, Then Skip to End of Block

Q34 In the last 12 months, on the days you shaved mice, how long did you spend doing this on a typical day?
_____ Hrs (1) _____ Mins (2)

Q35 In the past 12 months, on the days when you shaved mice, how many mice did you shave on a typical day?
1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q36 In the past 12 months, what personal protective equipment did you use when you shaved mice?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q37 In the last 12 months, when you shaved mice, did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q38 In last 12 months, were the mice who you shaved housed in IVCs or open cages?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q39 TASK 4: ANAESTHETISING MICE

Q40 In the last 12 months, during those periods when you had contact with mice, which of the following best describes how often you anaesthetised mice

Every day (1) At least once a week but not every day (2)

At least once a month but not every week (3) Less than once a month (4)

I did not anaesthetise mice (5)

If I do not anaesthetise mice Is Selected, Then Skip to End of Block

Q41 In the past 12 months, on the days when you anaesthetised mice, how much time did you spend doing this on a typical day?

_____ Hrs (1) _____ Mins (2)

Q42 In the past 12 months, on the days when you anaesthetised mice, how many mice did you anaesthetise on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q43 In the past 12 months, what personal protective equipment did you use when you anaesthetised mice?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q44 In the past 12 months, when you anaesthetised mice, did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q45 In the past 12 months, are the mice who you anaesthetised housed in IVCs or open cages?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q46 TASK 5: PERFORMING SURGERY ON MICE

Q47 In the past 12 months, during those periods when you had contact with mice, which of the following best describes how often you performed surgery (including dissection) on mice?

Every day (1) At least once a week but not every day (2)

At least once a month but not every week (3) Less than once a month (4)

I did not perform surgery (or dissection) on mice (5)

If I do not anaesthetise mice Is Selected, Then Skip to End of Block

Q48 In the past 12 months, on the days you performed surgery (including dissection) on mice, how much time did you spend on a typical day doing this?

_____ Hrs (1) _____ Mins (2)

Q49 In the past 12 months, on the days when you performed surgery (including dissection) on mice, how many mice did you perform surgery on, on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q50 In the past 12 months, what personal protective equipment did you use when you performed surgery (including dissection) on mice?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q51 In the past 12 months, when you performed surgery (including dissection) on mice, did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q52 In the past 12 months, are the mice on whom you performed surgery (including dissection) housed in IVCs or open cages?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q53 TASK 6: BEHAVIOURAL TESTING

Q54 In the past 12 months, during those periods when you had contact with mice, which of the following best describes how often you carried out behavioural testing on mice?

Every day (1) At least once a week but not every day (2)

At least once a month but not every week (3) Less than once a month (4)

I did not do behavioural work on mice (5)

If I do not cull mice Is Selected, Then Skip to End of Block

Q55 In the past 12 months, on the days you carried out behavioural testing on mice, how much time did you spend doing this on a typical day?

_____ Hrs (1) _____ Mins (2)

Q56 In the past 12 months, on the days when you did behavioural testing on mice, how many mice did you do this to on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q57 In the past 12 months, what personal protective equipment did you use when you did behavioural testing on mice?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q58 In the past 12 months, when you did behavioural testing on mice, did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q59 In the past 12 months, are the mice on whom you did behavioural testing housed in IVCs or open cages or a mixture?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q60 TASK 7: CULLING MICE.

Q61 In the past 12 months, during those periods when you had contact with mice, which of the following best describes how often you culled mice?

Every day (1) At least once a week but not every day (2)

At least once a month but not every week (3) Less than once a month (4)

I did not cull mice (5)

If I do not cull mice Is Selected, Then Skip to End of Block

Q62 In the past 12 months, on the days you culled mice, how much time did you spend doing this on a typical day?

_____ Hrs (1) _____ Mins (2)

Q63 In the past 12 months, on the days when you did cull mice, how many mice did you cull on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q64 In the past 12 months, what personal protective equipment did you use when you culled mice?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q65 In the past 12 months, when you culled mice, did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q66 In the past 12 months, are the mice whom you culled housed in IVCs or open cages or a mixture?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q67 TASK 8: CHANGING MICE CAGES

Q68 In the past 12 months, during those periods when you had contact with mice, which of the following best describes how often you changed mouse cages (transferred mice from dirty to clean cage)?

Every day (1) At least once a week but not every day (2)

Approximately two to three times a month (6) At least once a month but not every week (3)

Less than once a month (4) I didn't change mouse cages (5)

If I don't change mouse cages Is Selected, Then Skip to End of Block

Q69 In the past 12 months, on the days you spent transferring mice from dirty to clean cages, how much time did you spend doing this on a typical day?

_____ Hrs (1) _____ Mins (2)

Q70 In the past 12 months, on the days you spent changing mouse cages (transferring mice from dirty to clean cage), how many cages did you change on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q71 In the past 12 months, what personal protective equipment did you use when you changed mouse cages (transferred mice from dirty to clean cage)?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					

Hair net or mob cap (9)					

Q72 In the past 12 months, when you changed mice (transferred from dirty to clean cage), did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q73 In the past 12 months, are the mice whose cages you changed (transferred mice from dirty to clean cage) housed in IVCs or open cages or a mixture?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q74 WE REALISE THESE ARE BORING TO ANSWER BUT THE LIST OF TASKS IS NEARLY DONE! TASK 9: REMOVING DIRTY BEDDING FROM CAGES

Q75 In the past 12 months, during those periods when you had contact with mice, which of the following best describes how often you removed dirty bedding (in preparation for cage wash) from mouse cages?

Every day (1) At least once a week but not every day (2)

Approximately two to three times a month (6) At least once a month but not every week (3)

Less than once a month (4) I didn't remove dirty bedding from mouse cages (5)

If I don't remove dirty bedding... Is Selected, Then Skip to End of Block

Q76 In the past 12 months, on the days you spent removing dirty bedding from mouse cages, how much time on a typical day did you spend doing this?

_____ Hrs (1) _____ Mins (2)

Q77 In the past 12 months, on the days you spent removing dirty bedding from mouse cages, how many cages did you do this to on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q78 In the past 12 months, what personal protective equipment did you use when you remove dirty bedding from mouse cages?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q79 In the past 12 months, when you removed dirty bedding from mouse cages (before cage wash), did you do this on an open bench or under a hood?

Open bench only (1) Under a hood only (2) A mixture of open bench and under a hood (3)

Q80 In the past 12 months, did you remove dirty bedding from IVCs or open cages?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q156 TASK 10: CAGE WASH

Q157 In the past 12 months, during those periods when you had contact with mice, which of the following best describes how often you washed mouse cages?

Every day (1) At least once a week but not every day (2)

Approximately two to three times a month (6) At least once a month but not every week (3)

I didn't wash mouse cages (5) If I didn't wash mouse cages Is Selected, Then Skip to End of Block

Q158 In the past 12 months, on the days you spent washing mouse cages, how much time on a typical day did you spend doing this?

_____ Hrs (1) _____ Mins (2)

Q159 In the past 12 months, on the days you spent washing mouse cages, how many cages did you wash on a typical day?

1-10 (1) 11-20 (2) 21-50 (3) 51-100 (4) more than 100 (5)

Q160 In the past 12 months, what personal protective equipment did you use when you washed mouse cages?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					

Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q162 In the past 12 months, did you wash mouse cages which were IVCs or open cages?

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

Q81 TASK 11: ENTERING ROOMS WHERE MICE HOUSED WITHOUT HANDLING THEM

Q82 In the last 12 months: during those periods when you had contact with mice, which of the following best describes how often you went into rooms where mice are housed or handled (but did not handle them yourself) e.g. for observational work, to check them, to clean the rooms or replenish stocks

Every day (1) At least once a week but not every day (2)

Approximately two to three times a month (6) At least once a month but not every week (3)

I didn't go into rooms where others handle mice (5)

If I don't go into rooms where... Is Selected, Then Skip to End of Block

Q83 In the past 12 months, on the days when you went into rooms where mice are housed or handled (but did not handle them yourself), how much time on a typical day did you spend there? e.g. For observational work, to check them, to clean the rooms or replenish stocks

_____ Hrs (1) _____ Mins (2)

Q84 In the past 12 months, what personal protective equipment did you use when you go into rooms where mice are housed or handled (when you were not handling them yourself)? e.g. For observational work, to check them, to clean the rooms or replenish stocks

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q85 In the past 12 months, are the mice in the rooms which you go into (when you don't touch them yourself) housed in IVCs or open cages? e.g. For observational work, to check them, to clean the rooms or replenish stocks

IVC only (1) Open cages only (2) A mixture of IVCs and open cages (3)

I don't know (88)

Q86 In your current post do you handle pups or adult mice?

Pups only (1) Adult mice only (2) Both pups and adult mice (3) I don't handle mice (4)

Q87 In your current post do you handle female mice or male mice?

Female mice only (1) Male mice only (2) Both female and male mice (3) I don't handle mice (4)

Q88 THESE ARE THE LAST FEW QUESTIONS ABOUT YOUR WORK IN THE LAST 12 MONTHS

Q89 Over the last 12 months, what is the greatest amount of time in any one week you have spent in the animal facilities?

1-10 hours (1) 11-20 hours (2) 21-30 hours (3) 31-40 hours (4) 41-50 hours (5) more than 50 hours (6)

Q90 Over the last 12 months, what is the greatest amount of time in any one week you have handled (touched) mice? (including, for example, doing procedures on them)

1-10 hours (1) 11-20 hours (2) 21-30 hours (3) 31-40 hours (4) 41-50 hours (5) more than 50 hours (6) I did not handle(touch) mice (7)

Q91 Over the last 12 months, what is the maximum number of mice you have handled in any one week? If you handle the same mice on several occasions, count these separately e.g. 10 mice on 6 occasions each would be 60

1-10 (1) 11-50 (2) 51-100 (3) More than 100 (4) I did not handle mice (5)

Q92 Do you currently spend time in designated laboratories (rooms where mice are kept or handled outside the main animal facilities)?

Yes (1) No (2)

Q93 Which best describes the proportion of time in your current post you spend in the designated labs

I spend more time in designated labs than in the main animal facilities (1)

I spend more time in the main animal facilities than in the designated labs (2)

I spend equal amounts of time in designated labs and the main animal facilities (3)

Q94 Which type of personal protective equipment do you use (typically) when you work (go into) in the designated labs?

	Never (1)	Rarely (2)	Sometimes (3)	Most of the Time (4)	Always (5)
None (1)					
Gloves (2)					
Overshoes or special shoes (3)					
Lab coat/overall (4)					
Scrubs (5)					
Face mask (6)					
Powered helmet (7)					
Safety glasses or visor (8)					
Hair net or mob cap (9)					

Q95 THE NEXT SET OF QUESTIONS RELATE TO ANY PREVIOUS POSTS YOU HAVE HAD WHERE YOU HAVE WORKED WITH MICE.

This is my first post working with mice (1) I have had previous posts (2)

Q96 When did you first work with mice or in animal facilities containing mice? Do not count the occasional week or two in your training or studies (mm/yyyy)

Q97 Which type of personal protective equipment did you use in your first post when you worked with mice, mouse tissue or in rooms where mice are kept?

	None (1)	Gloves (2)	Over-shoes or special shoes (3)	Lab coat/ overall (4)	Scrubs (5)	Face mask (6)	Powered helmet (7)	Safety glasses or visor (8)	Hair net/mob cap (9)	N/A (10)
Going into animal facilities (1)										
Handling mice (2)										
Performing procedures on mice (e.g. shaving, culling) (3)										
Changing cages (move mice from dirty to clean cage) (4)										
Cleaning cages (tip sawdust out of dirty cage) (5)										
Going into a room where others handle mice (8)										

Q98 Which species have you worked with in previous posts? Tick as many as applicable

Mice (1) Rats (2) I have not worked with mice or rats in the past (4)

Q99 In your previous posts, what is the highest frequency you have ever....

	Every day (1)	At least once a week but not every day (2)	At least once a month but not every week (3)	Less than once a month (4)	Never (5)

Gone into an animal facilities containing mice? (2) Handled mice? (3) Removed dirty bedding from cages? (4)					

Q100 Which types of animal facilities have you worked in during previous posts?
IVC only facilities (1) Open cage only facilities (2) Both IVC and open cage facilities (3)
I have not worked in animal facilities in the past (4)
Q101 In your previous posts, what is the highest number of mice you have ever handled in one day?
1-10 (1) 11-50 (2) 51-100 (3) More than 100 (4) I did not handle mice in past posts (5)

Q102 THE NEXT SET OF QUESTIONS ARE ALL ABOUT YOUR HEALTH As with the rest of the questionnaire, the results are entirely CONFIDENTIAL and they will not be shared with your employer or occupational health department unless you choose to do so. Please answer them as accurately and honestly as you can.

Q103 Have you had asthma in the last 12 months?
Yes (1) No (2)
Q104 Did you have asthma as a child?
Yes (1) No (2)
Q105 Have you had hay fever in the last 12 months?
Yes (1) No (2)
Q106 Did you have hay fever as a child?
Yes (1) No (2)
Q107 Have you ever lived in a home where any of the following animals were kept as pets? tick as many as apply
Cat (1) Dog (2) Mouse (3) Rat (4) Gerbil (5) Hamster (6) Guinea pig (7) Rabbit (8)
None of the above (9)
Q108 Did you or do you ever have itchy eyes, sneezing, wheezing or chest tightness when you were near these pet animals?
Yes (1) No (2)
Q109 Which pet animals caused these symptoms? tick as many as apply
Cat (1) Dog (2) Mouse (3) Rat (4) Gerbil (5) Hamster (6) Guinea pig (7) Rabbit (8)
None of the above (9) I don't know (10)
Q110 Since starting work with laboratory animals have you had a blocked, itchy or runny nose or sneezing? (do not count colds or flu)
Yes (1) No (2)
Q111 What year did you first notice these symptoms? (yyyy)
Q112 What happens to these symptoms on weekends or holidays of one week or longer?
Get better (1) Get worse (2) No change (3)
Q113 Do you get these symptoms on contact with something at work?
Yes (1) No (2)
Q114 If you get a blocked, itchy or runny nose or sneezing on contact with something at work, then in your opinion, what causes these symptoms?
Q115 Since starting work with laboratory animals have you had itchy or runny eyes?
Yes (1) No (2)
Q116 What year did you first notice these symptoms? (yyyy)
Q117 What happens to these symptoms on weekends or holidays of one week or longer?
Get better (1) Get worse (2) No change (3)
Q118 Do you get these symptoms on contact with something at work?
Yes (1) No (2)
Q119 If you get itchy or runny eyes on contact with something at work, then, in your opinion, what causes these symptoms?
Q120 Since starting work with laboratory animals have you had tightness in the chest, or difficulty in breathing or wheezing or whistling in the chest?
Yes (1) No (2)
Q121 What year did you first notice these symptoms? (yyyy)
Q122 What happens to these symptoms on weekends or holidays of one week or longer?
Get better (1) Get worse (2) No change (3)
Q123 Do you get these symptoms on contact with something at work?
Yes (1) No (2)

Q124 If you get tightness in the chest, or difficulty in breathing or wheezing or whistling in the chest more on contact with something at work, then, in your opinion, what causes these symptoms?
Q125 Have you ever been scratched or bitten by a mouse?
Yes (1) No (2)
Q126 Have you ever had a swollen itchy rash (like a nettle sting) around the site of the scratch or bite?
Yes (1) No (2)
Q127 Has any health care professional ever told you that you are allergic to mice?
Yes (1) No (2)
Q128 What year did you first notice these symptoms? (yyyy)
Q129 Has any health care professional ever told you that you are allergic to any other laboratory animals?
Yes (1) No (2)
Q130 Which ones? tick all that apply
Rats (1) Guinea pigs (2) Rabbits (3) Other (4)
Q131 What year did you first notice these symptoms? (yyyy)
Q132 Have you had a mask fit test at work?
Yes (1) No (2)
Q133 THIS IS THE LAST SET OF QUESTIONS AND ARE A FEW GENERAL QUESTIONS ABOUT YOU

Q134 How many older siblings do you have? Don't count half-siblings or step-siblings and count a twin as a younger sibling
0 (1) 1 (2) 2 (3) 3 (4) 4 (5) 5 (6) more than 5 (7)
Q135 How many younger siblings do you have? don't count half-siblings or step-siblings and count a twin as a younger sibling
0 (1) 1 (2) 2 (3) 3 (4) 4 (5) 5 (6) 6 (7) 7 (8) more than 7 (9)
Q136 Do you live with any children?
Yes (1) No (2)
Q137 Do you smoke cigarettes?
Never (1) I used to smoke (2) I currently smoke (3)
Q138 What year did you start smoking? (yyyy)
Q139 What year did you stop smoking? (yyyy)
Q140 On average, how many cigarettes a day did you used to smoke?
Q141 On average, how many cigarettes a day do you smoke?
Q142 Which country were you born in?
Q143 Please choose one option that best describes your ethnic group or background?
White (12) Mixed / multiple ethnic groups (17) Asian / Asian British (22)
Black / African / Caribbean / Black British (28) Other ethnic group (32)
I do not wish to disclose this information (35)

Q144 You have now completed the questionnaire. Thank you for taking time to participate in this study, your help is very much appreciated. The SPIRAL team

2. SPIRAL consent form

Individually ventilated cages in laboratory animal facilities and the prevention of laboratory animal allergy: a proof-of-concept study.

(SPIRAL Study: (Safe Practice In Reducing Allergy in Laboratories)

Name:	**Date of birth:**

I have read and understood the Information document (version 3, 26/08/15) and the separate 'Genetics Information' (version 1.1 18/06/14). I have had the time to consider the information and ask questions and I am happy to take part. ☐

I understand that my participation is voluntary and I am free to withdraw at any time without giving a reason. ☐

I agree to undergo the following procedures:

Questionnaire ☐
Skin prick test ☐
Blood sample ☐
Spirometry ☐
FE_{NO} breathing test ☐

I understand that my samples are being given as a 'gift' and some may be stored and used for future research into laboratory allergy. Anonymous samples may be shared with collaborating laboratories. ☐

Name of volunteer (PRINT)	Signature	Date
Name of Investigator (PRINT)	Signature	Date

One copy to be kept by the researcher; one copy to be given to the patient.

SPIRAL study: **version 3, 26/8/15**
Dr J Feary, Royal Brompton and Harefield NHS Foundation Trust

List of abbreviations

ATS – American Thoracic Society

BTS – British Thoracic Society

CMF – Cast Metals Federation

CAS – Current asthma symptoms

CASCOT - Computer Assisted Structured Coding Tool

COIN - Corporate Operational Information System

COPD – Chronic obstructive pulmonary disease

COSHH – Control of Substances Hazardous to Health

CWH – Centre for Workplace Health

ECRHS – European Community Respiratory Health Survey

ERS – European Respiratory Society

FE_{NO} – Fractional exhaled nitric oxide

FEV_1 – Forced expiratory volume in 1 second

FVC – Forced vital capacity

HMW – High molecular weight

HSE – Health and Safety Executive

IgE – Immunoglobulin E

LAA – Laboratory animal allergy

LEV – Local exhaust ventilation

LMW – Low molecular weight

MDHS – Methods for the determination of hazardous substances

MRC – Medical Research Council

NSBHR – Non-specific bronchial hyperresponsiveness

OA – Occupational asthma

PEF – Peak expiratory flow

PPE – Personal protective equipment

RPE – Respiratory protective equipment

SIC -Specific Inhalation Challenge

SIgE – Specific immunoglobulin E

SIGN – Scottish Intercollegiate Guidelines Network

SOC - Standard Occupational Classification

SPIRAL – Safe Practice in Reducing Allergy in Laboratories

SRP – Strategic Research Programme

SWORD – Surveillance of Workplace Occupational Respiratory Diseases

TWA – Time-weighted average

WEL – Workplace exposure limit

WRC – Western red cedar

WRNS – Work-related nasal symptoms

WROS – Work-related ocular symptoms

WRRS – Work-related respiratory symptoms

WRS – Work-related symptoms

Bibliography

1. UK A. asthma.org.uk {online} London2019 [Available from: http://www. asthma.org.uk /asthma-facts-and-statistics.

2. Physicians. RCo. Why asthma still kills: the National Review of Asthma Deaths (NRAD) Confidential Enquiry report. London: Royal College of Physicians; 2014.

3. Asthma. GIf. Global Strategy for Asthma Management and Prevention. Wisconsin, USA: Global Initiative for asthma; 2018.

4. Society SIGNBT. SIGN 158: British guideline on the management of asthma. Edinburgh: Scottish Intercollegiate Guidelines Network; 2019.

5. Holgate S.T. WS, Postma D. S., Weiss S. T., Renz H., Sly P. D. Asthma. Nature Reviews Disease Primers. 2015;1:1-22.

6. Mukherjee M, Stoddart A, Gupta RP, Nwaru BI, Farr A, Heaven M, et al. The epidemiology, healthcare and societal burden and costs of asthma in the UK and its member nations: analyses of standalone and linked national databases. BMC medicine. 2016;14(1):113.

7. Barnes PJ, Jonsson B, Klim JB. The costs of asthma. European Respiratory Journal. 1996;9(4):636-42.

8. Sullivan PW, Slejko JF, Ghushchyan VH, Sucher B, Globe DR, Lin SL, et al. The relationship between asthma, asthma control and economic outcomes in the United States. Journal of Asthma. 2014;51(7):769-78.

9. Nicholson P.J., Cullinan P., Burge P.S., C. B. Occupational asthma: Prevention,identification & management: Systematic review & recommendations. London: British Occupational Health Research Foundation.; 2010.

10. Tarlo SM, Lemiere C. Occupational Asthma. The New England Journal of Medicine. 2014;370(7):640-9.

11. Fishwick D, Barber CM, Bradshaw LM, Harris-Roberts J, Francis M, Naylor S, et al. Standards of care for occupational asthma. Thorax. 2008;63(3):240-50.

12. Kogevinas M, Zock JP, Jarvis D, Kromhout H, Lillienberg L, Plana E, et al. Exposure to substances in the workplace and new-onset asthma: an international prospective population-based study (ECRHS-II). Lancet. 2007;370(9584):336-41.

13. Blanc PD, Annesi-Maesano I, Balmes JR, Cummings KJ, Fishwick D, Miedinger D, et al. The occupational burden of nonmalignant respiratory diseases. An official American Thoracic Society and European Respiratory Society statement. American journal of respiratory and critical care medicine. 2019;199(11):1312-34.

14. Anees W, Moore VC, Burge PS. FEV_1 decline in occupational asthma. Thorax. 2006;61(9):751-5.

15. Walters GI, Soundy A, Robertson AS, Burge PS, Ayres JG. Understanding health beliefs and behaviour in workers with suspected occupational asthma. Respiratory Medicine. 2015;109(3):379-88.

16. Bradshaw L, Barber C, Davies J, Curran A, Fishwick D. Work-related asthma symptoms and attitudes to the workplace. Occupational Medicine. 2007;57(1):30-5.

17. Ayres JG, Boyd R, Cowie H, Hurley JF. Costs of occupational asthma in the UK. Thorax. 2011;66(2):128.

18. Yranheikki E, Savolainen H. Occupational safety and health in Finland. Journal of Safety Research. 2000;31(4):177-83.

19. Anees W, Huggins V, Pavord ID, Robertson AS, Burge PS. Occupational asthma due to low molecular weight agents: eosinophilic and non- eosinophilic variants. Thorax. 2002;57(3):231.

20. Vandenplas O, Lantin A-C, D'Alpaos V, Larbanois A, Hoet P, Vandeweerdt M, et al. Time trends in occupational asthma in Belgium. Respiratory Medicine. 2011;105(9):1364-72.

21. Tarlo SM, Czyrka A, Ribeiro M, Vernich L, Luce CE, Liss G. Diisocyanate (iso) And Non-Diisocyanate (n-Iso) Sensitizer- Induced Occupational Asthma (oa) Frequency During 2003-07 In Ontario, Canada. American Journal of Respiratory and Critical Care Medicine. 2013;187.

22. Baur X, Chen Z, Liebers V. Exposure-response relationships of occupational inhalative allergens. Clinical and Experimental Allergy. 1998;28(5):537-44.

23. Executive HaS. Principles and guidelines to assist HSE in its judgements that duty-holders have reduced risk as low as reasonably practicable {online} 2001 [Available from: http://www.hse.gov.uk/risk/theory/alarp1.htm.

24. Executive HaS. EH40/2005 Workplace Exposure Limits. In: Executive HaS, editor. Containing the list of workplace exposure limits for use with the Control of Substances Hazardous to Health Regulations 2002 (as amended). Third Edition ed. Norwich, UK: TSO (The Stationary Office); 2018. p. 1-63.

25. Galea KS, Van Tongeren M, Sleeuwenhoek AJ, While D, Graham M, Bolton A, et al. Trends in Wood Dust Inhalation Exposure in the UK, 1985-2005. Annals of Occupational Hygiene. 2009;53(7):657-67.

26. Executive HaS. Work-related asthma statistics in Great Britain, 2019.: Health and Safety Executive; 2019.

27. Seed MJ, Carder M, Gittins M, Money A, Fishwick D, Barber CM, et al. Emerging trends in the UK incidence of occupational asthma: should we be worried? Occupational and environmental medicine. 2019;76(6):396-7.

28. Fishwick D, Barber CM, Bradshaw LM, Ayres JG, Barraclough R, Burge S, et al. Standards of care for occupational asthma: an update. Thorax. 2012;67(3):278.

29. Government U. The Control of Substances Hazardous to Health Regulations 2002. London: UK Government; 2002.

30. Gordon SB, Curran AD, Murphy J, Sillitoe C, Lee G, Wiley K, et al. Screening questionnaires for bakers' asthma—are they worth the effort? 1997;47(6):361-6.

31. Townsend MC. Spirometry in the Occupational Health Setting - 2011 Update. Journal of Occupational & Environmental Medicine. 2011;53(5):569-84.

32. Lange P. ÇY, Ingebrigtsen T. S., Vestbo J., Marott J.L. Long-term prognosis of asthma, chronic obstructive pulmonary disease, and asthma-chronic obstructive pulmonary disease overlap in the Copenhagen City Heart study: a prospective population-based analysis. Lancet Respiratory Medicine. 2016;4(6):454-62.

33. Redlich CA, Tarlo SM, Hankinson JL, Townsend MC, Eschenbacher WL, Von Essen SG, et al. Official American Thoracic Society technical standards: spirometry in the occupational setting. American journal of respiratory and critical care medicine. 2014;189(8):983-93.

34. Sumner J, Robinson E, Bradshaw L, Lewis L, Warren N, Young C, et al. Underestimation of spirometry if recommended testing guidance is not followed. Occupational Medicine. 2018;68(2):126-8.

35. Fishwick D, Sen D, Barker P, Codling A, Fox D, Naylor S. Health surveillance for occupational asthma in the UK. Occupational Medicine. 2016;66(5):365-70.

36. Curran AD. The role of nitric oxide in the development of asthma. International Archives of Allergy and Immunology. 1996;111(1):1-4.

37. Robinson D, Humbert M, Buhl R, Cruz AA, Inoue H, Korom S, et al. Revisiting Type 2‐high and Type 2‐low airway inflammation in asthma: current knowledge and therapeutic implications. Clinical & Experimental Allergy. 2017;47:161-75.

38. Mummadi SR, Hahn PY. Update on Exhaled Nitric Oxide in Clinical Practice. Chest. 2016;149(5):1340-4.

39. Wagener A, de Nijs S, Lutter R, Sousa A, Weersink E, Bel E, et al. External validation of blood eosinophils, FENO and serum periostin as surrogates for sputum eosinophils in asthma. Thorax. 2015;70(2):115-20.

40. Coman I, Lemière C. Fractional Exhaled Nitric Oxide (FeNO) in the Screening and Diagnosis Work-Up of Occupational Asthma. Current Treatment Options in Allergy. 2017;4(2):145-59.

41. Tossa P, Paris C, Zmirou-Navier D, Demange V, Acouetey D-S, Michaely J-P, et al. Increase in Exhaled Nitric Oxide Is Associated with Bronchial Hyperresponsiveness among Apprentices. American Journal of Respiratory and Critical Care Medicine. 2010;182(6):738-44.

42. Baatjies R, Jeebhay MF. Sensitisation to cereal flour allergens is a major determinant of elevated exhaled nitric oxide in bakers. Occupational and Environmental Medicine. 2013;70(5):310-6.

43. Van Der Walt A, Baatjies R, Singh T, Jeebhay MF. Environmental factors associated with baseline and serial changes in fractional exhaled nitric oxide (FeNO) in spice mill workers. Occupational and Environmental Medicine. 2016;73(9):614-20.

44. Adisesh L, Kharitonov S, Yates D, Snashell D, Newman-Taylor A, Barnes P. Exhaled and nasal nitric oxide is increased in laboratory animal allergy. Clinical and Experimental Allergy. 1998;28(7):876-80.

45. Jonaid BS, Pronk A, Doekes G, Heederik D. Exhaled nitric oxide in spray painters exposed to isocyanates: effect modification by atopy and smoking. Occupational and Environmental Medicine. 2014;71(6):415-22.

46. Dweik RA, Boggs PB, Erzurum SC, Irvin CG, Leigh MW, Lundberg JO, et al. An official ATS clinical practice guideline: interpretation of exhaled nitric oxide levels (FE_{NO}) for clinical applications. American Journal of Respiratory and Critical Care Medicine. 2011;184(5):602-15.

47. Travers J, Marsh S, Aldington S, Williams M, Shirtcliffe P, Pritchard A, et al. Reference ranges for exhaled nitric oxide derived from a random community survey of adults. Americal Journal of Respiratory and Critical Care Medicine. 2007;176(3):238-42.

48. Excellence NIfHaC. NG80: Asthma: diagnosis, monitoring, and chronic asthma management. NICE; 2017.

49. (SCOEL) TSCoOEL. Recommendation from the Scientific Committee on Occupational Exposure Limits: Risk assessment for Wood Dust. Brussels: European Commission: Employment, Social Affairs & Inclusion; 2003. Contract No.: SCOEL/SUM/102 final.

50. Kauppinen T, Vincent R, Liukkonen T, Grzebyk M, Kauppinen A, Welling I, et al. Occupational exposure to inhalable wood dust in the member states of the European Union. Annals of Occupational Hygiene. 2006;50(6):549-61.

51. Federation BW. British Woodworking Federation: industry statistics {online} 2018 [Available from: https://www.bwf.org.uk/publications/market-research/.

52. Black N, Dilworth M, Summers N. Occupational exposure to wood dust in the British woodworking industry in 1999/2000. Annals of Occupational Hygiene. 2007;51(3):249-60.

53. Simpson A. HS, Easterbrook E., and Wheeler J. Pilot project to research the need to update HSE on the occupational health risks in the woodworking industry. Health and Safety Executive; 2014. Contract No.: RR1011.

54. Ramazzini B. Diseases of Workers. 1st Ed ed. Chicago, Illinois: The University of Chicago Press; 1940.

55. Davidson JM. Toxic effects of iroko: An African wood. The Lancet. 1941;237(6124):38-9.

56. Hadfield EH. A study of adenocarcinoma of the paranasal sinuses in woodworkers in the furniture industry. Annals of the Royal College of Surgeons of England. 1970;46(6):301-19.

57. Chan-Yeung M. Fate of occupational asthma. A follow-up study of patients with occupational asthma due to Western Red Cedar (Thuja plicata). American Review of Respiriratory Disease. 1977;116(6):1023-9.

58. Chan-Yeung M, Lam S, Koener S. Clinical features and natural history of occupational asthma due to western red cedar (Thuja plicata). The American Journal of Medicine. 1982;72(3):411-5.

59. Torén K, Järvholm B. Effect of Occupational Exposure to Vapors, Gases, Dusts, and Fumes on COPD Mortality Risk Among Swedish Construction Workers: A Longitudinal Cohort Study: A Longitudinal Cohort Study. Chest. 2014;145(5):992-7.

60. IARC. IARC Monographs: Review of Human Carcinogens. Wood Dust. Lyon; France: International Agency for Research on Cancer; 2012. Contract No.: IARC Monographs - 100C.

61. Hubbard R, Lewis S, Richards K, Johnston I, Britton J. Occupational exposure to metal or wood dust and aetiology of cryptogenic fibrosing alveolitis. Lancet. 1996;347(8997):284-9.

62. Navaratnam V, Fleming KM, West J, Smith CJP, Jenkins RG, Fogarty A, et al. The rising incidence of idiopathic pulmonary fibrosis in the UK. Thorax. 2011;66(6):462.

63. Rake C, Gilham C, Hatch J, Darnton A, Hodgson J, Peto J. Occupational, domestic and environmental mesothelioma risks in the British population: a case-control study. British Journal of Cancer. 2009;100(7):1175-83.

64. Belin L. Sawmill alveolitis in Sweden. International Archives of Allergy and Applied Immunology. 1987;82(3-4):440-3.

65. Dykewicz MS, Laufer P, Patterson R, Roberts M, Sommers HM. Woodman's disease: hypersensitivity pneumonitis from cutting live trees. Journal of Allergy and Clinical Immunology. 1988;81(2):455-60.

66. Jones AR, S. Evans, G. . Review of Plant Dust and Chemical Respiratory Sensitizers. Health and Safety Laboratory: Health and Safety Laboratory; 2004. Contract No.: HE/03/06.

67. Ayars GH, Altman LC, Frazier CE, Chi EY. The toxicity of constituents of cedar and pine woods to the pulmonary epithelium. Journal of Allergy and Clinical Immunology. 1989;83(3):610-8.

68. Frew A, Chan H, Dryden P, Salari H, Lam S, Chan-Yeung M. Immunologic studies of the mechanisms of occupational asthma caused by western red cedar. Journal of Asthma and Clinical Immunology. 1993;92(3):466-78.

69. Rosenberg C, Liukkonen T, Kallas-Tarpila T, Ruonakangas A, Ranta R, Nurminen M, et al. Monoterpene and wood dust exposures: Work-related symptoms among Finnish sawmill workers. American Journal of Industrial Medicine. 2002;41(1):38-53.

70. Frew AJ, Chan H, Lam S, Chan-Yeung M. Plicatic acid induces histamine release from bronchial mast cells and basophils of patients with Western Red Cedar asthma. American Review of Respiratory Disease. 1992;145(4 PART 2):A21-A.

71. Chan-Yeung M. Mechanism of occupational asthma due to western red cedar (Thuja plicata). American Journal of Industrial Medicine. 1994;25(1):13-8.

72. Chan-Yeung M, Obata H, Dittrick M, Chan H, Abboud R. Airway inflammation, exhaled nitric oxide, and severity of asthma in patients with western red cedar asthma. American Journal of Respiratory and Critical Care Medicine. 1999;159(5):1434-8.

73. Kespohl S, Sander I, Merget R, Petersen A, Meyer HE, Sickmann A, et al. Identification of an obeche (Triplochiton scleroxylon) wood allergen as a class I chitinase. Allergy. 2005;60(6):808-14.

74. Ricciardi L, Fedele R, Saitta S, Tigano V, Mazzeo L, Fogliani O, et al. Occupational asthma due to exposure to iroko wood dust. Annals of Allergy Asthma & Immunology. 2003;91(4):393-7.

75. Kespohl S, Ochmann U, Maryska S, Nowak D, Bruening T, Raulf-Heimsoth M. Cross-reactive carbohydrate determinants as diagnostic tool for specification of occupational wood dust sensitisation. Allergy. 2007;62:282-.

76. Reijula K, Kujala V, Latvala J. Sauna builders' asthma caused by obeche. Thorax. 1994;49(6):622-3.

77. Skovsted TA, Schlunssen V, Schaumburg I, Wang P, Staun-olsen P, Skov PS. Only few workers exposed to wood dust are detected with specific IgE against pine wood. Allergy. 2003;58(8):772-9.

78. Mandryk J, Alwis KU, Hocking AD. Work-related symptoms and dose-response relationships for personal exposures and pulmonary function among woodworkers. American Journal of Industrial Medicine. 1999;35(5):481-90.

79. Milanowski J, Gora A, Skorska C, Krysinska-Traczyk E, Mackiewicz B, Sitkowska J, et al. Work-related symptoms among furniture factory workers in Lublin region (eastern Poland). Annals of Agricultural and Environmental Medicine. 2002;9(1):99-103.

80. Alexandersson R, Hedenstierna G. Pulmonary function in wood-workers exposed to formaldehyde - A prospective study. Archives of Environmental Health. 1989;44(1):5-11.

81. Jacobsen G, Schaumburg I, Sigsgaard T, Schlunssen V. Non-malignant respiratory diseases and occupational exposure to wood dust. Part I. Fresh wood and mixed wood industry. Annals of Agricultural and Environmental Medicine. 2010;17(1):15-28.

82. Jacobsen G, Schaumburg I, Sigsgaard T, Schlunssen V. Non-malignant respiratory diseases and occupational exposure to wood dust. Part II. Dry wood industry. Annals of Agricultural and Environmental Medicine. 2010;17(1):29-44.

83. Executive HaS. Work-related asthma in Great Britain, 2018. 2018.

84. Moher D, Liberati A, Tetzlaff J, Altman DG. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. PLoS Med. 2009;6(7):e1000097.

85. Network SIG. SIGN 50: A guideline developer's handbook. Edinburgh, Scotland: SIGN; 2011.

86. Liddle JW, M. Irwing, L. . Method for Evaluating Research and Guideline Evidence. Sydney, Australia: NSW Department of Health; 1996.

87. Nicholson PJ. How to undertake a systematic review in an occupational setting. Occupational and Environmental Medicine. 2007;64(5):353-8.

88. Schlunssen V, Sigsgaard T, Schaumburg I, Kromhout H. Cross-shift changes in FEV1 in relation to wood dust exposure: the implications of different exposure assessment methods. Occupational and Environmental Medicine. 2004;61(10):824-30.

89. Norrish AE, Beasley R, Hodgkinson EJ, Pearce N. A study of New Zealand wood workers: exposure to wood dust, respiratory symptoms, and suspected cases of occupational asthma. New Zealand Medical Journal. 1992;105(934):185-7.

90. Alwis KU, Mandryk J, Hocking AD. Exposure to biohazards in wood dust: bacteria, fungi, endotoxins, and (1-->3)-beta-D-glucans. Applied occupational and environmental hygiene. 1999;14(9):598-608.

91. Rongo LMB, Besselink A, Douwes J, Barten F, Msamanga GI, Dolmans WMV, et al. Respiratory symptoms and dust exposure among male workers in small-scale wood industries in Tanzania. Journal of Occupational and Environmental Medicine. 2002;44(12):1153-60.

92. Jacobsen G, Schlunssen V, Schaumburg I, Sigsgaard T. Increased incidence of respiratory symptoms among female woodworkers exposed to dry wood. European Respiratory Journal. 2009;33(6):1268-76.

93. Schlunssen V, Schaumburg I, Andersen NT, Sigsgaard T, Pedersen OF. Nasal patency is related to dust exposure in woodworkers. Occupational and Environmental Medicine. 2002;59(1):23-9.

94. Schlunssen V, Schaumburg I, Taudorf E, Mikkelsen AB, Sigsgaard T. Respiratory symptoms and lung function among Danish woodworkers. Journal of Occupational and Environmental Medicine. 2002;44(1):82-98.

95. Bohadana AB, Massin N, Wild P, Toamain JP, Engel S, Goutet P. Symptoms, airway responsiveness, and exposure to dust in beech and oak wood workers. Occupational and Environmental Medicine. 2000;57(4):268-73.

96. Pisaniello DL, Tkaczuk MN, Owen N. Occupational wood dust exposures, life-style variables, and respiratory symptoms. Journal of Occupational and Environmental Medicine. 1992;34(8):788-92.

97. Sripaiboonkij P, Phanprasit W, Jaakkola MS. Respiratory and skin effects of exposure to wood dust from the rubber tree Hevea brasiliensis. Occupational and Environmental Medicine. 2009;66(7):442-7.

98. Lipscomb HJ, Dement JM. Respiratory diseases among union carpenters: Cohort and case-control analyses. American Journal of Industrial Medicine. 1998;33(2):131-50.

99. Heikkila P, Martikainen R, Kurppa K, Husgafvel-Pursiainen K, Karjalainen A. Asthma incidence in wood-processing industries in Finland in a register-based population study. Scandinavian Journal of Work Environment & Health. 2008;34(1):66-72.

100. Schlunssen V, Schaumburg I, Heederik D, Taudorf E, Sigsgaard T. Indices of asthma among atopic and non-atopic woodworkers. Occupational and Environmental Medicine. 2004;61(6):504-11.

101. Perez-Rios M, Ruano-Ravina A, Etminan M, Takkouche B. A meta-analysis on wood dust exposure and risk of asthma. Allergy. 2010;65(4):467-73.

102. Ahman M, Holmstrom M, Ingelmansundberg H. Inflammatory markers in nasal lavage fluid from industrial arts teachers. American Journal of Industrial Medicine. 1995;28(4):541-50.

103. Shamssain MH. Pulmonary function and symptoms in workers exposed to wood dust. Thorax. 1992;47(2):84-7.

104. Carosso A, Ruffino C, Bugiani M. Respiratory diseases in woodworkers. British Journal of Industrial Medicine. 1987;44(1):53-6.

105. Talini D, Monteverdi A, Benvenuti A, Petrozzino M, Di Pede F, Lemmi M, et al. Asthma-like symptoms, atopy, and bronchial responsiveness in furniture workers. Occupational and Environmental Medicine. 1998;55(11):786-91.

106. Jacobsen GH, Schlunssen V, Schaumburg I, Sigsgaard T. Cross-shift and longitudinal changes in FEV1 among wood dust exposed workers. Occupational and Environmental Medicine. 2013;70(1):22-8.

107. Glindmeyer HW, Rando RJ, Lefante JJ, Freyder L, Brisolara JA, Jones RN. Longitudinal respiratory health study of the wood processing industry. American Journal of Industrial Medicine. 2008;51(8):595-609.

108. Jacobsen G, Schluenssen V, Schaumburg I, Taudorf E, Sigsgaard T. Longitudinal lung function decline and wood dust exposure in the furniture industry. European Respiratory Journal. 2008;31(2):334-42.

109. Schlunssen V, Kespohl S, Jacobsen G, Raulf-Heimsoth M, Schaumburg I, Sigsgaard T. Immunoglobulin E-mediated sensitization to pine and beech dust in relation to wood dust exposure levels and respiratory symptoms in the furniture industry. Scandinavian Journal of Work Environment & Health. 2011;37(2):159-67.

110. Eastman C, Schenker MB, Mitchell DC, Tancredi DJ, Bennett DH, Mitloehner FM. Acute Pulmonary Function Change Associated With Work on Large Dairies in California. Journal of Occupational and Environmental Medicine. 2013;55(1):86-91.

111. Glass WI, Power P, Burt R, Fishwick D, Bradshaw LM, Pearce NE. Work-related respiratory symptoms and lung function in New Zealand mussel openers. American Journal of Industrial Medicine. 1998;34(2):163-8.

112. Obata H, Dittrick M, Chan H, Chan-Yeung M. Sputum eosinophils and exhaled nitric oxide during late asthmatic reaction in patients with western red cedar asthma. European Respiratory Journal. 1999;13(3):489-95.

113. Dressel H, Gross C, de la Motte D, Sueltz J, Joerres RA, Nowak D. Educational intervention decreases exhaled nitric oxide in farmers with occupational asthma. European Respiratory Journal. 2007;30(3):545-8.

114. Maestrelli P, Boschetto P, Fabbri LM, Mapp CE. Mechanisms of occupational asthma. The Journal of Allergy and Clinical Immunology. 2009;123(3):531-42.

115. Kopferschmitt-Kubler MC, Ameille J, Popin E, Calastreng-Crinquand A, Vervloet D, Bayeux-Dunglas MC, et al. Occupational asthma in France: a 1-yr report of the Observatoire National de Asthmes Professionnels project. European Respiratory Journal. 2002;19(1):84-9.

116. Elder D, Abramson M, Fish D, Johnson A, McKenzie D, Sim M. Surveillance of Australian workplace Based Respiratory Events (SABRE): notifications for the first 3.5 years and validation of occupational asthma cases. Occupational Medicine-Oxford. 2004;54(6):395-9.

117. Wiggans RE, Evans G, Fishwick D, Barber CM. Asthma in furniture and wood processing workers: a systematic review. Occupational Medicine. 2016;66(3):193-201.

118. Malo JL, Cartier A, Desjardins A, Van de Weyer R, Vandenplas O. Occupational asthma caused by oak wood dust. Chest. 1995;108(3):856-8.

119. Lee LT, Tan KL. Occupational asthma due to exposure to chengal wood dust. Occupational Medicine-Oxford. 2009;59(5):357-9.

120. Chan-Yeung M, Leriche J, Maclean L, Lam S. Comparison of cellular and protein changes in bronchial lavage fluid of symptomatic and asymptomatic patients with red cedar asthma on follow-up examination. Clinical Allergy. 1988;18(4):359-65.

121. Cartier A. New causes of immunologic occupational asthma, 2012-2014. Current Opinion Allergy Clinical Immunology. 2015;15(2):117-23.

122. Campo P, Aranda A, Diaz-Perales A, Dona I, Ruiz M, Lisbona F, et al. Measurement of specific IgE to obeche wood dust (Triplochiton scleroxylon) in carpentry apprentices. Allergy. 2010;65:124-.

123. Stocks SJ, McNamee R, Turner S, Carder M, Agius RM. Assessing the impact of national level interventions on workplace respiratory disease in the UK: part 2-regulatory activity by the Health and Safety Executive. Occupational and Environmental Medicine. 2013;70:483-90.

124. Bohadana AB, Hannhart B, Ghezzo H, Teculescu D, Zmirou-Navier D. Exhaled nitric oxide and spirometry in respiratory health surveillance. Occupational Medicine-Oxford. 2011;61(2):108-14.

125. Wenzel S. Asthma phenotypes: the evolution from clinical to molecular approaches. Nature Medicine. 2012;18(5):716-25.

126. Lemiere C, Nguyen S, Sava F, D'Alpaos V, Huaux F, Vandenplas O. Occupational asthma phenotypes identified by increased fractional exhaled nitric oxide after exposure to causal agents. Journal of Allergy and Clinical Immunology. 2014;134(5):1063-7.

127. Executive HaS. Enforcement data: methods and quality statement {online} 2017 [Available from: hse.gov.uk/statistics/data-quality-statement.htm.

128. Douwes J, McLean D, Slater T, Pearce N. Asthma and other respiratory symptoms in New Zealand pine processing sawmill workers. American Journal of Industrial Medicine. 2001;39(6):608-15.

129. Elms J, Robinson E, Rahman S, Garrod A. Exposure to flour dust in UK bakeries: current use of control measures. Annals of Occupational Hygiene. 2005;49(1):85-91.

130. MATLAB. MATLAB and Statistics Toolbox Release 2012b ed. Natick, Massachusetts, United States: The MathWorks, Inc.

131. Bestall J, Paul E, Garrod R, Garnham R, Jones P, Wedzicha J. Usefulness of the Medical Research Council (MRC) dyspnoea scale as a measure of disability in patients with chronic obstructive pulmonary disease. Thorax. 1999;54(7):581-6.

132. Schatz M, Sorkness CA, Li JT, Marcus P, Murray JJ, Nathan RA, et al. Asthma Control Test: reliability, validity, and responsiveness in patients not previously followed by asthma specialists. Journal of Allergy and Clinical Immunology. 2006;117(3):549-56.

133. Miller MR, Hankinson J, Brusasco V, Burgos F, Casaburi R, Coates A, et al. Standardisation of spirometry. European Respiratory Journal. 2005;26(2):319-38.

134. Executive HaS. General methods for the gravimetric analysis of respirable, thoracic and inhalable aerosols: MDHS14/4. Bootle, Liverpool: Health and Safety Executive; 2014.

135. Statistics NOf. Standard Occupational Classification 2010 (SOC2010). Volume 2: the structure and coding index2010.

136. Research WIfE. Cascot: Computer Assisted Structured Coding Tool {online} 2015 [Available from: https://warwick.ac.uk/fac/soc/ier/software/cascot/.

137. Swanney MP, Ruppel G, Enright PL, Pedersen OF, Crapo RO, Miller MR, et al. Using the lower limit of normal for the FEV 1 / FVC ratio reduces the misclassification of airway obstruction. Thorax. 2008;63(12):1046-51.

138. Kerkhof M, Dubois AEJ, Postma DS, Schouten JP, Monchy JGR. Role and interpretation of total serum IgE measurements in the diagnosis of allergic airway disease in adults. Allergy. 2003;58(9):905-11.

139. Nerpin E, Olivieri M, Gislason T, Olin AC, Nielsen R, Johannessen A, et al. Determinants of fractional exhaled nitric oxide in healthy men and women from the European Community Respiratory Health Survey III. Clinical and Experimental Allergy. 2019;49(7):969-79.

140. Oliveros JC. Venny. An interactive tool for comparing lists with Vann's diagrams {online} 2007-2015 [Available from: https://bioinfogp.cnb.csic.es/tools/venny/.

141. Inc. S. IBM SPSS Statistics for Windows. Version 23. ed. Armonk:NY.: IBM Corp; 2015.

142. Armstrong BG. Effect of measurement error on epidemiological studies of environmental and occupational exposures. Occupation and Environmental Medicine. 1998;55(10):651-6.

143. Pearce N, Pekkanen J, Beasley R. How much asthma is really attributable to atopy? Thorax. 1999;54(3):268.

144. Hatcher JL, Cohen SD, Mims JW. Total serum immunoglobulin E as a marker for missed antigens on in vitro allergy screening. International forum of allergy & rhinology. 2013;3(100):782-7.

145. Tungu AM, Bråtveit M, Mamuya SH, Moen BE. Reduction in respiratory symptoms among cement workers: a follow-up study. Occupational Medicine. 2015;65(1):57-60.

146. Le Moual N, Kauffmann F, Eisen E, Kennedy SM. The healthy worker effect in asthma - Work may cause asthma, but asthma may also influence work. Am J Respir Crit Care Med. 2008;177(1):4-10.

147. Campo P, Aranda A, Rondon C, Dona I, Lisbona F, Rodriguez-Bada J, et al. Atopy, Specific Sensitization and Respiratory Symptoms In Apprentices of Furniture Industry Exposed to Diisocyanates And Wood Dust. Journal of Allergy and Clinical Immunology. 2010;125(2):AB119-AB.

148. Schlunssen V, Skovsted TA, Schaumburg I, Skov PS, Sigsgaard T. Wood dust sensitization among Danish woodworkers. American Journal of Industrial Medicine. 2004;46(4):408-9.

149. Li C-Y, Sung F-C. A review of the healthy worker effect in occupational epidemiology. Occupational medicine. 1999;49(4):225-9.

150. Chan-Yeung M. Immunologic and nonimmunologic mechanisms in asthma due to western red cedar (Thuja plicata). Journal of Allergy and Clinical Immunolology. 1982;70(1):32-7.

151. Upchurch S, Harris JM, Cullinan P. Temporal changes in UK birth order and the prevalence of atopy. Allergy. 2010;65(8):1039-41.

152. Kespohl S, Kotschy-Lang N, Tomm JM, von Bergen M, Maryska S, Bruening T, et al. Occupational IgE-Mediated Softwood Allergy: Characterization of the Causative Allergen. International Archives of Allergy and Immunology. 2012;157(2):202-8.

153. Torén K, Murgia N, Schiöler L, Bake B, Olin A-C. Reference values of fractional excretion of exhaled nitric oxide among non-smokers and current smokers. BMC pulmonary medicine. 2017;17(1):118.

154. Olin AC, Alving K, Toren K. Exhaled nitric oxide: relation to sensitization and respiratory symptoms. Clinical and Experimental Allergy. 2004;34(2):221-6.

155. Vandenplas O, Suojalehto H, Aasen TB, Baur X, Burge PS, de Blay F, et al. Specific inhalation challenge in the diagnosis of occupational asthma: consensus statement. European Respiratory Journal. 2014;43(6):1573-87.

156. Blanco I, Diego I, Bueno P, Fernandez E, Casas-Maldonado F, Esquinas C, et al. Geographical distribution of COPD prevalence in Europe, estimated by an inverse distance weighting interpolation technique. Int J Chronic Obstr Pulm Dis. 2018;13:57-67.

157. Knuchel-Takano A, Hunt D, Jaccard A, Bhimjiyani A, Brown M, Retat L, et al. Modelling the implications of reducing smoking prevalence : the benefits of increasing the UK tobacco duty escalator to public health and economic outcomes. Tobacco Control. 2017;27(e2):e124-e9.

158. Macfarlane E, Chapman A, Benke G, Meaklim J, Sim M, McNeil J. Training and other predictors of personal protective equipment use in Australian grain farmers using pesticides. Occupational and Environmental Medicine. 2008;65(2):141.

159. Turkeli A. History of Metal Casting {online} University of Marmaris2011 [Available from: http://mimoza.marmara.edu.tr/~altan.turkeli/files/cpt-1-history_of_metal_casting.pdf.

160. David Hey MO, and Martin Liddament. Forging The Valley. 1st Edition ed. Sheffield: Sheffield Academic Press; 1997.

161. Federation CM. http://www.castmetalsfederation.com/about {online} 2015 [Available from: http://www.castmetalsfederation.com/about.

162. Cooke J. SA, Yates T., and Llewellyn D. Exposure to substances hazardous to health in foundries. Bootle, Liverpool: Health and Safety Executive; 2017.

163. Löfstedt H, Westberg, H., Seldén, A. I., Rudblad, S., Bryngelsson,I., Ngo, Y., Svartengren, M. Nasal and Ocular Effects in Foundry Workers Using the Hot Box Method. Journal of Occupational and Environment Medicine. 2011;53(1):43-38.

164. Paris C, Ngatchou-Wandji J, Luc A, McNamee R, Bensefa-Colas L, Larabi L, et al. Work-related asthma in France: Recent trends for the period 2001- 2009. Occupational and Environmental Medicine. 2012;69(6):391-7.

165. Baur X, Bakehe P. Allergens causing occupational asthma: an evidence-based evaluation of the literature. International archives of occupational and environmental health. 2014;87(4):339-63.

166. Andjelkovich DA JD, Brown MH , Richardson RB , Miller FJ. Mortality of iron foundry workers: IV. Analysis of a subcohort exposed to formaldehyde. Journal of Occupational and Environmental Medicine. 1995;37(7):826-37.

167. Arif A, Delclos G. Association between cleaning-related chemicals and work-related asthma and asthma symptoms among healthcare professionals. Occupational and Environmental Medicine. 2012;69(1):35-40.

168. Burton C, Bradshaw L, Agius R, Burge S, Huggins V, Fishwick D. Medium-density fibreboard and occupational asthma. A case series. Occupational Medicine-Oxford. 2011;61(5):357-63.

169. Lee J. S. KHS, Choi B. S., and Park S. Y. A Case of Occupational Asthma in a Plastic Injection Process Worker. Annals of Occupational and Environmental Medicine. 2013;25:25.

170. Kim CW, Song JS, Ahn YS, Park SH, Park JW, Noh JH, et al. Occupational asthma due to formaldehyde. Yonsei Medical Journal. 2001;42(4):440-5.

171. Franko J, Jackson LG, Hubbs A, Kashon M, Meade BJ, Anderson SE. Evaluation of Furfuryl Alcohol Sensitization Potential Following Dermal and Pulmonary Exposure: Enhancement of Airway Responsiveness. Toxicological Sciences. 2012;125(1):105-15.

172. Hamzah NA, Mohd Tamrin SB, Ismail NH. Metal dust exposure and lung function deterioration among steel workers: an exposure- response relationship. International Journal of Occupational and Environmental Health. 2016;22(3):224-32.

173. Cherry NM, Hoyle J, Burgess G, Dippnall M, Smedley J, Pickering CAC, et al. Occupational Asthma in Foundry Workers. Annals of Occupational Hygiene. 2002;46(suppl1):373-6.

174. Liss GM, Bernstein DI, Moller DR, Gallagher JS, Stephenson RL, Bernstein IL. Pulmonary and immunologic evaluation of foundry workers exposed to methylene diphenyldiisocyanate (MDI). The Journal of Allergy and Clinical Immunology. 1988;82(1):55-61.

175. Servet K, Umit T, Halit C, Aziz G, Nurhan K. Prevalence of Occupational Asthma and Respiratory Symptoms in Foundry Workers. Pulmonary Medicine. 2013;2013(2013).

176. Löfstedt H, Westberg H, Seldén AI, Bryngelsson I, Svartengren M. Respiratory Symptoms and Lung Function in Foundry Workers Using the Hot Box Method: A 4-Year Follow-Up. Journal of Occupational and Environment Medicine. 2011;53(12):1425-9.

177. Nasrullah M, Mazurek JM, Wood JM, Bang KM, Kreiss K. Silicosis Mortality With Respiratory Tuberculosis in the United States, 1968–2006. American Journal of Epidemiology. 2011;174(7):839-48.

178. Rosenman K, Reilly M, Rice C, Hertzberg V, Tseng C-Y, Anderson H. Silicosis among foundry workers. Implication for the need to revise the OSHA standard. American Journal of Epidemiology. 1996;144(9):890-900.

179. Rosenman K, Reilly M, Henneberger PK. Estimating the total number of newly-recognized silicosis cases in the United States. Am J Ind Med. 2003;44(2):141-7.

180. Burge P. Occupation and chronic obstructive pulmonary disease (COPD). European Respiratory Journal. 1994:1032-4.

181. Rodríguez V, Tardón A, Kogevinas M, Prieto CS, Cueto A, García M, et al. Lung cancer risk in iron and steel foundry workers: a nested case control study in Asturias, Spain. American journal of industrial medicine. 2000;38(6):644-50.

182. Barbinova L, Baur X. Increase in exhaled nitric oxide (eNO) after work-related isocyanate exposure. International Archives of Occupational and Environmental Health. 2006;79(5):387-95.

183. Piipari R, Piirila P, Keskinen H, Tuppurainen M, Sovijarvi A, Nordman H. Exhaled nitric oxide in specific challenge tests to assess occupational asthma. European Respiratory Journal. 2002;20(6):1532-7.

184. Yacoub MR, Malo JL, Labrecque M, Cartier A, Lemière C. Occupational eosinophilic bronchitis. Allergy: European Journal of Allergy and Clinical Immunology. 2005;60(12):1542-4.

185. Hendrick DJ, Lane DJ. Occupational formalin asthma. British Journal of Industrial Medicine. 1977;34(1):11.

186. Pazdrak K, Górski P, Krakowiak A, Ruta U. Changes in nasal lavage fluid due to formaldehyde inhalation. International Archives of Occupational and Environmental Health. 1993;64(7):515-9.

187. Koskela K, Oksa PM, PhD, Sauni RM, PhD, Linnainmaa MP, Toivio PM, Lehtimaki LM, Prof, et al. Pulmonary Inflammation in Foundry Workers. Journal of Occupational & Environmental Medicine. 2015;57(2):124-8.

188. Franklin P, Dingle P, Stick S. Raised exhaled nitric oxide in healthy children is associated with domestic formaldehyde levels. Am J Respir Crit Care Med. 2000;161(5):1757-9.

189. Cotton R. aUR. Analysis of CT1 Foundry Dataset. Norwich: Health and Safety Executive; 2009.

190. Ulvestad B, Lund MB, Bakke B, Djupesland PG, Kongerud J, Boe J. Gas and dust exposure in underground construction is associated with signs of airway inflammation. European Respiratory Journal. 2001;17(3):416-21.

191. Cartier A, Malo JL, Doepner M, Nieboer E, Evans S, Dolovich J. Occupational asthma caused by nickel sulfate. The Journal of Allergy and Clinical Immunology. 1982;69(1):133-.

192. Löfstedt H, Westberg H, Seldén AI, Lundholm C, Svartengren M. Respiratory symptoms and lung function in foundry workers exposed to low molecular weight isocyanates. American Journal of Industrial Medicine. 2009;52(6):455-63.

193. AJ T. Respiratory irritants encountered at work. Thorax. 1996;51(5):541-5.

194. Fell AKM, Notø H, Skogstad M, Nordby K-C, Eduard W, Svendsen MV, et al. A cross- shift study of lung function, exhaled nitric oxide and inflammatory markers in blood in Norwegian cement production workers. Occupational and Environmental Medicine. 2011;68(11):799.

195. Coughlin SS. Recall bias in epidemiologic studies. Journal of Clinical Epidemiology. 1990;43(1):87-91.

196. Kuo HW, Chang CL, Liang WM, Chung BC. Respiratory abnormalities among male foundry workers in central Taiwan. 1999;49(8):499-505.

197. Iwatsubo Y, Matrat M, Brochard P, Ameille J, Choudat D, Conso F, et al. Healthy worker effect and changes in respiratory symptoms and lung function in hairdressing apprentices. Occupational and environmental medicine. 2003;60(11):831-40.

198. Fishwick DaC, A. Variability in the diagnosis of occupational asthma and implications for clinical practice. Current Opinion in Allergy and Clinical Immunology. 2008;8(2):140-4.

199. Meredith SK, Bugler J, Clark RL. Isocyanate exposure and occupational asthma: a case-referent study. Occupational and Environmental Medicine. 2000;57(12):830.

200. Belvisi MG, Baker K, Malloy N, Raemdonck K, Dekkak B, Pieper M, et al. Modelling the asthma phenotype: impact of cigarette smoke exposure. Respiratory research. 2018;19(1):89.

201. Johnson A, Moira CY, Maclean L, Atkins E, Dybuncio A, Cheng F, et al. Respiratory abnormalities among workers in an iron and steel foundry. British Journal of Industrial Medicine. 1985;42(2):94.

202. Fishwick D, Darby A, Hnizdo E, Barber C, Sumner J, Barraclough R, et al. COPD Causation and Workplace Exposures: An Assessment of Agreement among Expert Clinical Raters. Copd-Journal of Chronic Obstructive Pulmonary Disease. 2013;10(2):172-9.

203. Pronk A, Preller L, Doekes G, Wouters IM, Rooijackers J, Lammers JW, et al. Different respiratory phenotypes are associated with isocyanate exposure in spray painters. European Respiratory Journal. 2009;33(3):494-501.

204. Jarvis J SM, Stocks SJ, Agius RM. . A refined QSAR model for prediction of chemical asthma hazard. Occupational Medicine. 2015;23(8):659-66.

205. Michaud D. BM, and Perrault G. Characterization of Airborne Dust from Cast Iron Foundries by Physico-Chemical Methods and Multivariate Statistical Analyses. Air and Waste. 1993;43(5):729-35.

206. Feary J, and Cullinan, Paul. Laboratory animal allergy: a new world. Current Opinion in Allergy and Clinical Immunology. 2016;16(2):107-12.

207. Jones M. Laboratory Animal Allergy in the Modern Era. Current Allergy and Asthma Reports. 2015;15(12):1-8.

208. Nieuwenhuijsen MJ, Putcha V, Gordon S, Heederik D, Venables KM, Cullinan P, et al. Exposure- response relations among laboratory animal workers exposed to rats. Occupational and Environmental Medicine. 2003;60(2):104.

209. Feary J, Schofield S, Fitzgerald B, Canizales J, Jones M, Cullinan P. Laboratory Animal Allergy In The Contemporary World. American Journal of Respiratory and Critical Care Medicine. 2017;195.

210. Office UH. Animal Testing and Research {online}: UK Home Office; 2013 [updated 2018. Available from: https://www.gov.uk/guidance/research-and-testing-using-animals.

211. Executive HaS. EH76: Control of Laboratory Animal Allergy. Norwich, UK: Health and Safety Executive; 2011.

212. Tafuro F, Selis L, Goldoni M, Stendardo M, Mozzoni P, Ridolo E, et al. Biomarkers of respiratory allergy in laboratory animal care workers: an observational study. International archives of occupational and environmental health. 2018;91(6):735-44.

213. Hewitt RS, Smith AD, Cowan JO, Schofield JC, Herbison GP, Taylor DR. Serial exhaled nitric oxide measurements in the assessment of laboratory animal allergy. Journal of Asthma. 2008;45(2):101-7.

214. Lemiere C. Exhaled nitric oxide as a screening tool for occupational asthma. International Journal of Tuberculosis and Lung Disease. 2014;18(6):634-.

215. Feary JR, Schofield SJ, Canizales J, Fitzgerald B, Potts J, Jones M, et al. Laboratory animal allergy is preventable in modern research facilities. European Respiratory Journal. 2019;53(6):1900171.

216. Cullinan P, Cook A, Gordon S, Nieuwenhuijsen MJ, Tee RD, Venables KM, et al. Allergen exposure, atopy and smoking as determinants of allergy to rats in a cohort of laboratory employees. European Respiratory Journal. 1999;13(5):1139-43.

217. Nicholson PJ, Mayho GV, Roomes D, Swann AB, Blackburn BS. Health surveillance of workers exposed to laboratory animal allergens. Occupational Medicine. 2010;60(8):591-7.

218. Cullinan P, Lowson D, Nieuwenhuijsen MJ, Gordon S, Tee RD, Venables KM, et al. Work related symptoms, sensitisation, and estimated exposure in workers not previously exposed to laboratory rats. Occupational and Environmental Medicine. 1994;51(9):589.

219. Palmberg L, Sundblad B-M, Lindberg A, Kupczyk M, Sahlander K, Larsson K. Long term effect and allergic sensitization in newly employed workers in laboratory animal facilities. Respiratory Medicine. 2015;109(9):1164-73.

220. Kruize H, Post W, Heederik D, Martens B, Hollander A, Van Der Beek E. Respiratory Allergy in Laboratory Animal Workers: A Retrospective Cohort Study Using Pre- Employment Screening Data. Occupational and Environmental Medicine. 1997;54(11):830-5.

221. Peng RD, Paigen B, Eggleston PA, Hagberg KA, Krevans M, Curtin-Brosnan J, et al. Both the variability and level of mouse allergen exposure influence the phenotype of the immune response in workers at a mouse facility. The Journal of Allergy and Clinical Immunology. 2011;128(2).

222. Larese Filon F, Drusian A, Mauro M, Negro C. Laboratory animal allergy reduction from 2001 to 2016: An intervention study. Respiratory Medicine. 2018;136:71-6.

223. Korpi A, Lappalainen S, Kaliste E, Kalliokoski P, Reijula K, Pasanen AL. A multi‐faceted approach to risk assessment of laboratory animal allergens at two facilities. American Journal of Industrial Medicine. 2007;50(2):127-35.

224. Soumagne T, Laveneziana P, Veil-Picard M, Guillien A, Claudé F, Puyraveau M, et al. Asymptomatic subjects with airway obstruction have significant impairment at exercise. Thorax. 2016;71(9):804-11.

225. Coultas DB, Mapel D, Gagnon R, Lydick EVA. The Health Impact of Undiagnosed Airflow Obstruction in a National Sample of United States Adults. American Journal of Respiratory and Critical Care Medicine. 2001;164(3):372-7.

226. Lemiere C, D'Alpaos V, Chaboillez S, Cesar M, Wattiez M, Chiry S, et al. Investigation of Occupational Asthma Sputum Cell Counts or Exhaled Nitric Oxide? Chest. 2010;137(3):617-22.

227. Baur X, Barbinova L. Latex allergen exposure increases exhaled nitric oxide in symptomatic healthcare workers. European Respiratory Journal. 2005;25(2):309-16.

228. Świerczyńska-Machura D, Krakowiak A, Wiszniewska M, Dudek W, Walusiak J, Pałczyński C. Exhaled nitric oxide levels after specific inahalatory challenge test in subjects with diagnosed occupational asthma. International journal of occupational medicine and environmental health. 2008;21(3):219-25.

229. Pedrosa M, Barranco P, Lopez-Carrasco V, Quirce S. Changes in Exhaled Nitric Oxide Levels After Bronchial Allergen Challenge. Lung. 2012;190(2):209-14.

230. Mason P, Scarpa MC, Guarnieri G, Giordano G, Baraldi E, Maestrelli P. Exhaled nitric oxide dynamics in asthmatic reactions induced by diisocyanates. Clin Exp Allergy. 2016;46(12):1531-9.

231. van der Walt A, Singh T, Baatjies R, Lopata AL, Jeebhay MF. Work-related allergic respiratory disease and asthma in spice mill workers is associated with inhalant chili pepper and garlic exposures. Occupational and Environmental Medicine. 2013;70(7):446-52.

232. Demange V, Bohadana A, Massin N, Wild P. Exhaled nitric oxide and airway hyperresponsiveness in workers: a preliminary study in lifeguards. BMC pulmonary medicine. 2009;9(1):53.

233. Lund MB, Øksne PI, Hamre R, Kongerud J. Increased nitric oxide in exhaled air: an early marker of asthma in non- smoking aluminium potroom workers? Occupational and Environmental Medicine. 2000;57(4):274.

234. Olin AC, Andersson E, Andersson M, Granung G, Hagberg S, Toren K. Prevalence of asthma and exhaled nitric oxide are increased in bleachery workers exposed to ozone. European Respiratory Journal. 2004;23(1):87-92.

235. Johansson SGO, Bieber T, Dahl R, Friedmann PS, Lanier BQ, Lockey RF, et al. Revised nomenclature for allergy for global use: Report of the Nomenclature Review Committee of the World Allergy Organization, October 2003. The Journal of Allergy and Clinical Immunology. 2004;113(5):832-6.

236. Abrahamsen R, Fell AKM, Svendsen MV, Andersson E, Torén K, Henneberger PK, et al. Association of respiratory symptoms and asthma with occupational exposures: findings from a population- based cross- sectional survey in Telemark, Norway. BMJ Open. 2017;7(3).

237. Executive HaS. Occupational Lung Diseases in Great Britain, 2018. Health and Safety Executive; 2018 October 2018.

238. Fishwick D, Harris-Roberts J, Robinson E, Evans G, Barraclough R, Sen D, et al. Impact of worker education on respiratory symptoms and sensitization in bakeries. Occup Med (Lond). 2011;61(5):321-7.

239. Guy GB, Tania M, Ken RB. Eosinophils in the Spotlight: Eosinophilic airway inflammation in nonallergic asthma. Nature Medicine. 2013;19(8):977.

240. Anees W, Catton C, Burge PS, Robertson AS. Induced sputum and exhaled nitric oxide in workers with occupational asthma. Thorax. 2000;55:A60-A.

241. Lehtimaki L, Csonka P, Makinen E, Isojarvi J, Hovi S-L, Ahovuo-Saloranta A. Predictive value of exhaled nitric oxide in the management of asthma: a systematic review. European Respiratory Journal. 2016;48(3):706-14.

242. Smit LAM, Heederik D, Doekes G, Wouters IM. Exhaled nitric oxide in endotoxin- exposed adults: effect modification by smoking and atopy. Occupational and Environmental Medicine. 2009;66(4):251.

243. Barnes PJ, Dweik RA, Gelb AF, Gibson PG, George SC, Grasemann H, et al. Exhaled nitric oxide in pulmonary diseases a comprehensive review. Chest. 2010;138(3):682-92.

244. Sastre J, Costa C, del Garcia Potro M, Aguado E, Mahillo I, Fernandez-Nieto M. Changes in exhaled nitric oxide after inhalation challenge with occupational agents. J Investig Allergol Clin Immunol. 2013;23(6):421-7.

245. Stayner L, Steenland K, Dosemeci M, Hertz-Picciotto I. Attenuation of exposure- response curves in occupational cohort studies at high exposure levels. Scandinavian Journal of Work, Environment & Health. 2003;29(4):317-24.

246. Jeal H, Draper A, Harris J, Taylor A, Cullinan P, Jones M. Modified Th-2 responses at high-dose exposures to allergen - Using an occupational model. American Journal of Respiratory and Critical Care Medicine. 2006;174(1):21-5.

247. Zielen S, Devillier P, Heinrich J, Richter H, Wahn U. Sublingual immunotherapy provides long‐term relief in allergic rhinitis and reduces the risk of asthma: A retrospective, real‐world database analysis. Allergy. 2018;73(1):165-77.

248. Stenton SC, Beach, Jr., Avery A, Hendrick D. The value of questionnaires and spirometry in asthma surveillance programmes in the workplace. Occupational Medicine - Oxford. 1993;43(4):203-6.

249. Lo DK, Beardsmore CS, Roland D, Richardson M, Yang Y, Danvers L, et al. Lung function and asthma control in school-age children managed in UK primary care: a cohort study. Thorax. 2020;75(2):101-7.

250. Lemiere C, Chaboillez S, Bohadana A, Blais L, Maghni K. Noneosinophilic responders with occupational asthma: A phenotype associated with a poor asthma prognosis. Journal of Allergy and Clinical Immunology. 2014;133(3):883-5.

251. Dressel H, Gross C, de la Motte D, Sueltz J, Joerres RA, Nowak D. Educational Intervention in Farmers With Occupational Asthma: Long-term Effect on Exhaled Nitric Oxide. Journal of Investigational Allergology and Clinical Immunology. 2009;19(1):49-53.

252. Huang Y, Yang MC. Associations between occupational inhalation risks and FeNO levels in airway obstruction patients: results from the National Health and Nutrition Examination Survey, 2007- 2012. International Journal of COPD. 2017:3085-93.

253. Meo S, Alsaaran Z, Alshehri M. Effect of exposure to cement dust on Fractional Exhaled Nitric Oxide (FeNO) in non-smoking cement mill workers. Eur Rev Med Pharmacol Sci. 2014;18(10):1458-64.

254. Excellence. NIfHaC. DG12: Measuring fractional exhaled nitric oxide concentration in asthma: NIOX MINO, NIOX VERO and NObreath. London; 2014.

255. Florentin A, Acouetey DS, Remen T, Penven E, Thaon I, Zmirou-Navier D, et al. Exhaled nitric oxide and screening for occupational asthma in two at-risk sectors: bakery and hairdressing. International Journal of Tuberculosis and Lung Disease. 2014;18(6):744-50.