Dust exposure and respiratory health among Tanzanian coffee factory workers

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Scientific environment

This research focused on respiratory health among workers exposed to dust and endotoxin in coffee factories. It involved disciplines of occupational medicine and occupational hygiene. It was carried out under the research group of Occupational and Environmental Medicine at the Department of Global Public Health and Primary Care, University of Bergen.

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Isaiah 65:21, 23-24

21 They will build houses and live in them, they will plant vineyards and eat their fruit.

23 They will not toil in vain, nor bear children destined to disaster, for they are the race of Yahweh's blessed ones and so are their offspring.

24 Thus, before they call I shall answer, before they stop speaking I shall have heard.

"Deo Gloria!!!" "To God be the Glory"

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Abstract

Introduction: Exposure to organic dust may cause detrimental effects to the respiratory system of exposed workers. Organic dust is commonly contaminated with microbes and their derivatives such as bacteria and endotoxin, fungi, moulds and beta glucan. Few studies on exposure and health effects have been performed in primary coffee factories. The studies showed that processes in primary coffee factories cause emission of high dust levels. Work in coffee factories has been associated with respiratory health impairment.

Coffee beans are of two main types; Arabica and Robusta. Before coffee is brought to the factory it is processed at the farm. At the farms the harvested Robusta coffee cherries are mostly dried under the sun (called dry pre-processing) while Arabica coffee cherries are depulped using water (called wet pre-process) and then dried as parchment coffee. At the end of harvest season, remaining Arabica coffee cherries are dried without being depulped (dry processed).

The processes in primary coffee factories involves the pre-cleaning of coffee beans, the mechanical removal of the mesocarp and endocarp layers to get green coffee beans (GCB), the grading of the GCB, mixing them to produce a homogenous mixture by a process known as bulking and then packing in 60 kg bags. Loading and unloading of unprocessed coffee (parchment or coffee cherries) is done manually. For some processes, the machines are fed manually by production workers.

Objective: The aim of this research was to determine dust exposure levels and to assess respiratory health of the production workers in Robusta and Arabica primary coffee factories.

Materials and Methods: The research was conducted in Tanzania (in the Kilimanjaro and Kagera regions) in four primary coffee factories (factories; A, B, C and D) in three studies (in 2008 2009, and 2010). In the first study (2008) personal total dust (n=44) was sampled at a rate of 2 L/min from the breathing zone of the worker using side kick Casella pumps connected to closed-faced 25 mm conductive cassettes fitted with cellulose acetate filters. The samples were analysed gravimetrically. In addition, five samples were taken on glass fibre filters as pilot samples for analysis of endotoxin. We also assessed respiratory symptoms using an American Thoracic Society (ATS) standardized questionnaire among the production workers (n= 79) in comparison with a control group from a beverage factory (n=71).

In the second and third studies personal total dust (n= 149) was sampled by same methods as in 2008, using closed-faced 37 mm plastic cassettes which were fitted with polycarbonate filters. Dust samples were gravimetrically measured and further analysed for endotoxin. Airway inflammation was assessed using NIOX MINO device in both studies. In the third study, lung function and respiratory symptoms were assessed among production workers (n=138) in four primary coffee factories and two control factories (n=120). A portable spirometer was used for lung function testing.

Results: Personal total dust levels ranged from 0.25 to 36 mg/m^3; geometric mean GM = 2.50mg/m^3. Seventeen per cent of the samples were above the occupational exposure limit. There was a high correlation between total dust and endotoxin (r = 0.62). Endotoxin levels were higher when processing dry pre-processed coffee (mainly Robusta GM=10,800 EU/m^3) than when processing wet pre-processed Arabica (GM=1,350 EU/m^3). All endotoxin samples exceeded the health-based recommended value of 90 EU/m^3. Using a mixed model analysis dry pre-processing was shown to increase the total dust and endotoxin levels by a factor of 2.5 and 7.2, respectively.

Chronic respiratory symptoms were higher among coffee workers than controls. Having at least one asthma symptom was higher among the coffee workers compared to controls with odds ratio (OR) of 4.3 (95% CI; 1.9 – 9.9). Robusta coffee workers had higher prevalence of asthma symptoms (38%) than Arabica coffee workers (12%) (OR; 3.5, 95% CI; 1.4 – 9.0). Lung function parameters were not significantly different between coffee workers and controls. Nevertheless, in a linear regression model, controlling for age, height and type of coffee, there was a decrease in FEV_1 and FEV_1/FVC ratio related to an increase in cumulative total dust and endotoxin. In second study (2009) coffee workers had higher FE_{NO} levels than the controls, however, this was not found in the subsequent year 2010.

Conclusion: This study revealed that work in coffee factory is associated with high dust and endotoxin exposure which may be associated with impairment of respiratory health. Processing dry pre-processed coffee increases the exposure levels significantly. Reduction of dust exposure is recommended.

List of publications

I. Sakwari G, Bråtveit M, Mamuya SHD & Moen BE (2011) Dust exposure and chronic respiratory symptoms among coffee curing workers in Kilimanjaro: a cross sectional study. *BMC Pulm Med.*; Nov. 24:11:54.

II. Moen BE, Sakwari G, Mamuya SH, Akwilina AV, Larsson L, Pehrson C, Mashalla YJ & Bråtveit M. (2012) Respiratory Inflammation Among Workers Exposed to Airborne Dust With Endotoxins in a Coffee Curing Factory. *J Occup Environ Med.*;54 (7):847-50.

III. Sakwari G, Mamuya SHD, Bråtveit M, Larsson L, Pehrson C & Moen BE. (2012) Personal Exposure to Dust and Endotoxin in Robusta and Arabica Coffee Processing Factories in Tanzania. *Ann Occup Hyg.*; 57(2)173-83.

IV. Sakwari G, Bråtveit M, Mamuya SHD & Moen BE. (2013) Respiratory Symptoms, fractional exhaled NO and lung function among workers in Robusta and Arabica coffee factories. (In Press *J Occup Environ Med*)

Abbreviations and Terms

ATS	American Thoracic Society
ACGIH	American Conference of Governmental Industrial Hygienists
BHR	Bronchial hyperresponsiveness
BOT	Bank of Tanzania
CFC	Closed-faced cassette
CFU	Coli forming units
ERS	European Respiratory Society
ERHCS	European Respiratory Health Community Survey
EU	Endotoxin Unit
FEV_1	Forced Expiratory Volume in One Second
FVC	Forced Vital Capacity
FE_{NO}	Fractional Exhaled Nitric Oxide
GCB	Green Coffee Beans
GM	Geometric Mean
L	Litre
LAL	Limulus Amoebocyte Lysate
MEF_{50}	Maximum Expiratory Flow at 50%
NIOH	Norwegian Institute for Occupational Health
OEL	Occupational Exposure Limit
OR	Odds Ratio
OSHA-TZ	Occupational Health and Safety Authority-Tanzania
PEFR	Peak Expiratory Flow Rate
PD_{20}	Provocative Dose for 20% Reduction in FEV_1
RPE	Respiratory protective equipment
SKC pump	Side Kick Casella pump
SPSS	Statistical Package for Social Science
TCB	Tanzania Coffee Board
TaCRI	Tanzania Coffee Research Institute
US	United States

UK United Kingdom

Aerodynamic diameter of a particle is the diameter of 1unit density sphere that will have a dropping velocity in the air similar to the particle in question/of interest.

Bulking is a process of mixing coffee in churning machines to get a homogenous distribution of coffee in a consignment.

Catador is a machine that pneumatically separates coffee beans by gravity. It separates heavy from fluffy or lighter beans.

Coffee curing is a term used to mean primary coffee processing.

Dry pre-processed coffee is the coffee that has been dried straight after harvest. This is known as the dried coffee cherries.

Endotoxin is a component on a cell wall of gram negative bacteria. Commonly found in organic dust.

Fractional exhaled nitric oxide is a biomarker of airway inflammation. It is a result of nitric oxide synthase enzyme from both constitutive and induced forms. The induced nitric oxide synthase significantly increases the exhaled nitric oxide.

Hopper is a funnel-shaped bin which is fed with coffee on the top side, stores the coffee temporarily and discharges it to the processing line from the bottom side.

Wet pre-processed coffee is the coffee that has been depulped, fermented and washed after harvest and then dried. This is also called parchment coffee.

Contents

1. Introduction

1.1 Coffee

Coffee comes from a perennial tree that grows in tropical zones above 1000 m altitude and in subtropical areas (1). Coffee is an important crop worldwide and it is used as a beverage in all countries in the world (2). The major producers of coffee worldwide are Brazil, Vietnam, Ethiopia, Columbia and Indonesia. The major producers and exporters of coffee in Africa are Ethiopia, Uganda, Ivory Coast, Cameroon and Tanzania. Tanzania exports about 1% of the coffee available at the global market (3). Most of the coffee growing countries export their coffee. Worldwide a large number of workers are employed in the coffee sector but exact numbers are difficult to find. In Tanzania the number of workers in coffee sector is estimated to be above 2 million (4).

1.1.1 Types of coffee and growing areas

There are two main species of coffee; *Coffea Arabica* (Arabica) and *Coffea Canephora* (Robusta), which make up 99% of the coffee at the global market (5). There are other species of coffee that are of lower quantity*: Mascarocoffea* grown in Madagascar and *Coffea Liberica*, grown in West and Central Africa (1) to mention a few.

The coffee species often grown in East Africa are Arabica and Robusta, and these are also the ones cultivated in Tanzania. Arabica coffee growing areas in Tanzania are Mara, Kilimanjaro, Arusha, Tanga, Mbeya, Ruvuma, Iringa, Njombe and Kigoma while Robusta is grown in Kagera (Fig.1).

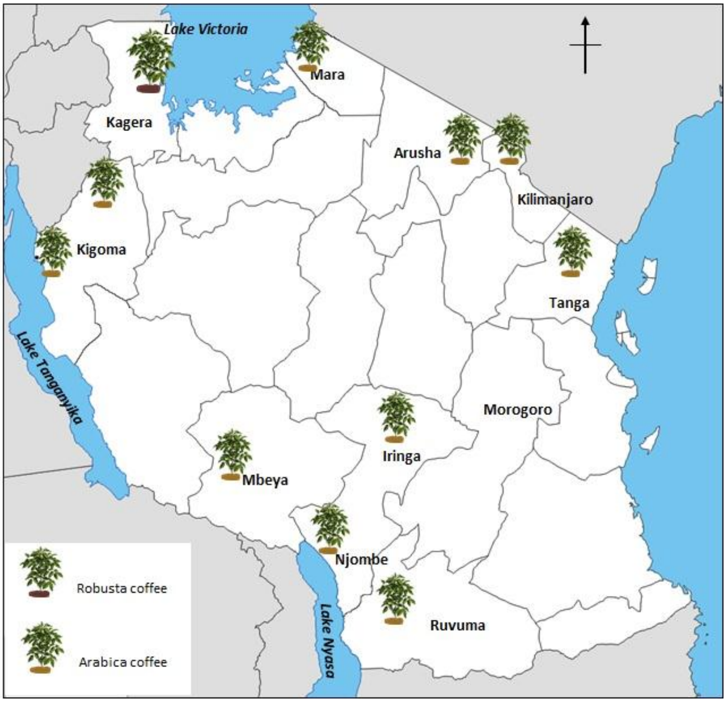

Figure 1: Map of Tanzania showing coffee growing areas.

1.2 Coffee history

History tells that coffee originated from Ethiopia (Abyssinia) from where it was taken to Yemen by Arab merchants earlier than 15th century. Coffee was later introduced to the East Indies by Dutch colonists in the 17th century and later from Java to the French colonies, from France to Latin America and back to Africa by missionaries in 19th century (2;6). This coffee was mostly Arabica species.

In 1880s missionaries introduced Arabica Coffee in the Morogoro region in Tanzania but it failed to grow. In 1898, German missionaries introduced Arabica plants in the rural Moshi district at Kilema Parish in the Kilimanjaro region (2;6). There it grew and spread across the slopes of Mount Kilimanjaro, Arusha and much later in 1970s it was introduced to the southern regions of Ruvuma and Mbeya.

Robusta has been grown in the north-western region of East Africa since the 18th century, before colonialism. A ceremonial ritual in the houses of chiefs or for an honoured guest in people's homes included the offering of green Robusta coffee bean to chew. This was practised by native people in central Africa and in Bunyoro Empire (6) (currently Uganda and a part of Tanzania called Kagera region). Later, the roasting of coffee was introduced in these areas during colonial era and the coffee business began. Chewing green coffee is still practised in the Kagera region and in coffee growing regions in Uganda.

Coffee production in Tanzania has fluctuated due to unstable world market prices and economies throughout the years. Production of coffee was highest in the 1960s to early 1980s when the coffee prices were high. After this time prices dropped and production decreased until the prices started to rise again in the late 1990s. Apart from the market prices, the annual coffee yield is dependent on the weather conditions—such as availability of adequate rainfall—in the particular season. It is estimated that the coffee produced in Tanzania is 48,000 tonnes annually, but may vary between 35,000 tonnes and 68,000 tonnes (4).

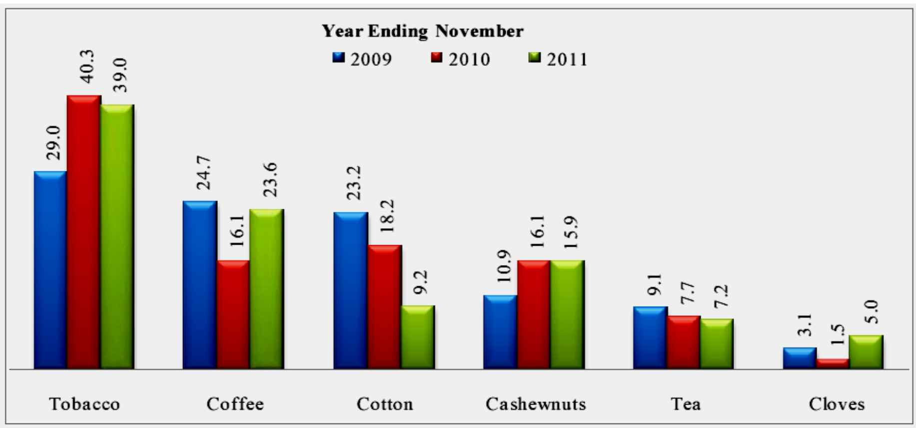

Figure 2: Triennial percentage of cash crop exports revenues in Tanzania. (Source: Bank of Tanzania Dec 2011) (7)

Currently coffee is the second most economically important cash crop in Tanzania that earns about 24% of revenues from cash crop export (7) (Fig. 2). About 90% of the exported coffee is from small scale farms, and 10 % is from the coffee estates (3;8). Coffee is a major source of income among coffee growers. It is evident to date that coffee producing areas have much better quality of living conditions compared to other communities (2).

1.3 Coffee processing

In Tanzania, the coffee season is once per year, while in other countries there can be two seasons per year. The coffee fruits (coffee cherries) are harvested by manual picking when they are ripen.

Coffee is processed in three stages before it is grounded and made ready to drink. After harvest, the coffee is first pre-processed at the farms (9). Secondly, the coffee is taken to the primary processing factories where it is hulled to get green coffee which is exported. Lastly, in the secondary processing factories usually located in the importing countries, the coffee is roasted and grounded (Fig. 3).

1.3.1 Pre-processing at farms

Immediately after the harvest, coffee cherries are pre-processed. In Tanzania Arabica coffee cherries are usually wet pre-processed while Robusta coffee cherries are dry pre-processed. At the end of the season the last harvest of Arabica can be dry pre-processed.

Wet pre-processing

In this stage Arabica coffee cherries are de-pulped in a machine using water on the same day of harvest. The coffee beans are fermented for 12-36 hours to remove the mucilage layer (a sticky, sugary layer around the coffee parchment cover), washed and dried on a wire mesh under the sun. The drying may take 7-14 days depending on the weather. The moisture content for properly dried beans must be 12.5% or less. The dried Arabica coffee beans are called parchment coffee. These are sold to merchants or collected by farmers' union group and are taken to the coffee factories for primary processing i.e. hulling and grading.

Dry pre-processing

Tanzanian Robusta coffee and occasionally the last harvests of Arabica coffee (1% of Arabica coffee) are dry pre-processed. After the harvest, the coffee cherries are cleaned (farmers pick out leaves and the raw cherries), and are spread in a layer of 10cm or less on the earth or concrete floor (most often on mats over the earth ground in Tanzania). The cherries are spread out in the morning, carefully mixed during the day and heaped together in the evening. The drying process may take fourteen days or more, depending on the weather conditions. As for Arabica, the moisture content has to be 12.5% or less. The dried Robusta and/or dried Arabica are called dried cherries. These are sold and are brought to the primary factories for further processing.

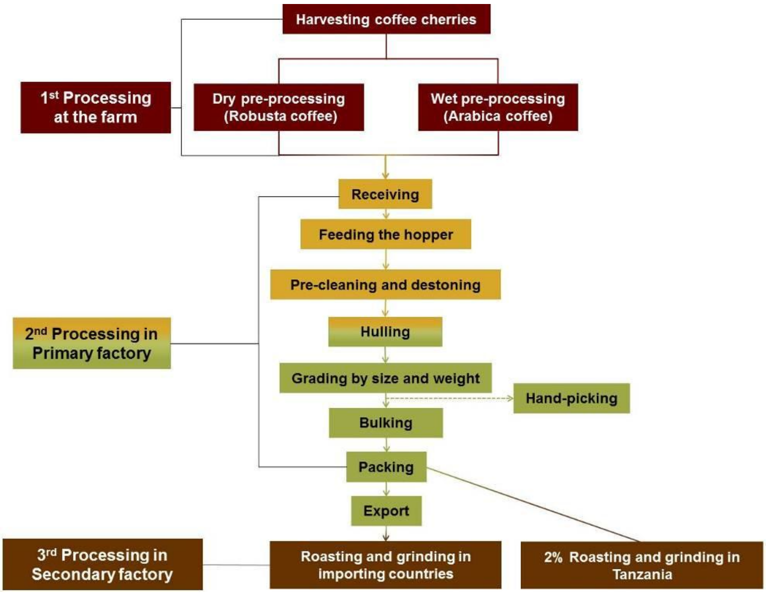

Figure 3: Schematic diagram for coffee processing.

1.3.2 Processing in primary coffee factories

The farmers bring the coffee to the primary coffee factories through farmers' unions or private buyers. Robusta coffee cherries and Arabica parchment coffee is pre-cleaned, destoned, hulled, graded by size and weight, and bulked to get a homogenous mixture of green coffee beans (GCB) the state in which coffee is exported . The GCB for export are packed in 60 kg bags. The activities in coffee processing factories are similar regardless of the type of machinery used or the type of coffee.

Handling parchment and coffee cherries in a primary coffee factory

This first activity in the coffee curing process involves measuring the moisture content of the coffee being received, and then the parchment or dried cherries bags from the trucks are manual unloaded, weighed and stacked in a storage area. Some factories have bucket elevators which transport coffee bags to the stacking storey where they are manually stacked. The parchment or dried cherries bags produce dust upon handling.

Feeding the hopper and hulling

In this stage, the coffee bags are cut and the coffee is poured on top of the hopper. The hopper is a chute at the very beginning of coffee processing where coffee is poured in and held temporarily to assure a continuous supply of coffee to the processing line. One or two workers feed the coffee into the hopper and monitor it to maintain a constant flow of coffee into the hopper. In some factories coffee is poured gradually from a short distance from the bags, while in other factories, the bags are emptied vigorously from a higher level (Fig. 4).

Figure 4: Pouring coffee in the hopper.

The coffee is then conveyed by gravity drop or by bucket elevators to the pre-cleaner machines, where all debris is removed mechanically by sieve plates. Thereafter cleaned coffee is conveyed by bucket elevators to the destoner where the stones and all non-coffee heavy objects are removed through mechanical sieves. The debris are collected by the workers and taken to the waste disposal. The cleaned coffee is then conveyed by bucket elevators to the huller machine. The hullers are machines that break and peel the pericarp, mesocarp and endocarp layers from the parchment coffee and the coffee cherries to get the green coffee beans.

The empty bags are folded and returned to the farmers or to the coffee estates. Jute bags are used in Arabica coffee growing areas and polypropylene bags in Robusta areas. Dust is produced from the coffee when it is poured from the bags, fed into the hopper, and from the pre-cleaners, the destoners, huller machines and when folding the emptied bags.

Sorting by size and grading by weight of green coffee beans

The green coffee is sorted by size on a table grader or roller grader. The table grader machine is a newer method while a roller grader is an old method, and it is still used in the old factories. The table grader has sieving plates with different pore size which separate coffee in different sizes giving rise to different grades from best to low grade; peaberries, AA, AB, C, TT, E and UG.

Figure 5: Work at the gravity table.

After the coffee has been sorted by size, the heavy beans are separated from the lighter beans. This process is done on a gravity table (Fig. 5) or by a machine called a catador.

Dust can be produced especially from the table and gravity graders as the machines are shaking continuously and are not enclosed.

Bulking of green coffee beans

Bulking is a process whereby different coffee grades, or coffee from different farmers is mixed to get a homogenous mixture. Upon request, coffee is bulked according to the specifications provided; while for normal bulking, the whole batch is mixed. In some factories, bulking of coffee is done at regular basis; while in others, it is done upon request from the coffee owner or buyer. In some factories coffee is manually fed into the bulking machines (Fig. 6).

Figure 6: Pouring coffee in the bulking machine.

Hand picking

Sometimes, the coffee might require hand picking. This is a manual process performed by women (Fig. 7). The women remove destroyed, broken or discoloured beans that were not removed previously by the machines. Coffee has to be hand-picked at least twice by two different women. In some factories, hand picking is performed by three different women before it can be bagged. One factory has a colour separator machine that is used instead of hand picking. This machine rejects discoloured beans out of the production line. This is rarely used.

Figure 7: Hand picking.

Packing

Coffee that has been handpicked is fed into the bulking machine for mixing before it is conveyed by bucket elevators to the storage tanks above the bulking machines. The coffee is weighed automatically and the workers manually opened the bucket (Fig. 8) to pour the green coffee beans into the new 60 kg jute bags. The bags are sewn and stacked, ready for shipping.

Figure 8: Packing GCB.

Husk handling

Fine and coarse dust particles are produced during coffee processing. The dust is conveyed via pipes to husk storage tanks. One or two workers load the husks into bags that are then loaded into trucks or sometimes the workers shuffle the husk directly onto the trucks. The husks from Arabica coffee are used to make fuel blocks for cooking, and some husks are also used as fuel in the production of burnt bricks. Husks from Robusta coffee are used as a mulching layer in farms. Workers in the husk area are exposed to dust when unloading husk material from the tanks and loading it onto bags (Fig. 9) or trucks.

Figure 9: A worker filling sacks with husk.

Cleaning coffee grade F and Sweeping

In the sorting of green beans by size and weight, the smaller beans that are not restrained in any of the sieve plates, as well as much of the husk material and dust, are brought out of the production line and into bags. This coffee grade (grade F) is fed into a blower machine to remove fluffy beans, husks and dust. This activity is not necessary in the process line; it is done on request and mostly in the old plants. Workers who feed the hopper and those who monitor the destoner also carry out this task on alternate shifts. Production of dust in this task might be high since most of the husk material and dust are blown out of the machines.

Sweeping is a continuous activity since the spilled beans and dust make the floor slippery. Sweeping is done manually with dry brooms and at some factories with compressors which might lead to high dust exposure. In some of the factories there are workers who are hired as sweepers, but in others the production workers sweep their own working area.

1.3.3 Processing in the secondary factories

The secondary coffee processing factories are found in the importing countries and only seldom in coffee growing countries. For instance, there is only one roasting factory in Tanzania and 98% of all coffee is exported. The raw material for roasting is the green coffee beans (GCB). The exported GCB after arrival in the importing countries are poured mechanically into a hopper (10). The GCB are cleaned, by removing fine dust and undesired particles. The cleaned coffee is then roasted at high temperatures, grounded at different sizes, mixed and packed for consumption and exportation (10;11).

1.4 Coffee curing factories in Tanzania

In the 1920s, colonial rulers in Tanzania established the first primary coffee processing factory in the Kilimanjaro region. This factory was meant to process Arabica coffee from the whole of East Africa (Kenya, Tanzania and Uganda). Another primary coffee processing factory was established in 1935 in the Kagera region for Robusta coffee processing. In the 1970s other factories were established in Mbeya and Mbinga. At the time of this writing (2013) there are about 18 coffee processing factories in Tanzania; 4 in Kilimanjaro, 1 in Arusha, 4 in Kagera, 3 in Mbeya and 3 in Iringa, 1 in Ruvuma, 1 in Mara and 1 in Kigoma. There are other small-scale factories, but most send their coffee to the large factories due to high running costs.

1.5 Dust fraction and health-related sampling

Dust is composed of solid particles that become suspended in the air and are generated by the mechanical or natural disintegration of organic or inorganic materials (12;13) . The particles in dust are categorized in different sizes based on their aerodynamic diameter. This is the diameter of a sphere of 1 unit density that sediments at the same velocity in the air as the said particle (12;14). The aerodynamic diameter determines the behaviour of the particle, which is of importance in inhalation exposure.

The inhalable dust fraction is the fraction of total airborne particles that are inhaled through the nose and the mouth. Bacteria, fungi and some allergens may fall into this category. These may bring about upper airway symptoms such as nasal irritation and rhinitis (15;16). Total dust is a fraction that was used previously that has now been replaced by inhalable fraction. The inhalable fraction includes particles with sizes that are of respirable and thoracic fraction (17).

Thoracic dust fraction includes the inhaled particles that can reach beyond the larynx in the respiratory system, with estimated aerodynamic diameter median of 11.64 µm. Particles of this fraction deposit on the airways: the bronchi and bronchioles and may cause effects like bronchitis and bronchial constriction. The respirable dust fraction includes the particles that will reach the alveolar region and they have an estimated aerodynamic diameter median of 4.25µm. However, very small particles (<0.5 µm aerodynamic diameter) may be expired out during normal breathing as they remain suspended in the air for a longer time (17;18), however, some will remain and can be very harmful to the lung tissue (14;17). Only fibres can reach the alveoli, even if they have large aerodynamic diameter when they move in streamline along the bronchioles (12).

Sampling of dust is also based on dust fraction, that is, inhalable dust, thoracic dust and respirable dust. For exposures that have very fine dust the respirable fraction is preferred, while for exposures that brings effect to the airways and lung tissue inhalable fraction is preferred. The sampled inhalable fraction will include 50% of particles with an aerodynamic diameter of ≤ 100 µm. Sampling the thoracic fraction will include 50% particles that have an aerodynamic diameter of 10 µm or less, while the respirable fraction will include 50% particles with ≤ 4 µm (14;18;19).

1.6 Occupational exposure limit values for organic dust and endotoxin

Organic dust consists of particles of plant or animal origin that have become airborne. This may include flour, pollen, coffee tea wood dusts. Organic dust may also be contaminated with bacteria, fungi, moulds, viruses and their derivatives such as beta-glucan and endotoxin. Inhalation of organic dust might cause organic dust toxic syndrome (ODTS), hay fever, byssinosis, and airway inflammation leading to asthma and reduced lung function (20-22).

Endotoxin is a component of the cell wall of gram negative bacteria. Exposure to endotoxin has been associated with chronic respiratory symptoms, reduced lung function (23-25), asthma (26), and increased levels of FE_{NO} among non-atopic workers (27) Occupational exposures to endotoxin have been studied among farmers, workers in agricultural industries, waste management and from laboratories. The health effects have been observed at levels above 100 EU/m^3 (23;28;29).

Occupational exposure limit value (OEL) is the maximum concentration to which a person can be exposed with no or minimal adverse effects (14;17). In most countries, OELs are based on health, economic and technical feasibility of control measures. Even though the OEL may not provide protection for every worker from developing a disease these levels give protection to the majority (14). The OELs for organic dust have been established in many developed countries; for instance, the recommended occupational exposure limit in Norway is 5 mg/m^3 (30), expressed in total dust fraction. Sampling of the total dust fraction has been found to underestimate the inhalable fraction by a factor of 1.7 to 4 (31). However, some occupational exposure limits are still expressed in total dust values.

At the moment, there is no OEL for endotoxin. Recently, a health-based recommended value for endotoxin of 90 EU/m^3 was suggested in the Netherlands (32). These values have not been established in most of the developing countries. Hence, in this study the recommended values referred to are 5 mg/m^3 for organic dust and 90 EU/m^3 for endotoxin, respectively. This value is derived from inhalable dust fraction.

1.7 Exposure in coffee factories

The raw material in the primary coffee factories is parchment coffee or dried coffee cherries. The dust in coffee factories is considered to be mainly organic dust as coffee is of plant origin. This coffee dust might be contaminated with fungi, moulds, gram-positive and gram-negative bacteria as well as other materials resulting from contamination during pre-processing and storage. The microbial components have been isolated from coffee at different stages of processing; parchment, dried cherries and husks in primary coffee factories (5;33;34) and from the dust in green coffee warehouse (10) and silos (35). Furthermore, species of gram negative and gram positive bacteria have been found in personal total dust samples taken from a primary

Arabica coffee factory in Tanzania (36). High levels of bacteria were found when handling parchment coffee. Exposures to fungal spores were highest when sorting green coffee, compared to other activities (36). Hence, a worker in a coffee factory may be exposed to dust consisting of components from the coffee beans, such as allergens, as well as fungi and their mycotoxin, moulds and beta-glucans, bacteria and endotoxin (37;38).

1.7.1 Dust and endotoxin exposure levels in coffee factories in previous studies

Two studies have measured dust levels in primary coffee factories; one in Uganda and the other in Papua New Guinea. The total dust exposure ranged from 1 – 58 mg/m^3 (39;40). Dust exposure in secondary coffee factories was first documented in 1970s (41) in the former Yugoslavia (Croatia) and ranged from 10 – 63 mg/m^3 in the green coffee areas (Table 1). A study from New Orleans reported relatively low mean total dust levels in both green coffee (0.48mg/m^3) and roasted coffee (0.44mg/m^3) areas. In recent years inhalable dust has been measured in secondary factory silos and haulage, and these levels ranged from 1.0 – 15.6 mg/m^3 (35). A study in Swedish coffee factories reported dust levels higher than the Swedish occupational exposure limit of organic dust of 5mg/m^3

It has been reported that dust in primary coffee factories have particles with a diameter ranging from less than 5 µm to 400 µm (40), and in secondary factories from 0.4 µm to 100 µm (42). Exposure level in the factories presumably varies with processes, activities, machinery as well as the crop being processed (35). Factor that influence exposure levels have not been studied specifically in coffee factories. Several determinants of exposure, such as; dustiness of the process, activities involved, raw materials, design of machine, and weather conditions have been reported from a collection of agricultural industries (43). The determinants of exposure have also been reported in other specific industries such as potato industry (44), pig farms (45), grain farms (46) and in waste management (47).

Endotoxin exposure has been measured and reported in secondary coffee factories in the Netherlands (GM 134 EU/m^3; range 12.4 – 2030 EU/m^3) (43) but not in primary coffee factories. Microorganisms such as bacteria, fungi and their derivatives (Ochratoxin A) and moulds have been isolated from dust samples from secondary coffee factories, from parchment coffee and from dried coffee cherries from the farms (5;10;33).

The studies done in primary coffee factories did not describe activities and machinery in the factories. Furthermore, determinants for dust and endotoxin exposure in coffee factories were not reported. In order to reduce the exposure levels it is important to know the determinants to institute proper and effective control measures.

1.8 Respiratory health among coffee workers

Both chronic and acute respiratory symptoms have been presented among workers in primary and secondary coffee factories (Table 1).

1.8.1 Acute and chronic respiratory symptoms in primary coffee factories

High prevalence of acute respiratory symptoms was reported among coffee workers in primary coffee factories in Sri Lanka (48) and Uganda (39). These symptoms included cough, sneezing, difficulty in breathing and running nose. Rhinitis has also been reported as an acute effect among primary coffee factory workers in Uganda (39).

Chronic respiratory symptoms have been reported among workers in primary coffee factories in Uganda (39) and Papua New Guinea (40) where workers had higher prevalence of chronic cough with or without phlegm, dyspnoea and wheezing compared to the control groups (Table 1). However, in these studies there were some methodological uncertainties such as mixing both primary and secondary factory workers as well as Robusta and Arabica. These studies also lacked information about the participants such as age, duration of employment (39), description of workers' activity in the factories and incomplete description on the assessment of chronic respiratory symptoms (39;40). There is also difference in processing activities. The processes in Sri-Lanka are mainly manual while in Tanzania these are done by machines (48).

1.8.2 Acute and chronic respiratory symptoms in secondary coffee factories

The first study on acute respiratory symptoms among secondary coffee processing workers was carried out in 1950 among nine workers in US who had acute allergic rhinitis (49). Acute cough, sneezing and throat irritation were reported among Croatian male green coffee workers (41;50) in subsequent years.

Chronic respiratory symptoms among workers handling green coffee in secondary coffee factories were also reported in the study from 1950. Chronic respiratory symptoms were also reported in subsequent years among secondary coffee processing workers handling green coffee and roasting coffee. These were chronic cough with or without phlegm, chronic bronchitis, wheezing, dyspnoea and asthma (10;35;41;42;51) (Table 1).

A German study among workers handling imported green coffee found that when the workers were handling imported green Arabica coffee reported twice as much complains of respiratory symptoms than when they handled Robusta coffee (35). It has been discussed that coffee dust can give allergy. Recently, an allergen has been identified from Arabica green coffee bean (52). It is not known whether allergens in Arabica are comparable to those in Robusta coffee or if there are differences in allergic development from the two types of coffee beans. It has been suggested that the allergen concentration are higher in green coffee beans compared to roasted coffee beans (53). Hence, it is expected to be more sensitization and development of allergic reaction among those handling green coffee compared to roasted coffee.

Table 1: An overview of studies done in primary and secondary processing factories presenting respiratory health effects among coffee workers.

Year	Author(s)	Country	Study design	Group of workers included	Persons studied	Dust levels (mg/m^3)	Respiratory health Effects
Studies done in primary coffee processing factories							
1985	Smith D et al. (40)	New Papua Guinea	Cross sectional	parchment coffee processing workers	69	Total Dust 0.7-10	Chronic cough 8.7% vs. 1.5% in controls FEV_1 drop of >10% among 19% coffee workers vs. 4% controls
1988	Uragoda CG (48)	Sri Lanka		Parchment coffee processing workers	38	-	Prevalence of acute symptoms among coffee workers; cough 84% sneezing 74%, running nose 55%
1996	Sekimpi DK, Agaba EF, Okot-Nwang M, Ogaram DA. (39)	Uganda	Cross sectional	Parchment coffee processing workers and coffee roaster	959	Personal total dust 10.8 - 58 Stationary:- 0.1 – 25.7 Personal respirable dust:- 0.7 – 15.2	Chronic respiratory symptoms was 8.8% in coffee workers and 2.3% in controls OR_{adj} of 3.99 (95% CI; 1.2 – 12.9) Acute symptoms: rhinitis 29% vs. 6% OR_{adj} 6.08(95% CI; 2.9 – 12.7) Post shift decrease in FEV_1 2.67 vs. -1.35 and PEFR 1.21 vs. -1.97
				Non coffee workers	128	-	
Studies in Secondary coffee processing factories							
1950	Figley KD and Rawling FFA (49)	USA	Case study	Patients who worked in coffee roasting plants	7	-	Allergic rhinitis, asthma
				Non coffee factories	2		
1961	Kaye M and Freedman SO (54)	USA	Case study	Coffee manufacturing workers suspected allergic in contact with coffee	39	-	16/36- Asthmatic, 69% allergic to raw chaff, 46% to raw bean vs. non symptomatic who had no asthma, 12% allergic to raw chaff, 2% to raw bean
				coffee workers with no allergic symptoms in contact with coffee	56		-

Year	Author(s)	Country	Study design	Group of workers included	Persons studied	Dust levels (mg/m^3)	Respiratory health Effects
				Office workers or workers with no contact to coffee	17		
1970	Van Toon (55)	The Netherlands	Case study	Patient who worked in a coffee roasting factory > 20yrs	1	-	Alveolitis
1978	Karr RN (56)	New Orleans	Matched Case control study	Coffee factory workers and controls	8 8	-	
1979	Zuskin et al. (41)	Former Yugoslavia	Cross sectional	Green coffee workers and coffee roasters	72	Total dust 1.4 - 62.3 (AM=11.2) Respirable fraction average was 3%	Chronic cough 42% and chronic bronchitis 23% among exposed vs. 7% and 3% among controls. Change in FEV_1 and FVC was -2.1 % and -1.3% for green coffee workers (p=0.01&0.05)
1981	Zuskin E, Valic F, Kanceljak B. (57)	Zagreb Yugoslavia	Cross sectional	Green coffee workers and coffee roasters	45	Not measured	Chronic cough 40% coffee workers vs. 7% controls. Asthma 8.9% vs. 0. Pre and post shift MEF_{50} decrease (0.38 l/s) among coffee workers
1982	Osterman K, Zetterstrom O and Johansson SGO. (11)	Sweden	Cross sectional Study I Study II		25 129	5.9 -6.3	Sensitization was pronounced among green coffee workers than control; study I (67% vs. 27%), study II (50% vs. 22%)
1982	Jones RN et al. (51)	USA-New Orleans	Cross sectional	 Coffee processing workers Green coffee workers Mixed (green and roasters) coffee roasters	372	Mean total dust levels 0.25 - 0.48 0.44 and 0.48 0.25 and 0.43 0.40 and 0.48	Low respiratory symptoms were common among current smoking coffee workers 49% vs. 22% among ex-smokers and non-smokers. Chronic bronchitis was 7% smoking coffee workers vs. 3% non-smokers Residual FEV_1 was lower for those who had worked longer time in coffee factory

Year	Author(s)	Country	Study design	Group of workers included	Persons studied	Dust levels (mg/m^3)	Respiratory health Effects
1985	Zuskin E, Kanceljak B, Skuric Z, Butkovic D. (58)	Zagreb (Former Yugoslavia)	Cross sectional	Green coffee workers and coffee roasters	9	Not measured	4 of 9 workers who had + skin test had a reduction in VC at 5 min after inhaling green coffee. Acute reduction in FEV_1 among health subjects exposed to coffee dust
1991	Thomas KE et al. (10)	United Kingdom	Cross sectional	Green coffee warehouse workers and roasters	197	Total dust 3.2 Respirable 0.22 Total dust 1.3 Respirable 0.15	Wheeze, cough and dyspnoea was reported by 12.7% of the exposed workers
1993	Zuskin E, Schachter NE, Kanceljak B, Witek TJ, Jr., and Fein E (50)	Croatia	Cross sectional	Green coffee workers and controls	103	Total dust 0.77- 35.56 Respirable dust 0.12 – 3.5	Chronic cough 57 % among green coffee workers vs. 11% in controls, chronic bronchitis 52% vs. 8%, chronic phlegm 62% vs. 9% Acute symptoms; throat irritation 61.9% and cough 57% among the exposed workers.
1998	Larese F, Fiorito A, Casasola F et al. (42)	Italy	Cross sectional	Green coffee workers Roasters Office workers	31 37 44	Personal total dust for green coffee 0.07 – 0.3	20% (6 cases) GCB workers had work related asthma or rhinitis ($p<0.01$). 26% of the green coffee workers were sensitized to GCB vs. 3% roasted coffee workers and 4% clerk workers. Common allergic symptoms were similar in all groups. Lung function was similar in all groups
2009	Oldenburg M, Bittner C, Baur X (35)	Germany	Cross sectional	Coffee silos workers Haulage workers Decaffeinating workers	19 24 17	Inhalable dust 2.6 – 15.7	Coffee worker exposed to >10mg/m^3 had erythematous 21.4%, 25% rhinoconjuctivitis and significantly lower PD_{20} was needed for bronchial hyperresposiveness. Post week decrease in FEV_1 of 3.9% among those exposed to <=10mg/m^3 & increase of 1.4% among >10mg/m^3

1.8.3 Lung function changes and airway inflamation among coffee workers

Acute reduction in lung function parameters after the work shift has been reported among workers in one primary (39) and three secondary coffee factories (41;51;58). However, other studies have not observed any acute change across the shift (35;40).

Significant difference between the mean FEV_1 among coffee workers and controls has been reported among workers in primary factories in Papua New Guinea (40) as well as a decrease in FEV_1 and FVC across the working week i.e. from Monday morning before the work shift to Friday afternoon after the work shift among green coffee workers in Germany (35). The decreases in lung function have been reported with very low exposures in US (51) and very high exposure levels in Croatia (41). However, more often current exposure does not reflect the reduced lung function changes, but rather the previous exposure.

Exposure to organic dust has been shown to cause airway inflammation. This can be assessed in occupational settings by a non-invasive approach by measuring the amount of exhaled nitric oxide in the exhaled air (59). This has not been reported among coffee workers; nevertheless it has been reported among workers exposed to organic dust and endotoxin from other workplaces (27;60).

2. Rationale and objectives of the study

2.1 Rationale

Only two studies have measured dust exposures in primary coffee factories and only three studies have assessed aspects of respiratory health of the workers in such factories. None of these studies have been done in Tanzania despite the fact that coffee is one of the major contributors to the country's revenue. It is estimated that more than 2 million people are directly or indirectly employed in the Tanzanian coffee sector. Due to poor records it is not known exactly how many are employed in coffee factories and coffee storage facilities.

Even though dust in coffee factories is mainly of organic origin none of the previous studies have described the association between possible endotoxin exposure levels and respiratory health of workers in primary coffee factories. In order to reduce exposure levels among workers, effective control measures are important. Hence there is a need to address the exposure type, exposure levels and in addition factors that determine the exposure levels. There are no studies which have addressed exposure determinants in Robusta or Arabica factories.

Coffee dust may cause respiratory health problems. It is known that organic dust and endotoxin exposure might cause inflammation in the airways of exposed workers (27) leading to respiratory health impairment such as development of asthma, chronic respiratory symptoms and reduced lung function. Hence, this study was set to assess lung function and to measure an inflammatory biomarker in order to study the relationship between the exposure and respiratory health effects among primary coffee factory workers. More knowledge about these factors may provide important information in the implementation of preventive measures in the industry.

2.2 Objectives of the study

2.2.1 General objective

- To obtain knowledge on dust exposure and respiratory health of workers in primary coffee processing factories in Tanzania.

2.2.2 Specific objectives

- To measure personal total dust and endotoxin levels, and explore their exposure determinants in Robusta and Arabica primary coffee factories (Papers 1 & 3).

- To assess chronic respiratory symptoms among coffee workers compared to a control group (Papers 1 & 4).

- To examine and compare lung function and airway inflammation among Robusta and Arabica coffee workers and a control group (Papers 2 & 4).

3. Methods

3.1 Study area

This study was done in coffee factories in two of the coffee growing regions in Tanzania; Kilimanjaro and Kagera (Fig. 1). Due to differences in culture and socioeconomic status, one control factory was selected from each region.

There are four participating coffee factories namely A, B, C and D and two control factories; one beverage and one fish factory. Coffee factories A, B and the beverage factory are from an Arabica coffee growing area in the Kilimanjaro region, and factories C, D and the fish factory are from a Robusta growing area in the Kagera region. Another control group was made from a nearby hotel and hospital in the Kilimanjaro region.

3.2 Study design

This study is made up of three cross sectional studies conducted in four primary coffee factories in three time periods. Study I (2008), was done in the two Arabica coffee factories (factories A and B) (Paper 1). In this study, personal total dust samples were taken, and interviews for acute and chronic respiratory symptoms were conducted among the coffee workers. In addition, samples for endotoxin analysis (n=5) were taken as a pilot study (Fig. 10).

Based on the findings from study I, the second and third studies were developed. The second cross sectional study (2009) was conducted in factory A. In this study, personal total dust sampling was conducted in the coffee factory (Papers 3&4). Assessments of airway inflammation was performed among the non-smoking coffee workers and among a control made of workers from a nearby hotel and healthcare workers who were attending a seminar at a hotel (Paper 2).

The third cross sectional study was done in 2010 in four coffee factories (A, B, C, and D) and in two control factories (The beverage factory and the fish factory) (paper 3 & 4). This study included factories from both two types of coffee. In this study personal total dust sampling was done in three of the coffee factories (B, C and D) and in the two control factories. Interview on respiratory symptoms, lung function test and assessment of airway inflammation were performed in all factories (Fig. 10).

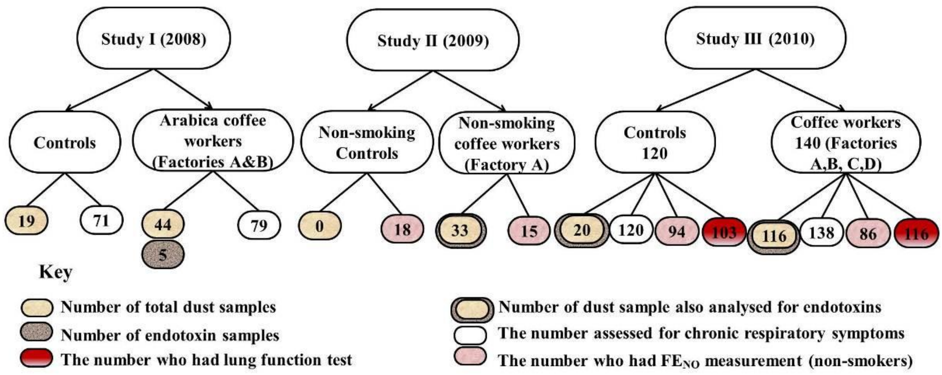

Figure 10: A schematic summary of data collection in coffee factories (A, B, C and D) and control factories (beverage factory, healthcare and hotel workers and fish factory).

3.2.1 Description of participating factories

Factory A

Factory A was established in 1997 in Moshi town with machines manufactured in 1996/97. On average, the factory hulls 2,500 - 5,500 tonnes of parchment coffee per season. The factory operates fully from July to March each year. They have mainly one shift from 8:00 to 17:00 with an exception during the peak season when they have two shifts. The factory has two premises close to each other; one for the factory and the other for the storage of both parchment and green coffee beans.

The huller and gravity table machines are fitted with exhaust pipes and hoods to reduce dust concentrations in the workplace atmosphere. General ventilation in the factory is of natural type with door openings, ventilation block layers on the walls and openings along the roof.

Factory B

Factory B is located in Moshi Town in the Kilimanjaro region. It was established in the 1920s, the first coffee factory in Tanzania. The factory processes an average of

5,000 tonnes of parchment coffee a year, which is 10% of its capacity. The factory hulls coffee from farmers' unions, small-scale coffee growers, private parchment coffee buyers and estate coffee growers in North East Tanzania.

The factory has three floors. The ground floor has machines for pre-cleaning, destoning, blowing grade F coffee and bulking. The first floor is equipped with hulling machines and catadors. There is also one hopper and a colour separator chamber. The second floor has the grading machines: the roller grader and gravity table. Catadors begin in this floor and proceed to the first floor. There is one hopper opening in this area as well. The remaining area is used for drying coffee that did not meet the humidity criteria.

The factory has both old and new machinery that on some occasions are operated simultaneously; however, the old machines are used frequently. There are ventilation block layers and ventilation fans on the walls. However, only one ventilation fan was working in 2008/09 and two were working in 2010 and these were operated intermittently. There are hoods in the bulking and pre-cleaning areas that were out of order.

Factory C

Factory C was established in 1935 in the Kagera region. In 2005, the production machines were substituted by newer ones (made in 1996/1997) in the hopper, pre-cleaners, hullers, and graders. The bulking machines were not changed. The factory operates from June to January/ March depending on the season. It operates on one shift 7:00to 17.00, and an average of 10 - 15 tonnes is processed in a day.

The factory has two separate production halls. The hopper and pre-cleaners are in hall one. The hullers, hand-hulled coffee hopper and gravity table are in hall two; with bulking machines and packing in an extended area of this hall. The destoners, hullers and gravity table are fitted with dust exhaust hoods. Ventilation is by natural cross ventilation. There is one ventilation fan in the huller area. Husks from the huller and fluffy cherries from the hopper are extracted by an exhaust pipe to the disposal area outdoors.

Factory D

Factory D is relatively new, established in 2005 in Kagera region, with machines made in 1986 and 1987, which are installed in one production hall. The factory is operated in two shifts, that is, from 7:00 to 18:00 and from 6:30 to 18:30 am. At an average both shifts process 100 tonnes per day.

Ventilation in factory D is through grid blocks, 0.5 x 1.5 m in size, which is along the outer walls, and also by a 0.15 m high opening of between the roof and two outer walls. There are also two wide open-doors on one of the walls. Husks and dust from the pre-cleaner, hullers and graders are conveyed by a pipe to the piling area at the rear side of the building.

The control groups

1.1.1.1.1 The beverage factory

The factory is located in the Kilimanjaro region in Moshi town, approximately 4 km from the coffee factories. The factory was established in 1997 and it produces beverages. Most of the works are mechanical. However, loading and unloading of boxes and crates on the production lines, and in trucks are done manually. The factory employs about 300 workers and about 150 causal workers. Heavy lifting and stacking is done by battery powered forklift trucks inside and by diesel powered forklift trucks outside the building.

The factory produces soft drink for the North East regions of Singida, Dodoma, Kilimanjaro, Arusha and Tanga.

1.1.1.1.2 The fish factory

This factory is situated about 4 km from the coffee factory in Bukoba – Kagera region. The factory has about 120 employees in the fish production line. The fish plant skins, debones, freezes and packs fresh and frozen fish for export. The fish are manually beheaded, degutted, skinned and are either conveyed on belts to a quick tunnel freezing machine or to manual packing as fresh fillets. All the packed fish are stored in cold rooms and freezers, ready to be transported.

Hotel and healthcare workers

The hospital where the controls in the second study work is 6km from the factory. The hotel is located near the factory. It offers restaurant and accommodation services. The workers are involved in housekeeping, serving and reception work. Cleaners, cooks were not included in the study.

3.3 Sample size

The sample size was different in the three studies. In first study sample size was calculated based on the prevalence of cough among coffee workers (42%) and among controls (7%) (41). A statistical power of 97% at a significant level of 0.05 gave an estimated sample size of 80 individuals in each of the exposed and the control groups.

The second study was a pilot study for exhaled nitric oxide, where all non-smoking coffee workers were included with a similar number of controls.

In the third study the three outcomes to be considered were the prevalence for chronic respiratory symptoms, FE_{NO} levels and lung function among coffee workers compared to a control group. The sample size was estimated based on lung function (FVC arithmetic mean 3.3 L, standard deviation (SD) 0.5 among exposed and 3.5 L (0.5) among controls) (61). This mean difference gave a sample size of 200 in the exposed group and 120 in the control group at a statistical power of 86% with a significant level of 0.05. This number was higher than any of the other outcomes of interest.

Our sampling unit was a production line worker in both coffee factories and control factories. In the second study, the sampling unit for control was a worker that had not been exposed to coffee dust or worked in any coffee factory.

3.4 Selection of participants

The number of employees in production lines in the coffee factories varies depending on the yield in the particular season. In the first study coffee factories from Kilimanjaro (A and B) had a total number of 45 (permanent production line workers) and 61 (permanent production line workers, including guards and supervisors who have offices in the production area), respectively. The number was significantly reduced in the second and third studies (27 and 45 production workers in A and B, respectively). The coffee factories in Kagera had 30 and 55 production worker in factories C and D respectively. The control factory in Kilimanjaro, a beverage factory, had 300 permanent employees and about 100 casual workers whereas the fish factory in Kagera had 120 workers. Only permanent employed workers were accessible for the studies.

All production workers in the coffee factories were invited to participate. In the exposure assessment, workers were selected randomly from section leaders' lists in factories B, C and D. In the first study, in factory A, participants with odd numbers were selected from the leaders list for dust sampling, while in factory B they were selected randomly from the section leader's list. In the second study, in factory A, only the non-smoking production workers were included. In the control factory, participants for both exposure and respiratory health assessment were randomly selected from different sections using the leader's list. In the control factory, a fraction of workers were randomly selected to fit the sample size required. In the second study a control group was made from non-smoking workers in a hotel near the factory (n=12) and health facility workers attending a seminar (n = 6).

The control groups in all studies were assumed to have a similar socioeconomic status to the coffee workers except for the hospital workers in the second study.

3.5 Exposure assessment and laboratory analyses

Personal total dust samples were collected in three studies during this research (Fig 10). In the first study samples collected were analysed for total dust (n=63) and endotoxin (n=5) as a pilot study.

In the second and third studies, the samples were analysed both gravimetrically and for endotoxin. In all studies, total dust samples were collected throughout the working shift from the breathing zone of the participant. Side Kick Casella (SKC) pumps were operated at a flow rate of 2 L/min. Most of the coffee workers rotate between different activities, hence the workers' activities during sampling was recorded.

3.5.1 Dust sampling and analyses first study

In the first study, three-piece closed-faced conductive 25 mm cassettes were used for personal total dust sampling (Fig. 12). Cellulose acetate filters of 0.8 µm pore size were used for total dust samples (n=45). Glass fibre filters of 0.2 µm pore size (n=5) were used to sample total dust which was analysed for endotoxin.

The dust samples were transported to Norway as hand luggage and then sent to Denmark for analysis at Eurofins Laboratory. These samples were gravimetrically analysed using a microbalance (AT261 Mettler Toledo) with a detection limit of 0.01 mg/m^3 for 8 hr. sampling time. The results were calculated as mg/m^3 using the sampled volume. One sample was not analysed since the pump stopped during working sampling.

Figure 11: Sampling of personal total dust during packing.

The endotoxin samples were stored in a refrigerator before being transported to Norway as hand luggage. They were then sent to the laboratory at the Norwegian

Institute of Occupational Health (NIOH), Oslo, for extraction and analysis of endotoxin.

Filters for endotoxin were immersed in non-pyrogenic water with 0.05% of Tween-20 and were shaken on a rotary shaker for 1hour. The endotoxin extract was analysed using the Kinetic chromogenic LAL test (62).

3.5.2 Dust sampling and analyses in the second and third studies

In the second and third studies, three-piece CFC, 37 mm diameter (Fig. 12), were used for total dust sampling. Polycarbonate filters with pore size of 0.8 µm were used.

In the second study dust sampling was conducted in factory A for three days when wet pre-processed Arabica coffee was processed and one day when dry pre-processed Arabica coffee was processed. In the third study dust was sampled when processing wet pre-processed Arabica in factory A and B and dry pre-processed Robusta factory C and D.

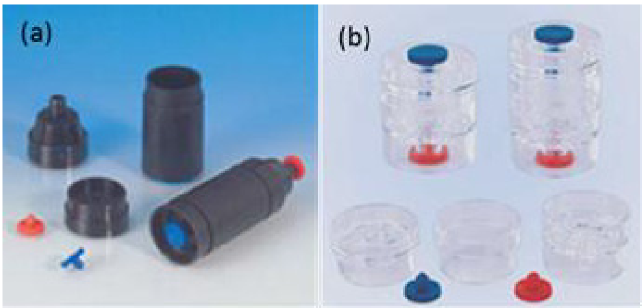

Figure 12: (a) Three piece conductive (25mm) cassettes and (b) three piece plastic (37mm) cassettes (Source: Pall Cooperation-Life Science).

Samples were stored at low temperatures after collection. Samples from the second study (n=33) were stored in a refrigerator and transported to Norway where they were kept frozen before transport to Lund Medical Laboratory for analysis. Samples from third study (n=116) were stored in a freezer at the end of each sampling day (-8º C). These were transported with freeze packs, to Dar es Salaam stored in a freezer and later transported to Norway packed with freeze packs. The samples were then sent to Lund medical laboratory in Sweden for analysis. The filters were kept frozen until the analysis day. Endotoxins were extracted from all the dust samples collected in second and third study. The analysis was by kinetic chromogenic Limulus Amebocyte lysate (LAL) test as described by Lane at al. 2004 (63).

3.6 Assessment of respiratory health

Respiratory health was assessed by questionnaire interview, lung function tests and measurement of FENO as a biomarker for airway inflammation.

3.6.1 Assessment of chronic respiratory symptoms

All participants who agreed to participate in the first and third study were interviewed on work history, education, previous respiratory illnesses, cooking fuel use, handling of pesticides, smoking habits, chronic respiratory symptoms and occurrence of asthma symptoms. Questions adapted from the ATS standardised questionnaire for assessing respiratory symptoms among adults were used (64) and in addition specific on questions asthma were used from the ERCHS questionnaire (65). All questions were translated to Kiswahili and translated back to English by different persons. The translated English version questionnaire was compared to the original version and any questions that did not have similar meanings were corrected (Appendix 2).

3.6.2 Assessment of airway inflammation

Fractional exhaled nitric oxide (FE_{NO}) was assessed as an inflammatory biomarker. It has been suggested that smoking lowers the levels of FE_{NO} (27); hence FE_{NO} was measured among the non-smokers only. All non-smokers who participated in respiratory symptom assessments were invited to participate. The assessment was done according to ATS/ERS guidelines (66) using a portable device, NIOX MINO (Aerocrine AB, Solna, Sweden), which measures FE_{NO} via an electrochemical sensor. However, due to time and economic constraints only one measurement was taken per person. Studies have shown that a single measurement using NIOX MINO is adequate (67). Room NO was <5 ppb on the days of FE_{NO} measurements and on occasions when the room NO was higher than 5 ppb FE_{NO} measurements were not performed. Recommended FE_{NO} values for adults have been established; FE_{NO} level <25 ppb is considered to be normal; 25-50 ppb represent intermediate values and should be interpreted with caution and levels above 50 ppb indicate persistently high values which may be related to the presence of airway inflammation (68). None of the participants were on corticosteroid medications.

3.6.3 Lung function testing

Figure 13: A participant performing lung function manoeuvre.

Lung function was measured by spirometer for all who agreed to participate in the study. The tests were performed in accordance with ATS guidelines for spirometry (69). The test was performed using a portable spirometer (SPIRARE3® sensor model SPS 320 by Diagnostica AS, Oslo, Norway) connected to a laptop. The manoeuvres were done in standing position. Each worker was asked to inhale deeply to maximum lung capacity and exhale forcefully into the device. Three successful manoeuvres were recorded and the best were retained for analysis (Fig. 12). The lung function parameters that were recorded were forced expiratory volume in one second (FEV_1), Forced vital capacity (FVC) and the ratio FEV_1/FVC.

3.7 Statistical analysis

Data were analysed using SPSS Windows version 15.0 and 18.0 (Paper 1) and IBM SPSS version 19 for Windows (Paper 2 – 4).

Categorical variables were compared using chi-square test and Fischer's exact tests. The prevalence ratio of categorical variables was calculated by using the generalized linear models' Poisson regression model with robust estimator (Paper 1). Binary logistic regression with robust estimator was used to calculate odds ratios (Paper 4). The adjusting variables were age, smoking habit, years worked in dusty factories other than coffee and presence of previous respiratory illness.

The distribution of measured values for total dust, endotoxin and FE_{NO} were skewed, and therefore were log transformed before analysis. Continuous variables were compared between the groups by independent t-test (Paper 1 – 4). Correlations between continuous variables were calculated by Pearson correlation tests.

Multiple linear regression models were used to assess adjusted associations between FE_{NO} levels (Paper 2 & 4), lung function parameters (FEV_1, FVC and FEV_1/FVC) (Paper 4) and independent variables. Independent variables in these models were; age, height (Paper 2 & 4), work in Arabica coffee factory versus Robusta and cumulative exposure to total dust or endotoxin (Paper 4). Variables with p value of less than 0.2 in any of the models for lung function parameters were retained in all three models for lung function. Similarly, in the FE_{NO} model, only the variables with a p-value <0.2 were retained in the final model.

To account for repeated samples from individual workers and factories, linear mixed models were used to estimate the determinants of exposure and their effect on exposure levels. The individual worker and factory were entered as random effects, while tasks, type of coffee and factory-related factors such as production rate, vigorous pouring of coffee, and having more than one machine in one room, were entered as fixed factors. Factors that had a p-value of more than 0.2 were removed from the models. Separate models were developed for total dust and endotoxin (Paper 3). The significant level was set to <0.05.

3.8 Ethical clearence

The first study was granted ethical clearance by the Regional Committee for Medical and Health Research Ethics West Norway and the National Institute for Medical Research in Tanzania. The second and third studies were granted ethical clearance by the Muhimbili University of Health and Allied Sciences Research Ethics Committee and the Regional Committee for Medical and Health Research Ethics, West Norway.

In all studies, permission to conduct the study was also obtained from the factories management where the objectives of the study were also explained in detail. The objectives of the study were explained to each worker. Those who agreed to participate were requested to give a written consent.

4. Summary of Results

4.1 Paper 1

Total dust exposure levels in the two Arabica coffee factories (A and B) were higher than in the control factory (GM; 1.23 mg/m^3 vs. 0.21 mg/m^3). The variability in total dust exposure was higher in factory B than A but the mean levels were not significantly different (GM 1.54 vs. 0.99 mg/m^3, respectively). Sweeping was the task with highest exposure to dust compared to other tasks (GM 8.2, median 7.55 mg/m^3). The use of respiratory protective equipment (RPE) was low among the coffee workers (33%), despite the dust produced during the processes. The RPE used were disposable masks that were not classified according to the quality of protection. In factory B 30% of those who did not have a mask, used a piece of cloth as their RPE.

Coffee workers had a higher prevalence of morning cough with sputum (23%) and chest tightness (27%) compared to controls (10% and 13%, respectively) with adjusted prevalence ratios (PR; 2.5; 95% CI; 1.0 – 5.5 and 2.4; 1.1 – 5.9, respectively).

4.2 Paper 2

This study was conducted in one Arabica coffee factory to assess fractional exhaled nitric oxide (FE_{NO}) levels of the workers, and to measure both personal total dust and endotoxin exposure levels.

Total dust and endotoxin levels were high among coffee workers (AM; 4.2 mg/m^3 and 6807 EU/m^3, respectively). Controls were working in a hospital and in one hotel nearby, hence dust and endotoxin exposures were not assessed from their workplaces. Total dust and endotoxin levels were significantly higher when hulling dry pre-processed Arabica than wet pre-processed Arabica (AM; 5.5 vs. 3.6 mg/m^3 and 14,253 vs. 1,990 EU/m^3, respectively). FE_{NO} mean levels were higher among coffee workers than controls (24 ppb vs. 14 ppb). Age adjusted levels of log transformed FE_{NO} were higher among male coffee workers compared to male controls (linear regression coefficient of 0.79 (95% CI 0.21 -1.36)), indicating 2.2 times higher FE_{NO} levels among coffee workers compared to controls.

4.3 Paper 3

This paper describes total dust and endotoxin exposure levels in Arabica and Robusta primary coffee factories and based on findings from Paper 2, it describes the determinants of exposure. Total dust and endotoxin levels were higher in Robusta factories (geometric mean (GM); 3.42 mg/m^3 and 10800 EU/m^3) than in Arabica factories (GM; 2.10 mg/m^3 and 1380 EU/m^3). Total dust levels correlated significantly with endotoxin levels with a correlation coefficient of 0.62. Sweeping, work at the hopper, the huller and the gravity table were the four tasks with highest dust exposure.

In a linear mixed model handling dry pre-processed coffee increased endotoxin exposure by a factor of 7 compared to handling wet pre-processed coffee, but had relatively lower effect on total dust levels (2.5 times higher exposure). Different tasks emerged as determinant for total dust and endotoxin exposure. Work at the hopper was a significant determinant for increased levels of both total dust (1.7 times higher) and endotoxin (2.8 times higher). Work at the huller increased endotoxin levels by a factor of 2.4, while work at the gravity table and sweeping increased total dust exposure by a factor of 1.5 and 3.5, respectively. Tasks and pre-processing method explained 30% and 71% of the total variability in dust and endotoxin exposure, respectively. Factory related factors did not remain in the model.

4.4 Paper 4

In the previous papers (Papers 1-3) there was a question still not answered: Is there a difference between Robusta and Arabica coffee workers in prevalence of respiratory symptoms, airway inflammation and their vital capacity? This paper reports the prevalence of chronic respiratory symptoms among Robusta and Arabica coffee workers. Coffee workers had higher prevalence of cough with sputum, chest tightness, wheezing, and asthma symptoms than the controls; the prevalence being highest among Robusta coffee workers. The Robusta coffee workers reported higher prevalence of at least one asthma attack in the past twelve months compared to Arabica coffee workers (38%vs. 18%, respectively), adjusted for age, height, years worked in dusty factories other than coffee and presence of previous respiratory illness (OR_{adj} 3.5; 95% CI; 1.4 – 9.0).

The age and height adjusted lung function parameters were not different comparing coffee workers and controls or when comparing Robusta and Arabica coffee workers. However, in the multiple linear models adjusted for age, height, years worked in dusty factories other than coffee and presence of previous respiratory illness, there was a significant decrease in FEV_1/FVC ratio related to cumulative dust and a significant decrease in FEV_1 and FEV_1/FVC with increased cumulative endotoxin. There was no difference in FE_{NO} levels between the groups.

5. Discussion

5.1 Main discussion

This study reports high personal total dust and endotoxin exposure among coffee workers with 17% of the collected samples exceeding the OEL for organic dust (5 mg/m^3) (30). Ninety nine per cent of the samples analysed for endotoxin (n=154) exceeded the health-based recommended exposure value of 90 EU/m^3 (32). The study reports higher prevalence for chronic respiratory symptoms among coffee workers than the controls (Papers 1 and 4); with the highest prevalence among those handling dry pre-processed Robusta coffee (Paper 4). Among the coffee workers, FEV_1 decreased significantly with increase in cumulative endotoxin exposure, while FEV_1/FVC ratio showed a small but significant decrease when both cumulative dust and endotoxin exposure increased (Paper 4). The level of FE_{NO} was higher among Arabica coffee workers in a high production year, however, these results were not observed in the subsequent year (Paper 4) when the production was lower (Fig. 14).

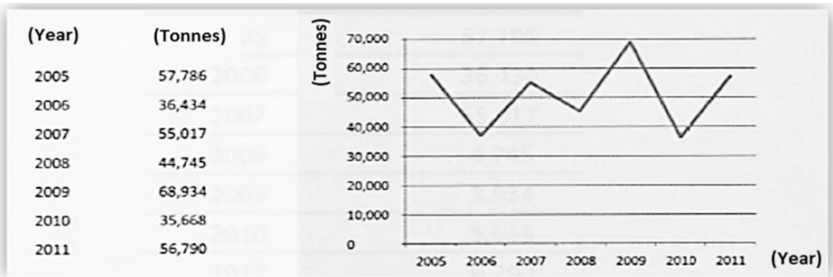

Figure 14: Seasonal variation of coffee production from 2005-2011(Source: TCB, "*Miaka 50 ya Uhuru na mapinduzi ya kilimo cha kahawa*").

5.1.1 Dust exposure

The present study has shown high personal total dust exposure in the coffee factories (GM 2.5, range 0.24 – 36 mg/m^3). Total dust levels were not different between factories processing similar type of coffee however; the levels were significantly higher in Robusta factories compared to Arabica factories (GM 3.42 vs. 2.10 mg/m^3).

The total dust exposure level in the primary coffee factories in the present study are lower than those found in secondary coffee factories in Croatia in 1979 (AM=11.2

mg/m^3) (41) and Sweden (5.9 – 6.3mg/m^3) in the green coffee areas as well as in primary and secondary factories in Uganda (Range:10 mg/m^3 – 58 mg/m^3) (39) . The difference could be due to different technologies over the years (three decades ago), processing stage in secondary factories and the amount of coffee being processed in the Ugandan factories. In the Croatian factories unloading bags was manual similar to our study but the mean level for this task was not given. At the time of the Ugandan study, Uganda was the biggest exporter of Robusta coffee in Africa, and this could have been dry pre-processed (70) which could also explain the difference in exposure levels. The total dust levels found in the present study are similar to the those measured in a green coffee warehouse in the UK (10) when unloading green coffee bags from lorries and tipping the GCB in a cleaner (3.2 mg/m^3 vs. 3.6mg/m^3).

In the present study, several tasks such as work at the hopper, sweeping, and sorting coffee at the gravity table were associated with higher exposures than the others. Packing was not associated with increased exposure levels contrary to findings reported from primary coffee factories in Papua New Guinea (40). Though tasks associated with higher exposure to dust are different in the Papua New Guinea study (40), the sorting task was associated with higher exposure similar to the present study.

5.1.2 Endotoxin exposure

Endotoxin levels were very high in the coffee factories (GM = 3500 EU/m^3; range 42-75,083 EU/m^3). Ninety nine per cent of the samples exceeded the recommended health based occupational exposure value of (90 EU/m^3) (32). There was high variability in endotoxin exposure levels in our study. No studies have previously described endotoxin exposure in primary coffee factories. However, higher levels of endotoxin have been reported in other agricultural industries. A high variability in endotoxin levels has been also been found in agriculture industries such as potato processing (GM; 279; range 0.5 – 62,227 EU/m^3 and 232; 10 – 29,080 EU/m^3) (44), agricultural seed processing (1800; 10 – 274,000 EU/m^3) (71) and in grain processing factories (360; 95 – 149,060) (72). The mean endotoxin level from the present study was higher than those presented from the other agricultural industries. Even though, one should be careful in comparing endotoxin results analysed in different laboratories due to differing analysis procedures (73).

Tasks associated with higher dust levels were similarly associated with higher endotoxin levels except for sweeping. Other studies on organic dust exposure in hemp processing plant and in horse stables reported higher dust and endotoxin when sweeping or cleaning (74;75). This difference with the present study might be due to different types of workplaces and different cleaning methods. Furthermore, there were few samples analysed for endotoxin from sweeping tasks in the present study as sweeping was done only when needed and in specific areas.

5.1.3 Determinants of exposure

The present study reported certain tasks in coffee factories as determinants of exposure for total dust and endotoxin. Nevertheless, these were not similar for both total dust and endotoxin exposures; work at the hopper was the only determinant for both exposures (Paper 3). Similarly the study in coffee factories in Papua New Guinea indicated that the tasks of, husking, sorting and packing were associated with 10 times increase in dust exposure level in the coffee factory. The effect of 10 is higher than that of 2.4 or 3.5 attributable to tasks in the present study (Paper 3). The difference could be that tasks were carried out differently, with different types of machines and working environment in the two studies.

Studies in other agricultural industries have also shown tasks to be determinants of exposure (43;45;46). In some of studies (43;46) the determinants for dust and endotoxins exposures were also not the same. In the present study factors like the machine with or without lid cover and vigorous activity did not explain the exposure levels in the coffee factories. This differs from the results reported from a potato processing factory where open rolling mills were associated with high endotoxin exposure compared to the closed ones (44).

The present study further showed that the type of pre-processing method at the farms had a significant effect on total dust and endotoxin exposure levels. Processing of dry pre-processed coffee was associated with a 2.5 times increase in dust exposure and

7.2 times increase in endotoxin. Processing either Robusta or Arabica coffee did not emerge as a determinant of exposure in the mixed models. These results indicate that the type of coffee had a lesser importance as a determinant of exposure compared to dry or wet pre-processed coffee raw material. There have been no other studies conducted to assess the determinants of exposure in coffee factories. However, results from other agricultural industries and farms have reported the type of raw material as a significant determinant for increase in exposure (43;46).

The determinants in our study explained much of the between-worker variability for both dust (21%) and endotoxin (68%) in the mixed effects models. On the contrary the models only explained 6% and 14% of the within-worker variance for dust and endotoxin. This is comparable to other studies in agricultural industries where none or very little within-worker variability was explained (43;47) meaning there is not much variability between day to day work. The determinants in this study, the tasks and processing dry pre-processed coffee best explained the total variance for both dust and endotoxin exposure.

5.1.4 Chronic respiratory symptoms among coffee workers

In our study coffee workers had a higher prevalence of cough day and night, chest tightness, wheezing and at least one attack of asthma symptoms in the past 12 months compared to the control group. The prevalence for some of the chronic respiratory symptoms found in our study was in line with those found in secondary coffee factories in Croatia (cough with phlegm 42%), the US (lower respiratory symptoms 37%) and in primary coffee factories in Uganda (chest tightness 22%)(39;41;51). However, lower prevalence of chronic cough (8.7% and 7.1%) has been reported from primary coffee factories in Papua New Guinea (40) and Uganda (39), from a secondary coffee factory in UK (wheeze, cough, dyspnoea 12.7%) (10) and among dock workers handling green coffee (76). This might be due to differences in processes, tasks, exposure levels as well as the group of workers selected for the assessment. For example the dock workers (76) were not being continuously exposed.

In the present study we have reported higher odds ratio for both chronic respiratory symptoms and asthma symptoms (Paper 4). Similar to our study higher odds ratio for cough with sputum, have been reported previously among primary and secondary coffee factory workers (39).

Few studies have reported asthma symptoms among male coffee workers in secondary coffee factories (42;49;56). Zuskin et al. 1985 reported that 6 of 9 coffee workers with respiratory complains had occupational asthma (58). A prevalence of asthma (9%) was also reported among female workers in secondary coffee factory handling both green and roasted coffee (57). Recently an allergen has been confirmed in Arabica coffee (52). This might mean that the symptoms of wheezing, chest tightness and asthma like symptoms reported among coffee workers in the present study may be related to allergic reactions. However, allergy was not examined in the present study.

Tuberculosis is not uncommon in Tanzania, with an incidence 312/100,000 population yearly (77). The country protocol is to investigate TB for patients presenting with a cough of two weeks or more (78). A study by Ngadaya et al. has shown that the prevalence of TB is similar in those with cough less than and more than two weeks (79). This could mean that coffee workers presenting with chronic cough with sputum production without smear positive test results might be misdiagnosed. This is due to limited or lack of connection between the occupational diseases and communicable diseases. The results from this study show that workers in coffee factories presenting with cough for more than two weeks might have occupational exposure related diseases.

5.1.5 Lung function changes and airway inflammation

This study has reported a decrease in lung function parameters (FEV_1 and FEV_1/FVC) among coffee workers in association with increased cumulative exposure to total dust and endotoxin. Most of the studies on lung function among coffee workers assessed the pre- and post-shift values. Even though we did not compare pre and post shift values, the results from this study are comparable to those in other studies in coffee factories which reported decrease in lung function after a work shift in Uganda and Croatia (39;41), and after a work week (35) among green coffee workers in Germany. In all these studies the decrease of the lung function parameter observed relates to increase in exposure and duration of exposure. Other studies found an association between exposures and decrease in other lung function parameter (MEF_{50}) among coffee workers (58). However, we did not analyse for other parameters. In the present study, we reported no difference in lung parameters when comparing coffee workers and controls. This was similar to the findings in secondary coffee factories in Italy (42) and in primary coffee factories in Papua New Guinea (40) which found no difference in lung function parameters between the coffee workers and the controls.

Despite the fact that the coffee workers in the present study had a higher prevalence for respiratory symptoms and asthma, there was no association between the prevalence of symptoms and lung function parameters. Lung function decreases have been reported to be higher among coffee workers with positive skin prick test (58). This was also indicated in the green coffee warehouse study in Italy (76). In a longitudinal study done by Sherman et al. 1992 (80) it was shown that people above the age of 40 who have respiratory symptoms have a longitudinal decrease in FEV_1. However the normal population is probably different from the coffee factory workers who had demanding tasks and a mean age less than 40 years.

Airway inflammation has not been assessed among coffee workers before. In this study, Arabica coffee workers who were exposed to high levels of personal total dust and endotoxin had increased concentration of the airway inflammatory marker, FE_{NO}, compared to the controls (Paper 2). These results were similar to studies done among farmers and agricultural workers (27) and among healthy subjects (60) exposed to endotoxins and indicated an association between exposure to endotoxin and levels of $FE_{NO.}$ However, this association was not observed in the subsequent season (Paper 4) when coffee production was lower, resulting to a lower dust and endotoxin exposure among the Arabica workers compared to the previous season.

Levels of FE_{NO} among coffee workers might be influenced with the amount of dust exposure as illustrated in our two studies (Papers 2 and 4). Increase in FE_{NO} level is also associated with cumulative exposure to endotoxin as illustrated among agricultural workers exposed to endotoxin (27). Smit et al. 2009 found that endotoxin exposure was associated with increased FE_{NO} among non-smoking and non-atopic workers (27). We did not assess atopy among the participants which could have influenced the results. Thomas et al. 1991 found that 54% of the workers in the warehouse and roasting plant were atopic (10). It has also been suggested that repeated exposure to organic dust might change the inflammatory response of the airways (60).

There are uncertainties that were not solved with this study, like the influence on exposure to beta-glucans. Both endotoxin and beta-glucans are found in organic dust (73). Fungal contamination has also been found in organic dust including dust in coffee factories (10) and it has been associated with allergic asthma. It is not possible to say that these workers were exposed to endotoxin only. In the present study, we did not assess the fungi or beta-glucans content in the endotoxin samples. Another uncertainty is the unknown effect on expression of FE_{NO} after intermittent exposure to endotoxin in coffee factories. It has been shown that cessation of exposure to endotoxin for 1 year lead to decrease in FE_{NO} from the mean of 27 ppb to 17 ppb among exposed workers (81). More research is needed to study whether the period of three months free of exposure in coffee factories lead to reduced inflammation and improved lung function among the workers.

5.2 Methodological discussion

5.2.1 Study design

This study was conducted using a cross section design for three consecutive years, involving different coffee factories. The dynamics of coffee production (Fig.14) causes a major change in the number of workers in each factory from season to season, and a longitudinal study design was not feasible for this group of workers. Only a few numbers of employees in the production line have fixed positions, and most of the workers vary in number and identity depending on the season yield. These situations cannot be controlled as they depend on nature and the weather. In our study, we found that about 20% of the workers had worked in factories processing similar type of coffee other than the current one.

One cannot neglect the fact that there may be a healthy worker effect in the coffee factories. The demanding nature of work in coffee factories will not allow a person who experiences adverse health effects to keep on working. Hence, those who are working are likely to be the healthiest ones. This may have weakened the association between the chronic respiratory symptoms and/or asthma symptoms and exposure. Furthermore, due to variability in production between seasons, the exposure levels vary for the workers who are employed in consecutive seasons as well as those who are employed intermittently. Studies on cessation of endotoxin exposure have shown that there is improvement of respiratory symptoms and lung function with time of cessation among some of the exposed workers (82;83). However the effect of intermittent exposure to coffee dust is not known and may have an impact on the outcomes of our study.

Selection of participants and sampling

We selected production line workers who are exposed to coffee dust. Participants were representative of the production workers in each factory in that they were either randomly selected or all were included. Participants were selected without knowledge about their health state. Work in coffee factories is physically demanding. Hence, the study might have included the very healthy and physically fit part of population. However the demanding nature of tasks in coffee factories was present in all factories, which could mean that it might be the case in any other primary coffee factory. The influence of having healthy workers was reduced by using controls groups from equally demanding tasks in the control factories (84).

Control groups

One of the strengths of this study is to have control groups for comparison of the outcomes. To avoid misclassification of exposed and unexposed the controls in this study had never worked in coffee factories. The groups were similar to coffee workers in socioeconomic status except for Paper 2 where health facility workers and hotel workers were used, some of whom had a higher socioeconomic status. However, this was not expected to interfere much with our outcome of interest, which was FE_{NO}. It was important to have controls from the same geographical area with presumed similar food habits.

5.2.2 Exposure and respiratory health assessments

Assessment of dust and endotoxin exposure

Closed face cassettes (CFC) were used for total dust sampling. CFCs are reported to under estimate inhalable dust by a factor of 1.5 - 4.2 (31) and samples less than inhalable sampler when particles are of >30 μm (85). Even though the CFC has some limitation, they are of low cost and can be easily used in harsh environments (85). Sampling was done throughout the working shift, which was at times slightly more than 8 hours. For this reason and due to high dust levels in some tasks, some samples were labelled overloaded (n=34). However, both the dust from the cassette walls and on the filters was measured in the laboratory. This situation may, however, underestimate the real amount of dust on these filters.

Sampling time varied in the data collected. Two samples collected for 116 min and 240 min were due to overloading of dust or interruption of the processing activities. In some cases, the activity was closed earlier for the day due to power failure and the workers had other tasks that did not involve coffee processing hence, the pump was stopped for that day. In one instance, a worker had to leave due to an emergency for private reasons, hence the pump was stopped. These factors could have an effect on the dust levels calculated per shift for the specific samples as we calculated the volume based on the sampling time.

Storage and transportation of endotoxin filters was a challenge. Endotoxin samples were supposed to be kept under cold temperature from the time of sampling to the day of analysis. Transporting the samples from different places in Tanzania to Norway and then to Sweden while maintaining the temperature was not easy but we managed to meet the quality demands by using rented freezers at a vaccination office and private freezers and transport the samples with freeze packs. A recent a review on measurement of endotoxin at workplaces (86) recommended that samples should be delivered to the laboratory at most 24 hours after sampling. Due to the unavailability of accredited laboratory in Tanzania the samples were delivered in the laboratory more than 24 hours after sampling, however, these were stored at lower temperature.

Extraction of endotoxins may differ in different laboratories and this could introduce bias in the results (87). However, all samples were extracted in the same laboratory except for the five samples that were taken as pilot samples. When the five samples were excluded the exposure levels and determinants remained similar.

The cumulative exposure was calculated using seasons for both Arabica and Robusta workers. This procedure could have led to bias in the cumulative exposure estimates. Coffee seasons are generally longer for Arabica coffee than Robusta coffee, we assumed they had one season regardless of the number of months in the season; the exact number of months for the specific seasons worked in the coffee factory was not available. Furthermore, the exposure level was assumed to be the same in the previous years as it was in the years of the study. The limitation with this assumption is that dust levels may vary with the production rate which will vary with different seasons as well as the duration of the season. Nevertheless, Figure 14 shows variation in production in two consecutive years hence having data from 2008 and 2009, 2008 and 2010 may have reduced the magnitude of underestimation or overestimation of cumulative exposure for the Arabica factories.

Respiratory health assessment

We assessed respiratory symptoms using a standardized questionnaire through an interview. Scholars have documented that there can be underreporting and over reporting when a questionnaire is used in both interview and self-administered questionnaire (88). Furthermore, English is not common among regular Tanzanians hence the questionnaire had to be translated to Kiswahili and back to English before it could be used.

Interviewing is a preferred method of assessment in Tanzania, especially if it involves people of blue collar jobs. This is due to the fact that some may not understand the questions and may not return the questionnaire. According to DHS 2010 (89) about 27% of male aged 20-54 have incomplete or no education in Tanzania. However, using this method may introduce interviewer bias (88). In this study, the bias was reduced by using a standardized questionnaire (64) and one researcher interviewed all the participants to maintain uniformity.

Lung function was assessed by a portable spirometer at any time of the day following ATS guidelines (69). Diurnal variation could have interfered with the results; nevertheless this was similarly assessed among the controls. Some of the studies used a vitalograph (10) or stationary spirometer (40;51), which would have not been feasible for this study setting. Use of portable spirometer is suitable for occupational studies where workers are examined onsite.

Other means of examination such as x-ray, blood samples to assess immunological variables such as neutrophils and eosinophils or performed induced sputum tests to assess the inflammation in the workers. These methods were not used as each one had its limitation with regards to the study site. It was not possible to take all the participants to a regional hospital for x-ray in the study areas. The limitation of induced sputum is that it need some medical attention in case of a severe attach and also some cases fail to produce the sputum (90).

External validity

This study was about primary coffee processing factories, not farm processing or coffee roasting factories. It included old and new factories, factories processing Robusta and those processing Arabica coffee. This makes the result applicable to other Tanzanian coffee factories. The results may also be applicable to factories of similar nature processing similar types of coffee in other countries. Occurrence of adverse respiratory health effects among workers in the study factories might be found in workers in other similar coffee factories.

6. Conclusion and recommendations

6.1 Conclusion

This study revealed that work in coffee factory may be associated with exposure to total dust and endotoxin higher than OEL and the health-based recommended value, respectively. Handling dry pre-processed coffee increases the exposure levels of total dust and endotoxin significantly. This exposure might be associated with impairment of the respiratory health that is decreased lung function and higher prevalence of chronic respiratory symptoms. In the present study the association between airway inflammation and exposure was inconclusive.

6.2 Recommendations

Due to high levels of dust and endotoxin and higher prevalence of respiratory and asthma symptoms control measures are of importance. Control of dust levels by focusing on the determinants of exposure will also reduce the level of endotoxin in the workplace atmosphere.

Short term

- Provision of at least efficient respiratory protection masks (RPE) for the workers. Representative sample of workers should participate in choosing and testing the masks so as to make ownership of the equipment. Personal fitting of RPEs to the workers have to be done before any RPE is distributed for use.

- Regular education on use and importance of RPEs to the workers before commencement of work, and on posters may increase the adherence.

- Changing practices in some activities such as sweeping by using damp cleaning method will reduce the regurgitation of settled dust. Practice of good housekeeping; regular cleaning and dusting off places in the factory including the storage areas.

- Regular health and housekeeping inspections by the government body (OSHA-TZ) to ensure good working environment and regular medical examination of the workers.

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Long term

- Installation of exhaust and hoods on gravity tables, pre-cleaners, hullers and hoppers
- Repair of dust removal systems; exhaust fans and broken hoods.
- Lowering hoppers that are above the pouring level.
- Cooperation between coffee authorities, TCB, coffee farmers unions, and research centre (TaCRI) to ensure proper handling of coffee at farms during drying and storage to reduce growth of microorganisms.

7. Future perspective

The present study and others have shown that there might be a relationship between dust exposure and health effects among production workers in coffee factories. However, there are still unanswered questions. Even though longitudinal studies might be difficult in these settings, a larger study including all coffee factories in an area could be conducted for three to four consecutive seasons.

Due to fluctuations in coffee production between seasons the association between exposure to dust and endotoxin in coffee factories and airway inflammation needs to be studied in more detail. It is important to learn which impact the seasonal coffee processing has on the workers respiratory health, i.e. whether respiratory health recovers between seasons or not. The association between other harmful contents in coffee dust and allergy is worth studying.

In the present study we could not say which type of inflammation the workers may have, and other methods such as a study on sputum cell count would provide more information on this topic. This may lead to possible medical attention to improve the health of the workers which is our first priority.

The exposure of organic dust from coffee is throughout the production chain; from the farms, storage warehouses and transportation. Hence widening the horizon by studying the effects among these groups will also aid in understanding the health problems and how best to protect them. More could probably be done at the farms to reduce the occurrence of endotoxin in the coffee cherries. Thus, further studies could be done to assess drying and storage conditions in order to detect factors that could contribute to lower endotoxin levels.

References

(1) Willson KC, Smith R, Clifford MN, Clarke RJ. Coffee botany, biochemistry and production of beans and beverage. Westport, Connecticut: The AVI Publishing Company; 1985.

(2) Clarence-Smith W, Topik S. Global coffee economy in Africa, Asia and Latin America 1500 -1989. Cambridge: Cambridge University Press; 2003.

(3) Tanzania Coffee Research Institute. Tanzania Coffee Baseline Report. Sokoine University of Agriculture, Morogoro, Tanzania: Bureau of Agriculture Consultancy and Advisory Service; 2005.

(4) Tanzania Coffee Board. Annual Report 2009/2010. TCB 2011 [cited 2013 Mar 14];1-26. Available from: URL: http://www.coffeeboard.or.tz/news_publications.php

(5) Silva CF, Schwan RF, Dias SE, Wheals AE. Microbial diversity during maturation and natural processing of coffee cherries of *Coffea Arabica* in Brazil. Int J Food Microbiol 2000;60(2-3):251-60.

(6) Uribe AC. Brown Gold: The amazing story of coffee. New York: Random House; 1954.

(7) BOT. Monthly economic review, December 2011. Bank of Tanzania 2011 [cited 2013 Mar 4];18-19. Available from: URL: http://www.bot-tz.org/Publications/publicationsAndStatistics.asp

(8) Baffes J. Tanzania's coffee sector: constraints and challenges in a global environment. World Bank Group African Region work paper series 2003;(42):1-52.

(9) Matua J. Post harvest and processing of coffee in African countries. FAO 2000 [cited 2013 Mar 14];X6936/EAvailable from: URL: http://www.fao.org/docrep/003/X6939E/X6939e00.htm

(10) Thomas KE, Trigg CJ, Baxter PJ, Topping M, Lacey J, Crook B, et al. Factors relating to the development of respiratory symptoms in coffee process workers. Br J Ind Med 1991;48(5):314-22.

(11) Osterman K, Zetterstrom O, Johansson SGO. Coffee Worker's allergy. Allergy 1982;37(5):313-22.

(12) Gardiner K, Harrington JM. Occupational hygiene. 3 ed. Malden, Massachusetts: Backwell Publishing; 2005.

(13) ILO. Encyclopeadia of occupational health and safety. 4th ed. Geneva: ILO; 1998.

(14) WHO. Hazard prevention and control in work environment: airborne dust. WHO 1999 [cited 2012 Dec 12];1-27. Available from: URL: http://www.who.int/occupational_health/publications/airdust/en/index.html

(15) Occupational Hygiene- Gases,Vapours,Dusts and Fibres. In: Aw TC, Gardiner K, Harrington JM, editors. Occupational Health. 5 ed. Oxford: Backwell; 2007. p. 117-39.

(16) Bakirci N, Kalaca S, Francis H, Fletcher AM, Pickering CA, Tumerdem N, et al. Natural history and risk factors of early respiratory responses to exposure to cotton dust in newly exposed workers. J Occup Environ Med 2007;49(8):853-61.

(17) Cherrie W, Howie R, Semple S. Monitoring for health hazards at work. 4 ed. Oxford, UK: Wiley-Backwell; 2010.

(18) Mark D. The sampling of Aerosols: principles and methods. In: Gardiner K, Harrington J.M, editors. Occupational hygiene. 3 ed. Backwell; 2005. p. 185-207.

(19) America Conference of Governmental Industrial Hygienists. Particles size-selective sampling criteia for airborne particulate matter. Threshold limit

values for chemical substances and physical agents and biological exposure indices.Cincinnati, OH: ACGIH; 2012. p. 76-9.

(20) Von Essen S, Robbins RA, Thompson AB, Rennard SI. Organic dust toxic syndrome: an acute febrile reaction to organic dust exposure distinct from hypersensivity pneumonitis. Clin Toxicol 1990 1;28(4):389-420.

(21) Smit LA, Zuurbier M, Doekes G, Wouters IM, Heederik D, Douwes J. Hay fever and asthma symptoms in conventional and organic farmers in The Netherlands. Occup Environ Med 2007;64(2):101-7.

(22) Liebers V, Raulf-Heimsoth M, Bruning T. Health effects due to endotoxin inhalation (review). Arch Toxicol 2008;82(4):203-10.

(23) Vogelzang P, van Gulden J, Folgering H, Kolk JJ, Heederik D, Preller L, et al. Endotoxin exposure as a major determinant of lung function decline in pig farmers. Am J Respir Crit Care Med 1998;157(1):15-8.

(24) Thorn J. The inflammatory response in humans after inhalation of bacterial endotoxin: a review. Inflam Res 2001;50(5):254-61.

(25) Wang XR, Zhang HX, Sun BX, Dai HL, Hang JQ, Eisen EA, et al. A 20-year follow-up study on chronic respiratory effects of exposure to cotton dust. Eur Respir J 2005;26(5):881-6.

(26) Thorne PS, Kulhankova K, Yin M, Cohn R, Arbes SJ, Zeldin DC. Endotoxin exposure is a risk factor for asthma: The national survey of endotoxin in United States housing. Am J Respir Crit Care Med 2005;172(11):1371-7.

(27) Smit LA, Heederik D, Doekes G, Wouters IM. Exhaled nitric oxide in endotoxin-exposed adults: effect modification by smoking and atopy. Occup Environ Med 2009;66(4):251-5.

(28) Sikkeland LIB, Eduard W, Stangeland AM, Thorgersen EB, Haug T, Aukrust P, et al. Occupational exposure to bacterial single cell protein induces inflammation in lung and blood. Inhal Toxicol 2009;21(8):674-81.

(29) Smit LA, Spaan S, Heederik D. Endotoxin exposure and symptoms in wastewater treatment workers. Am J Ind Med 2005;48(1):30-9.

(30) The Norwegian Labour Inspection Authority. Regulation on remedial action and limit values (*Forskrift om tiltaks- og grenseverdier*). 2013. Report No.: 704.

(31) Martin JR, Zalk DM. Comparison of total dust/inhalable dust sampling methods for the evaluation of airborne wood dust. Appl Occup Environ Hyg 1998;13(3):177-82.

(32) Health Council of the Netherlands. Endotoxins; Health based recommended occupational exposure limit. The Hague: Health Council of the Netherlands; 2010.

(33) Bucheli P, Kanchanomai C, Meyer I, Pittet A. Development of Ochratoxin A during Robusta (*Coffea canephora*) coffee cherry drying. J Agric Food Chem 2000;48(4):1358-62.

(34) Suarez-Quiroz M, Gonzalez-Rios O, Barel M, Guyot B, Schorr-Galindo S, Guiraud JP. Study of Ochratoxin A-producing strains in coffee processing. Int J Food Sci & Tech 2004;39(5):501-7.

(35) Oldenburg M, Bittner C, Baur X. Health risks due to coffee dust. Chest 2009;136(2):536-44.

(36) Sakwari G. Dust exposures and respiratory health among coffee processing workers in Kilimanjaro, Tanzania. Master Thesis, University of Bergen; 2010.

(37) Douwes J, Thorne P, Pearce N, Heederik D. Bioaerosol health effects and exposure assessment: Progress and prospects. Ann Occup Hyg 2003;(3):187-200.

(38) Rylander R. Endotoxin and occupational airway disease. Curr Opin Allergy Clin Immunol 2006;6(1):62-6.

(39) Sekimpi DK, Agaba EF, Okot-Nwang M, Ogaram DA. Occupational coffee dust allergies in Uganda. African Newletter on Occupational Health and Safety 1996;6:6-9.

(40) Smith D, Brott K, Koki G. Respiratory Impairment in Coffee Factory-Workers in the Asaro Valley of Papua-New-Guinea. Br J Ind Med 1985;42(7):495-8.

(41) Zuskin E, Valic F, Skuric Z. Respiratory-Function in Coffee Workers. Br J Ind Med 1979;36(2):117-22.

(42) Larese F, Fiorito A, Casasola F, Molinari S, Peresson M, Barbina P, et al. Sensitization to green coffee beans and work-related allergic symptoms in coffee workers. Am J Ind Med 1998;34(6):623-7.

(43) Spaan S, Schinkel J, Wouters IM, Preller L, Tielemans E, Nij ET, et al. Variability in endotoxin exposure levels and consequences for exposure assessment. Ann Occup Hyg 2008;52(5):303-16.

(44) Zock J-P, Heederik D, Kromhout H. Exposure to dust, endotoxin and micro-organisms in the potato processing industry. Ann Occup Hyg 1995;39(6):841-54.

(45) Preller L, Heederik D, Kromhout H, Boleij J, Tielen MJ. Determinants of dust and endotoxin exposure of pig farmers: development of a control strategy using imperical modelling. Ann Occup Hyg 1995;39(5):545-57.

(46) Halstensen AS, Nordby KC, Wouters IM, Eduard W. Determinants of microbial exposure in grain farming. Ann Occup Hyg 2007 Oct;51(7):581-92.

(47) Wouters IM, Spaan S, Douwes J, Doekes G, Heederik D. Overview of personal exposure to inhalable dust, endotoxin, beta(1-->3)-glucan and fungal extracellular polysaccharides in the waste management chain. Ann Occup Hyg 2006;50(1):39-53.

(48) Uragoda CG. Acute Symptoms in Coffee Workers. J Trop Med Hyg 1988;91(3):169-72.

(49) Figley KD, Rawling FFA. Castor bean - an industrial hazard as a contaminant of green coffee dust and used burlap bags. Allergy 1950;21(6):545-53.

(50) Zuskin E, Schachter EN, Kanceljak B, Witek TJ, Fein E. Organic Dust Disease of Airways. Int Arch Occup Environ Health 1993;65(2):135-40.

(51) Jones RN, Hughes JM, Lehrer SB, Butcher BT, Glindmeyer HW, Diem JE, et al. Lung function consequenses of exposures and hypersensitivity in coffee workers who process green coffee beans. Am Rev Respir Dis 1982;125:199-202.

(52) Manavski N, Peters U, Brettschneider R, Oldenburg M, Baur X, Bittner C. Cof a 1: Identification, expression and immunoreactivity of the first coffee allergen. Int Arch Allergy Immunol 2012;159(3):235-42.

(53) Lemiere C, Malo JL, McCants M, Lehrer S. Occupational asthma caused by roasted coffee: Immunologic evidence that roasted coffee contains the same antigens as green coffee, but at a lower concentration. J Allergy Clin Immunol 1996;98(2):464-6.

(54) Kaye M, Freedman SO. Allergy to raw coffee - an occupational disease. Can Med Assoc J 1961 Mar 4;84(9):469-71.

(55) Van Toorn D. Coffee worker's lung. A new example of extrinsic alveolitis. Thorax 1970;25(4):399-405.

(56) Karr RM, Lehrer SB, Butcher BT, Salvaggio JE. Coffee worker's asthma: A clinical appraisal using the radioallergosorbent test. J Allergy Clin Immunol 1978;62(3):143-8.

(57) Zuskin E, Valic F, Kanceljak B. Immunological and respiratory changes in coffee workers. Thorax 1981;36:9-13.

(58) Zuskin E, Kanceljak B, Skuric Z, Butkovic D. Bronchial Reactivity in Green Coffee Exposure. Br J Ind Med 1985;42(6):415-20.

(59) Quirce S, Lemiere C, De Blay F, Del Pozo V, Gerth Van Wijk R, Maestrelli P, et al. Noninvasive methods for assessment of airway inflammation in occupational settings. Allergy 2010;65(4):445-58.

(60) Sundblad M, Larsson BM, Palmberg L, Larsson K. Exhaled nitric oxide and bronchial responsiveness in healthy subjects exposed to organic dust. Eur Respir J 2002;20:246-1.

(61) Mwaiselage J, Bråtveit M, Moen B, Mashalla Y. Cement dust exposure and ventilatory function impairment: an exposure-response study. J Occup and Environ Med 2004;46(7):658-67.

(62) Douwes J, Versloot P, Hollander A, Heederik D, Doekes G. Influence of various dust sampling and extraction methods on the measurement of airborne endotoxin. App Environ Microbiol 199;61(5):1763-9.

(63) Lane SR, Nicholls PJ, Sewell RDE. The measurement and health impact of endotoxin contamination in organic dusts from multiple sources: Focus on the cotton industry. Inhal Toxicol 2004;16(4):217-29.

(64) America Thoracic Society- Division of Lung Diseases. Recommended respiratory disease questionnaires for use with adults and children in epidemiological research (ATS-DLD-78-A). American Thoracic Society 1978 [cited 2013 Mar 4];Available from: URL: http://www.thoracic.org/statements/

(65) de Marco R, Zanolin ME, Accordini S, Signorelli D, Marinoni A, Bugiani M, et al. A new questionnaire for the repeat of the first stage of the European Community Respiratory Health Survey: a pilot study. Eurn Respir J 1999;14(5):1044-8.

(66) American Thoracic Society. ATS/ERS Recommendations for standardized procedures for the online and offline measurement of exhaled lower respiratory nitric oxide and nasal nitric oxide, 2005. Am J Respir Crit Care Med 2005;171(8):912-30.

(67) Menzies D, Nair A, Lipworth BJ. Portable exhaled nitric oxide measurement comparison with the "Gold Standard" technique. CHEST 2007;131(2):410-4.

(68) 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 (FENO) for clinical applications. Am J Respir Crit Care Med 2011;184(5):602-15.

(69) Miller MR, Hankinson J, Brusasco V, Burgos F, Casaburi R, Coates A, et al. Standardisation of spirometry. Eur Respir J 2005;26(2):319-38.

(70) International coffee organisation. Trade statistics. ICO 2012 December [cited 2013 Feb 16];Available from: URL: http://www.ico.org/trade_statistics.asp

(71) Smit LA, Wouters IM, Hobo MM, Eduard W, Doekes G, Heederik D. Agricultural seed dust as a potential cause of organic dust toxic syndrome. J Occup Environ Med 2006;63(1):59-67.

(72) Spaan S, Wouters IM, Oosting I, Doekes G, Heederik D. Exposure to inhalable dust and endotoxins in agricultural industries. J Environ Monit 2006;8(1):63-72.

(73) Radon K. The two sides of the 'Endotoxin coin'. Occupational and Environmental Medicine 2006;63(1):73-8.

(74) Fishwick D, Allan LJ, Wright A, Curran AD. Assessment of exposure to organic dust in a hemp processing plant. Ann Occup Hyg 2001;45(7):577-83.

(75) Samadi S, Wouters IM, Houben R, Jamshidifard AR, Van Eerdenburg F, Heederik DJJ. Exposure to inhalable dust, endotoxins, beta(1 -> 3)-glucans, and airborne microorganisms in horse stables. Ann Occup Hyg 2009;53(6):595-603.

(76) Zotti R, Patussi V, Fiorito A, Larese F. Sensitization to green coffee bean (GCB) and castor bean (CB) allergens among dock workers. Int Arch Occup Environ Health 1988;61(1-2):7-12.

(77) Onozaki I, Ishikawa N, Enarson DA. Measuring the Global Burden of Tuberculosis. In: Preedy V, Watson R, editors. Handbook of Disease Burdens and Quality of Life Measures. Springer New York; 2010. p. 1189-210.

(78) Ministry of Health and Social Welfare. Manual of the National Tuberculosis and Leprosy programme in Tanzania. 5 ed. Dar es Salaam, United Republic of Tanzania: 2006.

(79) Ngadaya E, Mfinanga G, Wandwalo E, Morkve O. Detection of pulmonary tuberculosis among patients with cough attending outpatient departments in Dar Es Salaam, Tanzania: does duration of cough matter? BMC Health Serv Res 2009;9(1):1-5.

(80) Sherman CB, Xu X, Speizer FE, Ferris BG, Weiss ST, Dockery DW. Longitudinal Lung Function Decline in Subjects with Respiratory Symptoms. Am J Respir Crit Care Med 1992 Oct 1;146(4):855-9.

(81) Sikkeland LIB, Eduard W, Skogstad M, Alexis NE, Kongerud J. Recovery from workplace-induced airway inflammation 1 year after cessation of exposure. Occupational and Environmental Medicine 2012 Jul 25.

(82) Skogstad M, Sikkeland LIB, Øvstebø R, Haug KBF, Heldal KK, Skare Ø, et al. Long-term occupational outcomes of endotoxin exposure and the effect of

exposure cessation. Occupational and Environmental Medicine 2012 Feb 1;69(2):107-12.

(83) Wang XR, Zhang HX, Sun BX, Dai HL, Pan LD, Eisen EA, et al. Is Chronic Airway Obstruction from Cotton Dust Exposure Reversible? Epidemiol 2004;15(6):695-701.

(84) Li C-Y, Sung E-C. A review of the healthy worker effect in occupational epidemiology. Occupational Medicine 1999;49(4):225-9.

(85) Gorner P, Simon X, Wrobel R, Kauffer E, Witschger O. Laboratory Study of Selected Personal Inhalable Aerosol Samplers. Ann Occup Hyg;54(2):165-87.

(86) Duquenne P, Marchand G, Duchaine C. Measurement of Endotoxins in Bioaerosols at Workplace: A Critical Review of Literature and a Standardization Issue. Ann Occup Hyg 2012;57(2):137-72.

(87) Chun DTW, Bartlett K, Gordon T, Jacobs RR, Larsson BM, Larsson L, et al. History and results of the two inter-laboratory round robin endotoxin assay studies on cotton dust. Am J Ind Med 2006;49(4):301-6.

(88) Bowling A. Mode of questionnaire administration can have serious effects on data quality. J Publ Health;27(3):281-91.

(89) Tanzania Bureau of Statistics. Tanzania National Demographic Heath Survey 2010. TBS 2011 [cited 2013 Feb 18];Available from: URL: http://www.nbs.go.tz/index.php?option=com_content&view=article&id=106:tdhs&catid=53:tdhs&Itemid=123

(90) Purokivi M, Randell J, Hirvonen MR, Tukiainen H. Reproducibility of measurements of exhaled NO, and cell count and cytokine concentrations in induced sputum. Eur Respir J;16(2):242-6.