

Original Article

# Exposure to Wood Dust, Microbial Components, and Terpenes in the Norwegian Sawmill Industry

Anne Straumfors<sup>1,2\*</sup>, Raymond Olsen<sup>1</sup>, Hanne Line Daae<sup>1</sup>, Anani Afanou<sup>1</sup>, Dave McLean<sup>2</sup>, Marine Corbin<sup>2</sup>, Andrea 't Mannetje<sup>2</sup>, Bente Ulvestad<sup>1</sup>, Berit Bakke<sup>1</sup>, Helle Laier Johnsen<sup>1</sup>, Jeroen Douwes<sup>2</sup> and Wijnand Eduard<sup>1</sup>

<sup>1</sup>Department of Chemical and Biological Working Environment, National Institute of Occupational Health, PO Box 5530, Majorstuen, 0304 Oslo, Norway; <sup>2</sup>Centre for Public Health Research, Massey University - Wellington Campus, P.O. Box 756, Wellington 6140, New Zealand

\*Author to whom correspondence should be addressed. Tel: (+47)-23-19-53-38; e-mail: [anne.straumfors@stami.no](mailto:anne.straumfors@stami.no)

Submitted 5 March 2018; revised 23 April 2018; editorial decision 24 April 2018; revised version accepted 14 May 2018.

## Abstract

Sawmill workers are exposed to wood dust (a well-known carcinogen), microorganisms, endotoxins, resin acids (diterpenes), and vapours containing terpenes, which may cause skin irritation, allergy, and respiratory symptoms including asthma. The health effects of most of these exposures are poorly understood as most studies measure only wood dust. The present study assessed these exposures in the Norwegian sawmill industry, which processes predominantly spruce and pine. Personal exposures of wood dust, resin acids, endotoxin, fungal spores and fragments, mono-, and sesquiterpenes were measured in 10 departments in 11 saw and planer mills. The geometric mean (GM) and geometric standard deviation (GSD) thoracic exposures were: 0.09 mg m<sup>-3</sup> dust (GSD 2.6), 3.0 endotoxin units (EU) m<sup>-3</sup> (GSD 4.9), 0.4 × 10<sup>5</sup> fungal spores m<sup>-3</sup> (GSD 4.2), 2 × 10<sup>5</sup> fungal fragments m<sup>-3</sup> (GSD 3.2), and 1560 ng m<sup>-3</sup> of resin acids (GSD 5.5). The GM (GSD) inhalable exposures were: 0.72 mg m<sup>-3</sup> dust (2.6), 17 EU m<sup>-3</sup> (4.3), 0.4 × 10<sup>5</sup> fungal spores m<sup>-3</sup> (3.8), and 7508 ng m<sup>-3</sup> (4.4) of resin acids. The overall correlation between the thoracic and inhalable exposure was strong for resin acid ( $r_p = 0.84$ ), but moderate for all other components ( $r_p = 0.34$ – $0.64$ ). The GM (GSD) exposure to monoterpenes and sesquiterpenes were 1105 µg m<sup>-3</sup> (7.8) and 40 µg m<sup>-3</sup> (3.9), respectively. Although mean exposures were relatively low, the variance was large, with exposures regularly exceeding the recommended occupational exposure limits. The exposures to spores and endotoxins were relatively high in the dry timber departments, but exposures to microbial components and mono- and sesquiterpenes were generally highest in areas where green (undried) timber was handled. Dust and resin acid exposure were highest in the dry areas of the sawmills. Low to moderate correlation between components ( $r_p$  ranging from 0.02 to 0.65) suggests that investigations of exposure–response associations for these components (both individually and combined) are feasible in future epidemiological studies.

**Keywords:** endotoxin; fungal spores; fungal fragments; monoterpenes; resin acids; sesquiterpenes; wood dust

## Introduction

Sawmill workers are exposed to wood dust, microorganisms, resin acids (diterpenes), and endotoxins, as well as vapours containing terpenes. Exposure to dust from both hard and soft wood may cause nasal and sino-nasal cancers (IARC, 2012) and possibly lung cancer (Barcenas *et al.*, 2005; Jayaprakash *et al.*, 2008). Pine dust exposure in sawmill workers has also been associated with skin irritation, allergy, and respiratory symptoms including asthma symptoms and lung function decline (Douwes *et al.*, 2006).

Wood mainly consists of cellulose, hemicelluloses, and lignin, but also large amounts of terpenes. Monoterpenes are released from the heartwood during sawing and planning. The most common monoterpenes are  $\alpha$ -pinene,  $\beta$ -pinene,  $\Delta^3$ -carene,  $\beta$ -phellandrene, and limonene, but the composition is dependent on tree species (Fengel and Wegener, 2003). Exposure to monoterpenes has been associated with irritation of the eyes, mouth and throat, chest tightness, reduced lung function, increased bronchial hyperactivity, and airway inflammation (Hedenstierna *et al.*, 1983; Johard *et al.*, 1993; Dahlqvist and Ulfvarson, 1994; Eriksson *et al.*, 1996). Both spruce and pine secrete resin acid through resin channels in the wood, which are largest in pine; resin acids exposure may therefore be highest in pine processing. There are two broad categories of resin acids (i.e. abietic and pimaric acids), with abietic acid (AA) being the predominant type in pine and spruce. Abietic acid has been associated with allergic sensitization, respiratory symptoms, and asthma (Ayars *et al.*, 1989; Hessel *et al.*, 1995; Demers *et al.*, 1997).

Before sawing, wood logs are stored outdoors in a humid environment to prevent cracking. As a result, logs are often colonized by fungi (Dickinson and Levy, 1990; Strong *et al.*, 2005) and bacteria (Rossell *et al.*, 1973). After sawing, fungi may continue to grow on the timber if the water content is high, as is often the case when it is stored before kiln drying and during the first few hours in the drier. This may lead to exposure to fungi when handling the dried timber, particularly when sorting, and to a lesser extent during sawing and planning of dried timber. Exposure to fungal spores has previously been associated with respiratory symptoms and allergic alveolitis among sawmill workers (Wimander and Belin, 1980; Eduard *et al.*, 1992, 1993; Halpin *et al.*, 1994; Halpin *et al.*, 1994).

Bacteria may grow on the inside and outside of trees and may become airborne during handling and processing of the timber, which may also represent a health risk (Dutkiewicz *et al.*, 2001). Gram-negative bacteria are

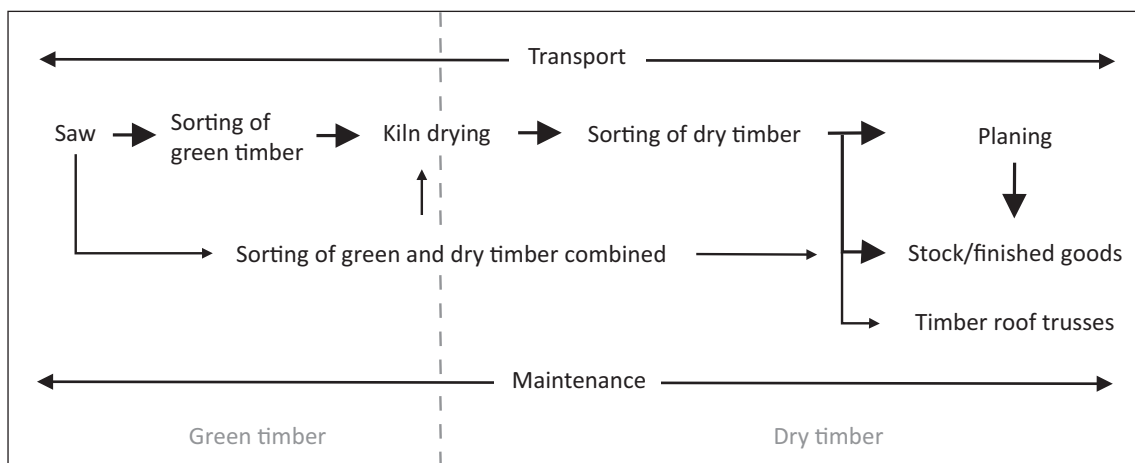
particularly relevant as they may contribute to exposure to endotoxins (Douwes *et al.*, 2000), which have potent pro-inflammatory properties and are associated with symptoms of the airways in a number of industries.

Despite the complex exposure situation involving wood dust and multiple wood-associated chemicals and organisms, with each exposure potentially able to contribute to adverse respiratory health, most studies continue to measure only wood dust. As a result, exposure levels of other relevant components and its impact on respiratory health remains largely unknown. The present study, which is part of a large longitudinal study on respiratory health of Norwegian sawmill workers, assessed exposure levels of dust, microorganisms, terpenes, and resin acids in 11 saw and planer mills, processing predominantly pine and spruce.

## Methods

### Sampling strategy

Eleven industrial sawmills, sorting, and planning companies in Norway, processing predominantly spruce (*Picea abies*) and/or pine (*Pinus sylvestris*) were included in the study. The selection was based on size, location, and wood type. In order to obtain a representative selection of sawmills, we recruited large- and medium-sized industrial sawmills from the two largest actors in the Norwegian wood industry and from independent sawmill companies. Small private sawmills connected to farms were not included. A schematic illustration of the process and the associated job groups are presented in Fig. 1. In total, 2305 full shift (duration 170–642 min, median 513 min) personal samples were collected involving 1–6 repeated measurements of 205 workers in 10 different departments (in some sawmills staff worked in both the saw and sorting of green timber departments; for these workers we created an addition category, see Table 2) in the period 2013–2016. Fourteen workers on each work shift carried small backpacks with sampling equipment for multiple measurements. Three sampling set types were used (Table 1). Sampling set type 1 collected samples for analysis of terpenes and thoracic dust, resin acids, endotoxin, and fungi. Sesquiterpenes were only collected in the saw and in the planning departments, where the concentrations *a priori* were thought to be highest. Sampling set types 2 and 3 collected inhalable and thoracic samples in parallel, primarily in the saw department, the sorting of dry timber department and the planning department, where the exposure to the main components were thought to be highest. A description of the work conducted in each department is presented in Table 2.



**Figure 1.** Process overview at saw-, sorting-, and planer mills. Arrows show the process direction, and the thickness of the arrows indicates which parts were most common. Maintenance and transport included work along the whole process line.

### Sampling

Thoracic samples were collected using BGI GK2.69 cyclones (BGI Inc., Waltham, MA, USA) mounted with Millipore 37 mm sampling cassettes (Merck Life Sciences, Darmstadt, Germany) at a flow rate of 1.6 l min<sup>-1</sup>. Inhalable samples were collected using 37 mm conical inhalable sampling cassettes (CIS; Casella Solutions, Kempston, UK) at a flow rate of 3.5 l min<sup>-1</sup>. Samples for dust and resin acid analysis were collected using polyvinylchloride filters (PVC, pore size 5 µm, Merck). Samples for endotoxin analysis were collected using glass fibre filters (GF/A, Merck), and samples for fungal analyses were collected using polycarbonate (PC) filters (Merck). PC filters with pore size 0.8 µm were used for the thoracic fraction, whilst for inhalable samples we used pore size 1.0 µm to reduce the resistance across the filter and maintain the required high flow throughout the work shift. Monoterpenes were collected using Anasorb CSC charcoal tubes (SKC Cat. no 226-01) (SKC Ltd, Dorset, UK) and sesquiterpenes were trapped on Tenax TA adsorbant tubes (Markes Int., Ltd., Llantrisant, RCT, UK), both at a flow rate of 50 mL min<sup>-1</sup>. After sampling, the tubes were sealed with rubber caps and swage lock caps, respectively, until analysis. The flow rates were recorded using a digital flow meter (Defender, SKC Inc., Eighty Four, PA, USA) before and after sampling. The average was used to calculate the volume sampled for inhalable samples and mono- and sesquiterpenes samples, whereas the designed flow rate of 1.6 l min<sup>-1</sup> for BGI GK2.69 cyclones was used for thoracic dust samples.

### Gravimetric measurement of aerosol mass

Gravimetric measurements were performed using a microbalance (Sartorius AG, MC 210p, Göttingen, Germany), and exposure levels expressed in mg m<sup>-3</sup>. Unexposed field blanks were included for every 10th sample. If a change in the mean weight of at least two field blanks was detected, we applied an equivalent correction to the field samples from the same series. The quality control procedure also included two certified weights and one set of in-house reference filters used for calibration prior to weighing each series. The gravimetric measurement was performed in a climate-controlled room with continuously monitored temperature (20 ± 1°C) and relative humidity (40 ± 2%). To ensure precise and comparable weighing conditions, all filters were equilibrated for a minimum of 2 days in the climate room prior to the weighing and mounting of the air sampling cassettes. Static electricity was discharged prior to weighing (Staticmaster®, NRD, LLC, NY, USA). The limit of detection (LOD), defined as three times the standard deviation of the blank filters, was 0.023 mg for thoracic dust and 0.011 mg for inhalable dust.

### Resin acid analysis

Resin acids were analysed using the same thoracic and inhalable dust samples that were collected to assess dust exposure. Resin acids were extracted using 2 ml methanol. Extracts were then filtered through a 0.20-µm nylon filter and transferred to 2 ml sample vials. Resin acids were subsequently separated by liquid chromatography and analysed using mass spectrometric detection

Table 1. Sampling overview.

Sample type	Exposure components	N	Sampling equipment	Analytical methods	Airflow
Sampling set type 1					
Thoracic particles	Dust/resin acids	389	BGI GK2.69/Millipore 37 mm/PVC 5µm	Gravimetry/LC-MS	1.6 l min <sup>-1</sup>
	Endotoxin	389	BGI GK2.69/Millipore 37 mm/GF/A	LAL	1.6 l min <sup>-1</sup>
	Fungal spores/fragments <sup>a</sup>	391	BGI GK2.69/Millipore 37 mm/PC 0.8 µm	FESEM	1.6 l min <sup>-1</sup>
Vapours	Monoterpenes	387	Anasorb CSC charcoal tubes	GC	50 ml min <sup>-1</sup>
	Sesquiterpenes <sup>b</sup>	58	Tenax TA adsorbent tubes	GC-MS	50 ml min <sup>-1</sup>
Sampling set type 2					
Thoracic particles	Dust/resin acids	113	BGI GK2.69/Millipore 37 mm/PVC 5 µm	Gravimetry/LC-MS	1.6 l min <sup>-1</sup>
Inhalable particles	Dust/resin acids	92	BGI GK2.69/Millipore 37 mm/GF/A	LAL	1.6 l min <sup>-1</sup>
	Endotoxin	112	CIS/PVC 5 µm	Gravimetry/LC-MS	3.5 l min <sup>-1</sup>
	Endotoxin	91	CIS/GF/A	LAL	3.5 l min <sup>-1</sup>
Sampling set type 3					
Thoracic particles	Fungal spores	85	BGI GK2.69/Millipore 37 mm/PC 0.8 µm	FESEM	1.6 l min <sup>-1</sup>
Inhalable particles	Fungal spores	85	CIS/PC 1.0 µm	FESEM	3.5 l min <sup>-1</sup>

FESEM, field mission scanning electron microscopy; GC-MS, gas chromatography-mass spectrometry; LC-MS, liquid chromatography-mass spectrometry; LAL, *Limulus* amoebocyte lysate assay.

<sup>a</sup>Fungal fragments were analyzed in two sawmills (*n* = 69).

<sup>b</sup>Sesquiterpenes were sampled in saw and planer departments only.



**Table 2.** Description of job groups.

Job group/department	Description
Saw	Debarked logs were entering the saw building, were measured and sawed into timber by bands saws, circular saw, and edge saw. Remote operation of the saws from enclosed control room, and trouble shooting and cleaning 'out' in the production areas
Sorting of green timber	Cut timber were transported on tracks and manually sorted based on quality by use of docking saw, and subsequently automatically sorted into piles by dimension. In some sawmills, the workers were operating the sorting remotely in an enclosed control room or in elevated, unprotected chairs with joysticks, but were out in the production area for trouble shooting and cleaning
Saw/sorting of green timber	For two sawmills, the job groups saw and sorting of green timber were combined due to work rotation between the departments
Kiln drying	Kiln drying of timber under controlled temperature and humidity. The work Included transportation of timber with forklift, operational control of the kiln dryer, fuelling of kiln, and cleaning of boiler
Sorting of dry timber	Dried timber was transported into the dry sorting building for manually sorting based on quality, labelling, and strapping. The work included cleaning of the production areas with compressed air or broom. Operators in this part of the sawmills were mostly placed in this dry end of the timberline regularly
Sorting of green and dry timber combined	Sorting of green and dry timber were done on the same production line, but not simultaneously. One sawmill had this solution. Both green and dry dust were present during work on this sawmill
Planing	Some of the sawmills had a planing department, where the dried timber were transported into and planed, profiled and resawed. The planer itself was enclosed and equipped with large dust extraction and ventilation systems, whereas the operator positions were not protected, sometimes equipped with local sprinkling to reduce dust levels and nuisance of the dry dust. Planed timber were strapped and wrapped
Stock/finished goods	Working in the stock/finished goods area included despatching, picking of material for customers, use of wrapping machine and forklift
Transport	Transport of logs and timber between departments, and wood chips, splinter and bark in the sawmill yard. Mainly outdoor work in forklift or truck with ventilated cabin
Maintenance	Maintenance workers were a heterogeneous group doing all kinds of repairs and maintenance work all over the sawmill and in the workshop during a day
Timber roof trusses	A separate department at one of the sawmills where roof trusses were constructed from dry timber. Work included transportation and cutting of timber, and assembly of roof trusses parts

(LC-MS) and atmospheric pressure chemical ionization (APCI) in negative mode. Five resin acids, i.e. 7-oxodehydroabietic acid (7-OXO), dehydroabietic acid (DHAA), levopimaric acid (LPA), AA, and isopimaric acid (IPA) were determined, and exposure levels were presented as ng m<sup>-3</sup> for each component individually, as well as the sum of all resin acids. However, as *P. abies* and *P. sylvestris* also contain the resin acids palustric acid (PALU) and neoabietic acid (NA) (Fengel and Wegener, 2003), which have identical masses as LPA, AA, and IPA, we determined the retention time of the resin acids PALU and NA by injecting analytical standards of PALU and NA, which showed that both co-eluted with AA. As the mass spectrometric signal intensities of PALU, NA, and AA were also almost identical, the resin acids PALU, NA, and AA were quantified as AA. The LODs were based on a signal-to-noise ratio of 3:1, and were 0.07 ng for 7-OXO, 0.67 ng for LPA, 0.65 ng for AA, 0.48 ng for IPA, and 0.26 ng for DHAA.

### Endotoxin analysis

Endotoxin was extracted using 5 ml endotoxin-free water with 0.05% Tween-20 and shaking at 500 rpm for 1 h, followed by centrifugation at 1000 G/rcf (Douwes et al., 1995). Supernatants were stored at -20°C until analysis. Extracts were diluted 10–50 times and analysed using the kinetic Limulus amoebocyte lysate (LAL) assay according to the manufacturers description (Lonza Ltd., Basel, Switzerland). Samples were analysed in quadruplicate consisting of two parallel analyses with and without 50 EU ml<sup>-1</sup> spike-in control. The concentrations in the extracts were determined by comparison with a five-point standard curve (0.005–50 EU ml<sup>-1</sup>). The LOD was 0.18–0.53 EU ml<sup>-1</sup> across analytical series. Exposure levels are presented as endotoxin units (EU) per m<sup>3</sup> (EU m<sup>-3</sup>).

### Fungal analyses

Fungal analyses were done by field emission scanning electron microscopy (FESEM). Due to different amounts

of dust on the filter, the thoracic, and inhalable PC filters were treated differently before analysis. A section of the thoracic filter samples was prepared for FESEM analysis without prior washing of the filter. Filters with inhalable dust were washed twice, first with 5 ml, and thereafter with 2 ml of phosphate-buffered saline containing 0.1% bovine serum albumin (BSA). A proportion (0.2, 0.5, or 0.7 ml) was filtered through a 25-mm polycarbonate filter (pore size 0.4  $\mu\text{m}$ ) which was prepared for FESEM analysis. Fungal fragments in the thoracic fraction of dust samples from two sawmills were immunogold labelled and analysed as previously described (Afanou *et al.*, 2018). Fungal spores were identified by their morphology. Spores and fragments were counted as described in Afanou *et al.* (2018). The lowest detectable number of particles was defined as 0.5 spores in 100 fields at 3000 $\times$  magnification, resulting in a LOD of 4018 spores for thoracic filters and 11 645 spores for inhalable filter.

### Monoterpene sampling and analyses

The capped charcoal sampling tubes were stored at  $-20^{\circ}\text{C}$  until analysis. Monoterpenes were desorbed from the charcoal tubes using 1.5 ml carbon disulphide ( $\text{CS}_2$ ) (Rathburn Cat.no. RG2027) at room temperature overnight and analysed using an Agilent 7890A gas chromatograph (GC) (Agilent Technologies, Santa Clara, CA, USA) with an Agilent HP-5 (25 m  $\times$  0.32 mm i.d., 1.05  $\mu\text{m}$  film thickness) capillary column and flame ionization detector (FID). The oven temperature was ramped as follows:  $35^{\circ}\text{C}$  for 8 min,  $6^{\circ}\text{C min}^{-1}$ ,  $240^{\circ}\text{C}$  in 1 min. 1  $\mu\text{l}$  was injected in splitless mode with helium as the carrier gas at a flow rate of  $1.5 \text{ mL min}^{-1}$ . The injector and detector temperature were both  $250^{\circ}\text{C}$ . Standard solutions of  $\alpha$ -pinene,  $\beta$ -pinene, 3-carene, p-cymene, and limonene were prepared in parallel with the samples. Dilution series of each standard were prepared containing a mixture of the monoterpenes at  $5 \times 10^{-4}$ ,  $1 \times 10^{-4}$ ,  $5 \times 10^{-5}$ , and  $1 \times 10^{-5} \text{ ml ml}^{-1}$  in  $\text{CS}_2$ . The desorption efficiency was corrected for by transferring 1.5 ml of the standard solution to a 2-ml sample glass containing coal from the main part of an unexposed charcoal tube. Data generated by GC/FID were collected and evaluated using the GC ChemStation software [B.03.01 (317); Agilent Technologies]. Monoterpenes present in the samples were identified by comparison with the retention time of the reference solutions and quantified against calibration curves with their, respectively, monoterpenes. No terpenes were found in field blanks, and adjustments for this were therefore not necessary. The LODs were based on a signal-to-noise ratio of 3:1, and varied from 0.07 to  $0.44 \mu\text{g ml}^{-1}$  across the different analytical series.

### Sesquiterpene analysis

Identification and quantification of the sesquiterpenes were carried out using a thermodesorption (TD) instrument (UNITY, series2) (TD, Markes Int., Ltd., Llantrisant, RCT, UK) connected to an Agilent 6890 gas chromatograph (GC) and an Agilent 5973 mass-selective detector (MS) (Agilent Technologies, Santa Clara, CA, USA). The sesquiterpenes were separated at a Varian, factor FOUR VF-5 ms capillary column (30 m, 0.32 mm i.d., 1.0  $\mu\text{m}$  film thickness). The GC was operated in constant pressure mode with an initial pressure to give a flow rate of  $1.8 \text{ mL min}^{-1}$  and helium as a carrier gas. The GC oven was initially set at  $40^{\circ}\text{C}$  for 7 min followed by an  $8^{\circ}\text{C min}^{-1}$  ramp to  $300^{\circ}\text{C}$  and finally held for 12 min. The MS was operated in the electron impact (EI, 70 eV) ionization mode. The temperature of the ion source and the quadrupole was 230 and  $150^{\circ}\text{C}$ , respectively. For identification of the sesquiterpenes, the MS was running in full scan mode (35–550 amu). As the total ion chromatogram (TIC) for sesquiterpenes are similar, and it is difficult to identify different sesquiterpenes from their TIC alone, the following compounds available were analyzed:  $\alpha$ -longipinen, (+)-longicyclen, isolongifolen, (-)- $\alpha$ -gurjunene, longifolene, trans- $\beta$ -farnesen, (-)-trans-caryophyllene, (+)- $\beta$ -cedrene, and  $\alpha$ -humulene (all from Merck). Several sesquiterpenes were detected, but only  $\alpha$ -longipinene could be identified by comparison with an available reference compound (mass spectra and retention time). Quantification of the sesquiterpenes was performed in SIM (single ion monitoring) mode. Three characteristic fragments of sesquiterpenes were monitored and used for identification and quantification:  $m/z$  133, 161, and 204.  $\alpha$ -longipinene exposure is expressed as  $\mu\text{g m}^{-3}$ . The rest of the detected, not identified sesquiterpenes (including  $\alpha$ -longipinene) are presented as  $\alpha$ -longipinene equivalents ( $\mu\text{g m}^{-3}$ ). Blank corrections were made using field and laboratory blanks. Agilent ChemStation software (E.02.02.) and the NIST 11 Mass Spectral Library together with retention times and reference compounds, was used for identifying and quantifying the sesquiterpenes.

### Statistical analyses

The exposure data were skewed and approximated a lognormal distribution. Therefore, we used ln-transformed values. Samples with values below LOD were replaced by the respective  $\text{LOD}/\sqrt{2}$  and adjusted for air volume (gravimetric and resin acid analysis) and dilution (endotoxin and monoterpene analysis). Samples with no observed spores were replaced by the LOD and adjusted for air volume. Samples with sesquiterpene values between zero and the lowest reference standard were calculated by using a best-fit cubic equation.

The exposure levels are expressed by geometric means (GMs) and geometric standard deviations (GSDs). Correlations between the different exposure components were assessed using Pearson correlation analyses, and a *P*-value below 0.05 was regarded as statistically significant. IBM SPSS statistics 25 was used for all statistical analyses (IBM, North Castle, NY, USA).

## Results

### Dust exposure

The mean thoracic dust exposure was 0.09 mg m<sup>-3</sup> and the mean inhalable dust exposure was 0.72 mg m<sup>-3</sup> (Table 3), with 10% of the inhalable samples and 0.6% of the thoracic samples exceeding the Norwegian occupational exposure limit (OEL) of 2 mg m<sup>-3</sup> for total softwood dust (Arbeidstilsynet, 2016). The thoracic dust exposure was highest for maintenance workers and workers sorting dry timber and lowest for transport workers and workers in the stock/finished goods department. The inhalable dust exposure was highest for workers involved in kiln drying and sorting of dry timber, and lowest for transport workers and workers in the stock/finished goods department. The correlation between inhalable dust and thoracic dust was moderate ( $r_p = 0.41$ ,  $P < 0.001$ ), and significant only in the planing department ( $r_p = 0.57$ ,  $P = 0.001$ ) when analysed by department.

### Microbial exposure

#### Endotoxin

Endotoxin exposure was relatively low for both thoracic and inhalable samples, but exposures varied considerably

as shown by the high GSDs (Table 4). The recommended health-based OEL for endotoxin (90 EU m<sup>-3</sup>, inhalable) (Health Council of the Netherlands, 2010) was exceeded in 4% of the thoracic samples and 11% of the inhalable samples. The thoracic exposure was highest in workers in the saw and sorting of green timber departments. Inhalable exposures were highest in the sorting of dry timber department, followed by the saw department. The correlation between thoracic and inhalable samples were moderate for all samples ( $r_p = 0.64$ ,  $P < 0.001$ ) and for the different departments (ranging from  $r_p = 0.39$ ,  $P < 0.05$  to  $r_p = 0.61$ ,  $P < 0.001$ ).

#### Fungal spores and fragments

Mean fungal spore exposures were all below the LOEL of 1 × 10<sup>5</sup> spores m<sup>-3</sup> (Eduard, 2009) (Table 4). However, large variance between measurements were observed, and 24% of both thoracic and inhalable measurements exceeded the LOEL. The mean thoracic exposure was highest in the department of saw/sorting of green timber, but was also elevated in the department of combined sorting of green and dry timber. The inhalable exposure was highest in the timber roof trusses department and the department of combined sorting of green and dry timber. However, there were only two measurements in both departments, with the highest level measured in the sorting of green timber department. Both the thoracic and inhalable exposure levels were also elevated in workers in the sorting of dry timber department, the sorting of dry and green timber combined, and the saw department. The maximum exposure was 5 × 10<sup>6</sup> thoracic spores m<sup>-3</sup> in the planing department and 1.7 × 10<sup>6</sup>

**Table 3.** Thoracic and inhalable dust exposure levels in the Norwegian sawmill industry.

Departments	Thoracic dust (mg m <sup>-3</sup> )			Inhalable dust (mg m <sup>-3</sup> )		
	N	<LOD (N)	GM(GSD)	N	<LOD (N)	GM(GSD)
All samples	501	53	0.09 (2.6)	112	0	0.72 (2.6)
Saw	65	1	0.08 (1.9)	29	0	0.67 (2.3)
Saw/sorting of green timber	46	6	0.06 (2.1)	17	0	0.74 (2.2)
Sorting of green timber	35	4	0.06 (1.9)	4	0	0.40 (2.4)
Kiln drying	25	4	0.09 (4.4)	2	0	8.49 (11.5) <sup>a</sup>
Sorting of dry timber	103	4	0.14 (2.0)	20	0	1.24 (2.4)
Sorting of green and dry timber combined	5	0	0.06 (1.5)	2	0	0.69 (0.2) <sup>a</sup>
Planing	86	3	0.09 (1.8)	29	0	0.68 (2.3)
Stock/finished goods	17	8	0.03 (3.2)	2	0	0.22 (0.2) <sup>a</sup>
Transport	50	20	0.04 (3.1)	2	0	0.31 (0.3) <sup>a</sup>
Maintenance	63	2	0.18 (2.8)	3	0	0.42 (1.8)
Timber roof trusses	6	1	0.06 (2.3)	2	0	0.74 (0.8) <sup>a</sup>

GM; geometric mean, GSD; geometric standard deviation; <LOD; below the limit of detection; N; number of samples.

<sup>a</sup>Arithmetic mean (standard deviation) due to only  $n = 2$ .

**Table 4.** Thoracic and inhalable microbial exposure in the Norwegian sawmill industry.

Departments	Endotoxin (EU m <sup>-3</sup> )			Fungal spores (×10 <sup>5</sup> m <sup>-3</sup> )			Fungal fragments (×10 <sup>5</sup> m <sup>-3</sup> )			
	N	<LOD (N)	GM (GSD)	N	<LOD (N)	GM (GSD)	N	Submicronic fragments <sup>b</sup>	Larger fragments	All fungal fragments
Thoracic samples										
All thoracic samples	481	124	3 (4.9)	476	77	0.4 (4.2)	69	0.2 (4.2)	1.7 (3.2)	2.0 (3.2)
Saw	62	4	7 (5.0)	57	7	0.5 (4.2)	16	0.1 (3.7)	0.8 (3.7)	0.9 (3.6)
Saw/sorting of green timber	36	4	4 (3.7)	39	1	1.1 (3.1)	17	0.3 (5.2)	2.5 (2.6)	2.9 (2.8)
Sorting of green timber	36	14	5 (8.2)	40	7	0.5 (3.8)	6	0.8 (5.9)	6.5 (2.1)	7.8 (2.2)
Kiln drying	25	10	1 (3.2)	5	1	0.4 (3.2)	10	0.1 (2.2)	1.2 (2.4)	1.4 (2.3)
Sorting of dry timber	94	21	3 (5.4)	99	9	0.6 (4.2)	18	0.1 (3.0)	1.9 (2.9)	2.0 (2.8)
Sorting of green and dry timber combined	5	3	1 (2.8)	5	0	0.8 (3.8)	0	-	-	-
Planning	86	23	2 (3.5)	82	7	0.5 (3.9)	2	0.2 (0.1) <sup>a</sup>	2.7 (0.8) <sup>a</sup>	2.9 (0.7) <sup>a</sup>
Stock/finished goods	18	7	1 (2.0)	17	7	0.1 (2.5)	0	-	-	-
Transport	50	26	1 (1.9)	48	21	0.1 (2.4)	0	-	-	-
Maintenance	63	10	3 (5.4)	63	12	0.3 (3.7)	0	-	-	-
Timber roof trusses	6	2	2 (10.2)	6	5	0.07 (2.1)	0	-	-	-
Inhalable samples										
All inhalable samples	16	16	17 (4.3)	86	12	0.4 (3.8)	-	-	-	-
Saw	27	0	35 (3.7)	21	2	0.4 (2.8)	-	-	-	-
Saw/sorting of green timber	7	0	28 (2.9)	10	1	0.5 (2.8)	-	-	-	-
Sorting of green timber	4	2	16 (3.9)	8	1	0.7 (5.3)	-	-	-	-
Kiln drying	2	0	1 (1.1) <sup>a</sup>	0	0	-	-	-	-	-
Sorting of dry timber	12	2	43 (2.1)	15	2	0.5 (3.9)	-	-	-	-
Sorting of green and dry timber combined	2	2	5 (2.6) <sup>a</sup>	2	0	3.6 (0.6) <sup>a</sup>	-	-	-	-
Planning	29	8	9 (3.1)	26	6	0.2 (3.3)	-	-	-	-
Stock/finished goods	1	0	47 (na)	0	-	-	-	-	-	-
Transport	2	0	4 (0.1)	0	-	-	-	-	-	-
Maintenance	3	1	5 (1.2)	2	0	0.8 (0.4) <sup>a</sup>	-	-	-	-
Timber roof trusses	2	1	4 (5.1) <sup>a</sup>	2	0	8.9 (12) <sup>a</sup>	-	-	-	-

GM, geometric mean, GSD, geometric standard deviation; &lt;LOD, below the limit of detection; N, number of samples.

<sup>a</sup>Arithmetic mean (standard deviation) due to only *n* = 2.<sup>b</sup>Twenty-eight samples were >LOD for submicronic fragments no samples <LOD for larger fragments.



inhalable spores  $\text{m}^{-3}$  in the timber roof trusses department. The levels of thoracic and inhalable spore exposures were weakly correlated when all samples were considered ( $r_p = 0.34$ ,  $P < 0.5$ ), but in the department for sorting dry timber, the correlation was stronger ( $r_p = 0.68$ ,  $P < 0.01$ ), whereas it was not significant (or not possible to analyze due to lack of measurements) in other departments.

Based on total particle numbers, there were more fungal fragments than spores in the thoracic samples. The exposure to both the submicronic and the larger fragments were highest in workers in the sorting of green timber department, followed by the saw/sorting of green timber and the planing department, although only two measurements were available for the latter. The correlation between the small and large fragments were strong in all samples ( $r_p = 0.71$ ,  $P < 0.001$ ), in the saw department ( $r_p = 0.81$ ,  $P < 0.001$ ), the saw/sorting of green timber department ( $r_p = 0.86$ ,  $P < 0.001$ ), and in the sorting of green timber department ( $r_p = 0.73$ ,  $P = 0.09$ ). Only moderate correlation was observed in the sorting of dry timber department ( $r_p = 0.56$ ,  $P = 0.01$ ), and weak in kiln drying ( $r_p = 0.37$ ,  $P = 0.3$ ).

Weak correlations between thoracic fungal spores exposure and fungal fragments exposure were observed ( $r_p = 0.34$ – $0.36$ ,  $P < 0.01$ ), except in the sorting of dry timber department, where the correlation was moderate between spores and SF, LF, or the sum of all fragments ( $r_p = 0.56$ – $0.61$ ,  $P < 0.01$ ).

### Resin acid exposure

The highest mean resin acid exposure was observed in workers in the sorting of dry timber department, the sorting of green and dry timber combined, and in the planing department, whereas the lowest exposures were observed in workers involved in transport and kiln drying (Table 5). Of all the different identified resin acids in the samples, the mean exposure to DHAA was the highest, in both the thoracic fraction and the inhalable fraction (Table 5). However, the highest single measurement was  $50 \mu\text{g m}^{-3}$  thoracic AA and  $88 \mu\text{g m}^{-3}$  inhalable AA in the dry sorting department. Two percent of the thoracic and 9% of the inhalable measurements exceeded the British long-term 8 h TWA OEL of  $50 \mu\text{g m}^{-3}$  rosin-based solder flux fume (HSE, 2011), mainly in the dry sorting department. The variance in exposure levels were considerable both within and between departments, but the relative abundance of the different resin acids was similar in both dust fractions. High correlations between the different individual resin acids were observed in both thoracic ( $r_p = 0.74$ – $0.97$ ,

$P < 0.001$ ) and inhalable ( $r_p = 0.73$ – $0.98$ ,  $P < 0.001$ ) samples, and the sum of all resin acids correlated strongly with all individual resin acids ( $r_p = 0.82$ – $0.99$ ,  $P = 0.001$  and  $r_p = 0.86$ – $0.99$ ,  $P < 0.001$  for thoracic and inhalable fractions, respectively). The resin acids in the thoracic fraction were highly correlated with the inhalable fraction ( $r_p = 0.84$ ,  $P < 0.001$ ). However, this was dependent on the department, and the correlation in the saw and in the saw/sorting of green timber department was only moderate ( $r_p = 0.42$ ,  $P < 0.05$  and  $r_p = 0.56$ ,  $P < 0.05$ , respectively).

### Terpene exposure

The exposure to terpenes was highest in the saw and sorting of green timber department, whereas the lowest exposure was observed in workers in the department for roof timber trusses, stock/finished goods, and transport (Table 6). Of the five monoterpenes that were identified,  $\alpha$ -pinene exposure was highest in all departments (Table 6). The relative pattern of terpene concentration was similar in all departments, except in the saw, sorting of green timber, stock/finished goods, and timber roof trusses departments, where the second highest monoterpene was  $\beta$ -pinene, and not 3-carene, as was found in the other departments (Table 6). Very high GSDs were observed for all terpenes, particularly, 3-carene. The number of monoterpene measurements below the detection limit were 4 for  $\alpha$ -pinene, 23 for  $\beta$ -pinene and 3-carene, 98 for p-cymene, and 36 for limonene. In general, nondetectable results were observed in departments where exposures were expected to be low, such as transport and stock/finished goods. However, p-cymene exposures were low throughout the saw- and planer mills. The correlations between all individual monoterpenes were high ( $r_p = 0.80$ – $0.96$ ,  $P < 0.001$ ), also across departments.

Sesquiterpenes exposures were highest in workers in the saw department, with exposures being nearly five times higher than in the planing department (Table 6).  $\alpha$ -longipinene was the only sesquiterpene that could be identified by a reference compound, but the mean  $\alpha$ -longipinene exposure represented only 4% of the mean exposure of all determined sesquiterpenes (and 5 and 3% of the mean total sesquiterpene exposure in the saw and planing department, respectively). The correlation between  $\alpha$ -longipinene and the sum of all sesquiterpenes was high ( $r_p = 0.97$ ,  $P < 0.001$ ). The sesquiterpene exposure was very low compared to the monoterpene exposure (Table 6). All single, as well as the sum of all terpenes, were below the present OEL for monoterpenes of  $140 \text{ mg m}^{-3}$  (Arbeidstilsynet, 2016).

**Table 5.** Thoracic and inhalable resin acid exposure levels in the Norwegian sawmill industry.

Departments	N	7-OXO (ng m <sup>-3</sup> )		DHAA (ng m <sup>-3</sup> )		LPA (ng m <sup>-3</sup> )		AA (ng m <sup>-3</sup> )		IPA (ng m <sup>-3</sup> )		All resin acids (ng m <sup>-3</sup> )	
		<LOD	GM (GSD)	<LOD	GM (GSD)	<LOD	GM (GSD)	<LOD	GM (GSD)	<LOD	GM (GSD)	<LOD	GM (GSD)
<b>Thoracic samples</b>													
All thoracic samples	502	7	76 (5.9)	0	698 (5.0)	2	101 (7.6)	12	282 (9.1)	8	217 (7.6)	0	1561 (5.5)
Saw	65	0	74 (2.8)	0	683 (2.8)	0	134 (3.2)	0	325 (3.1)	0	291 (83.6)	0	1589 (3.0)
Saw/sorting of green timber	46	0	94 (3.3)	0	648 (2.1)	0	79 (4.0)	1	232 (3.6)	1	191 (3.9)	0	1392 (2.3)
Sorting of green timber	35	0	83 (2.3)	0	443 (2.2)	0	80 (3.0)	0	218 (82.7)	0	157 (2.7)	0	1027 (2.3)
Kiln drying	25	0	23 (3.9)	0	176 (3.4)	1	15 (4.6)	1	33 (5.7)	1	37 (4.6)	0	358 (3.1)
Sorting of dry timber	103	1	35.3 (3.5)	0	3499 (4.3)	0	841 (5.5)	0	2622 (4.8)	1	1539 (84.9)	0	9260 (4.3)
Sorting of green and dry timber combined	5	2	168 (1.5)	0	2089 (1.5)	0	680 (1.9)	0	1937 (1.6)	0	1160 (1.6)	0	6097 (1.6)
<b>Inhalable samples</b>													
Planing	85	0	139 (2.1)	0	1093 (2.3)	0	163 (3.7)	0	594 (3.2)	0	376 (3.1)	0	2511 (2.6)
Stock/finished goods	18	1	8 (5.3)	0	166 (3.6)	0	8 (3.4)	4	11 (6.8)	3	9 (6.4)	0	252 (3.1)
Transport	50	2	6 (4.4)	0	81 (3.7)	0	7 (2.9)	6	11 (5.2)	2	12 (4.0)	0	147 (3.1)
Maintenance	64	1	71 (4.9)	0	677 (2.9)	1	76 (3.9)	0	284 (3.9)	0	227 (3.3)	0	1450 (3.1)
Timber roof trusses	6	0	2 (27.6)	0	31 (7.9)	0	5 (3.5)	0	10 (7.5)	0	9 (5.8)	0	61 (7.3)
<b>All inhalable samples</b>													
All inhalable samples	112	0	52.9 (3.6)	0	3127 (3.9)	0	535 (6.1)	0	1640 (5.4)	0	1277 (4.8)	0	7508 (4.4)
Saw	29	0	42.0 (3.1)	0	2245 (2.5)	0	333 (3.1)	0	952 (2.9)	0	896 (3.0)	0	5074 (2.7)
Saw/sorting of green timber	17	0	55.8 (2.3)	0	3116 (2.0)	0	610 (2.9)	0	1562 (2.4)	0	1433 (2.4)	0	7585 (2.2)
Sorting of green timber	4	0	28.9 (2.8)	0	1391 (1.8)	0	200 (3.2)	0	504 (2.3)	0	536 (2.6)	0	3192 (1.9)
Kiln drying	2	0	8.9 (107) <sup>a</sup>	0	413 (456) <sup>a</sup>	0	24 (28) <sup>a</sup>	0	102 (11.1) <sup>a</sup>	0	116 (126) <sup>a</sup>	0	745 (828) <sup>a</sup>
Sorting of dry timber	20	0	157.1 (2.8)	0	1287.5 (2.9)	0	354.5 (4.8)	0	969.6 (3.7)	0	600.2 (3.59)	0	3588.6 (3.2)
Sorting of green and dry timber combined	2	0	102.3 (474) <sup>a</sup>	0	971.2 (4443) <sup>a</sup>	0	303.0 (1337) <sup>a</sup>	0	989.3 (4867) <sup>a</sup>	0	566.1 (2313) <sup>a</sup>	0	2731.7 (13434) <sup>a</sup>
<b>All thoracic and inhalable samples</b>													
Planing	29	0	72.4 (2.1)	0	416.9 (2.5)	0	69.3 (3.6)	0	260.8 (3.2)	0	165.4 (3.1)	0	1018.7 (2.7)
Stock/finished goods	2	0	11.5 (134) <sup>a</sup>	0	60.5 (646) <sup>a</sup>	0	11.6 (156) <sup>a</sup>	0	33.5 (44.7) <sup>a</sup>	0	2.73 (350) <sup>a</sup>	0	1444 (1733) <sup>a</sup>
Transport	2	0	5 (25) <sup>a</sup>	0	190 (86) <sup>a</sup>	0	31 (25) <sup>a</sup>	0	107 (67) <sup>a</sup>	0	81 (50) <sup>a</sup>	0	446 (254) <sup>a</sup>
Maintenance	3	0	20.1 (3.0)	0	100.1 (2.9)	0	9.2 (2.5)	0	35.9 (3.6)	0	22.8 (3.8)	0	189.2 (3.1)
Timber roof trusses	2	0	2.5 (15) <sup>a</sup>	0	6.9 (16) <sup>a</sup>	0	5 (1.1) <sup>a</sup>	0	1.7 (2.0) <sup>a</sup>	0	2.1 (2.5) <sup>a</sup>	0	13.7 (31) <sup>a</sup>

GM, geometric mean; GSD, geometric standard deviation; <LOD, below the limit of detection; N, number of samples.  
<sup>a</sup>Arithmetic mean (standard deviation) due to only  $n = 2$ .

Table 6. Terpene exposure levels in the Norwegian sawmill industry.

Departments	N	$\alpha$ -Pinene ( $\mu\text{g m}^{-3}$ )		$\beta$ -Pinene ( $\mu\text{g m}^{-3}$ )		3-Carene ( $\mu\text{g m}^{-3}$ )		p-Cymene ( $\mu\text{g m}^{-3}$ )		Limonene ( $\mu\text{g m}^{-3}$ )		Sum all mono- terpenes ( $\mu\text{g m}^{-3}$ )		$\alpha$ -Longipinene ( $\mu\text{g m}^{-3}$ ) <sup>e</sup>		All sesqui- terpenes ( $\mu\text{g m}^{-3}$ ) <sup>e</sup>	
		GM (GSD)		GM (GSD)		GM (GSD)		GM (GSD)		GM (GSD)		GM (GSD)		GM (GSD)		GM (GSD)	
All samples	409	611 (8.2)	112 (7.0)	144 (10.1) <sup>a</sup>	21 (6.4)	66 (7.0)	1105 (7.8)	1.8 (3.4)	40 (3.9)								
Saw	38	4956 (5.0)	1188 (3.5)	335 (10.2)	114 (5.7)	607 (4.0)	7788 (5.0)	3.4 (2.7)	74 (2.9)								
Saw/sorting of green timber	38	1811 (11.2)	341 (7.0)	997 (17.1)	52 (10.4)	193 (8.0)	6554 (11.4)	-	-								
Sorting of green timber	32	3771 (4.0)	1072 (2.8)	569 (9.0)	140 (4.0)	585 (3.5)	6619 (4.1)	-	-								
Kiln drying	23	437 (2.9)	83 (2.9)	102 (3.6) <sup>b</sup>	14 (2.9)	45 (2.2)	852 (2.3)	-	-								
Sorting of dry timber	91	201 (3.5)	38 (2.8)	48 (5.7)	7 (3.0)	20 (3.0)	342 (3.4)	-	-								
Sorting of green and dry timber combined	3	846 (2.7)	42 (2.8)	285 (2.6)	21 (2.8)	46 (3.4)	1244 (2.7)	-	-								
Planing	57	1012 (6.7)	110 (4.3)	295 (9.1)	32 (4.8)	83 (4.9)	1614 (6.4)	0.6 (3.4)	16 (3.9)								
Stock/finished goods	15	31 (2.5)	9 (1.7)	8 (2.7)	3 (1.4)	8 (2.6)	69 (2.0)	-	-								
Transport	47	105 (3.3)	21 (2.7)	31 (4.0) <sup>c</sup>	5 (1.9)	11 (2.9)	212 (2.9)	-	-								
Maintenance	61	1230 (5.6)	201 (6.6)	262 (6.2) <sup>d</sup>	37 (4.3)	136 (5.0)	387 (5.4)	-	-								
Timber roof trusses	4	17 (3.4)	6 (1.4)	5 (1.7)	2 (1.1)	3 (1.2)	37 (1.9)	-	-								

GM, geometric mean; GSD, geometric standard deviation.

The number of monoterpene measurements below the detection limit were 4 for  $\alpha$ -pinene, 23 for  $\beta$ -pinene and 3-carene, 98 for p-cymene, and 36 for limonene. N, number of samples. <sup>a</sup> $n = 387$ ; <sup>b</sup> $n = 21$ ; <sup>c</sup> $n = 59$ ; <sup>d</sup> $n = 43$ ; <sup>e</sup> $n = 4$ .

### Correlations between exposure components

The sum of all fungal fragments, the sum of all resin acids, the sum of all monoterpenes, and the sum of all sesquiterpenes were used to assess the correlations between all exposure components. The exposure components correlated weakly to moderately within both the thoracic and inhalable fraction (Table 7).

### Discussion

To our knowledge, this article describes the most extensive exposure characterization ever conducted in the saw and planer mill industry. Five resin acids, five monoterpenes, several sesquiterpenes, endotoxin, dust loads, fungal spores, and fungal fragments were measured in personal samples from 10 different departments in 11 Norwegian companies, totalling nearly 2300 samples. The results showed that although mean exposure levels were relatively low for both the thoracic and inhalable fractions, the within and between department variance was

generally large. The recommended OELs of 2 mg m<sup>-3</sup> for wood dust (total dust), 90 EU m<sup>-3</sup> for endotoxin (inhalable), and 1 × 10<sup>5</sup> fungal spores m<sup>-3</sup> were exceeded in a proportion of all workers suggesting that at least some of these exposures may contribute to adverse health effects in this industry.

Inhalable dust and monoterpene exposures were comparable to previously reported exposure levels of 0.5–2.2 mg m<sup>-3</sup> dust and 0.1–138 mg m<sup>-3</sup> monoterpenes in sawmills processing spruce or pine (Eriksson *et al.*, 1996; Teschke *et al.*, 1999; Demers *et al.*, 2000; Douwes *et al.*, 2000; Liljelind *et al.*, 2001; Rosenberg *et al.*, 2002; Yamanaka *et al.*, 2009; Faerden *et al.*, 2014). Exposure for thoracic dust has previously been reported only in the US wood processing industry (Kalliny *et al.*, 2008), which showed higher exposures (0.29 mg m<sup>-3</sup>) in sawmills processing softwood than we found in our study (0.09 mg m<sup>-3</sup>).

A previous study showed that 9–21% of the total terpene emissions in sawdust from *P. abies* and *P. sylvestris* was sesquiterpenes (Granström, 2010).

**Table 7.** Pearson correlation between ln-transformed exposure components in the Norwegian sawmill industry.

Exposure component		Dust	Endotoxin	Fungal spores	Fungal fragments	Resin acids	Monoterpenes	Sesquiterpenes
Thoracic exposure								
Dust	$r_p$	1	0.22 <sup>a</sup>	0.22 <sup>a</sup>	0.02	0.54 <sup>a</sup>	0.12 <sup>b</sup>	0.04
	$N$	501	478	388	50	500	385	58
Endotoxin	$r_p$	0.22 <sup>a</sup>	1	0.39 <sup>a</sup>	0.50 <sup>a</sup>	0.13 <sup>a</sup>	0.15 <sup>a</sup>	0.39 <sup>a</sup>
	$N$	478	481	389	50	479	366	49
Fungal spores	$r_p$	0.22 <sup>a</sup>	0.39 <sup>a</sup>	1	0.36 <sup>a</sup>	0.26 <sup>a</sup>	0.13 <sup>b</sup>	0.14
	$N$	388	389	476	69	389	366	48
Fungal fragments	$r_p$	0.02	0.50 <sup>a</sup>	0.36 <sup>a</sup>	1	0.08	-0.09	0.43
	$N$	50	50	69	69	50	36	7
Resin acids	$r_p$	0.54 <sup>a</sup>	0.13 <sup>a</sup>	0.26 <sup>a</sup>	0.08	1	0.25 <sup>a</sup>	0.07
	$N$	500	479	389	50	502	384	58
Monoterpenes	$r_p$	0.12 <sup>b</sup>	0.15 <sup>a</sup>	0.13 <sup>b</sup>	-0.09	0.25 <sup>a</sup>	1	0.65 <sup>a</sup>
	$N$	385	366	366	36	384	387	56
Sesquiterpenes	$r_p$	0.04	0.39 <sup>a</sup>	0.14	0.43	0.07	0.65 <sup>a</sup>	1
	$N$	58	49	48	7	58	56	58
Inhalable exposure								
Dust	$r_p$	1	0.24 <sup>b</sup>	-	-	0.55 <sup>a</sup>	0.15	0.19
	$N$	112	91	-	-	112	19	9
Endotoxin	$r_p$	0.24 <sup>b</sup>	1	-	-	0.35 <sup>a</sup>	-	-
	$N$	91	91	-	-	91	0	0
Resin acids	$r_p$	0.55 <sup>a</sup>	0.35 <sup>a</sup>	-	-	1	-0.61 <sup>a</sup>	0.67
	$N$	112	91	-	-	112	19	9

-, not analysed, no sample to compare.

<sup>a</sup>Correlation is significant at the 0.01 level.

<sup>b</sup>Correlation is significant at the 0.05 level.

However, in the current study, sawmill workers were on average exposed to 30 times lower levels of sesquiterpenes than monoterpenes. It is unclear whether sesquiterpenes, at these levels, may contribute to adverse health effects, but as sesquiterpene exposure levels correlated only moderately with monoterpenes it is feasible to assess this in future epidemiological analyses.

The GM of  $7.5 \mu\text{g m}^{-3}$  inhalable resin acid exposure observed in our study was similar to the GM of  $8.0 \mu\text{g m}^{-3}$  reported by Teschke and colleagues, one of only a few studies in the sawmill industry that measured resin acid exposure (Teschke *et al.*, 1999). The GM of  $1.5 \mu\text{g m}^{-3}$  thoracic resin acid exposure was lower than what was measured by Teschke *et al.*, but the GM of both thoracic and inhalable exposure was higher in several departments, particularly the dry sorting and planing department.

Mean inhalable endotoxin exposures in our study were higher than previously observed in New Zealand sawmills (Douwes *et al.*, 2000), and we also found greater variance between workers as shown by greater GSDs. Thoracic endotoxin exposures were, as expected, lower than inhalable levels, but 4% of the samples nonetheless exceeded the recommended inhalable OEL of 90 EU  $\text{m}^{-3}$ .

The mean fungal spore (GM  $0.41 \times 10^5$  spores  $\text{m}^{-3}$ ) exposure was higher than previously reported in a 10-year follow-up study of exposures related to hypersensitivity pneumonitis by Færden *et al.* (2014). The maximum exposure of  $5 \times 10^6$  thoracic spores  $\text{m}^{-3}$  and  $1.7 \times 10^6$  inhalable spores  $\text{m}^{-3}$  was, however, lower than the maximum of  $1 \times 10^7$  observed in Norwegian sawmills in the eighties (Eduard *et al.*, 1992). The variation in the composition of fungal particles in thoracic air samples from two of the sawmills included in the present study has recently been described, and revealed that by number of particles, fragments dominated the total count (Afanou *et al.*, 2018). The GM of  $2 \times 10^5$  fungal fragments  $\text{m}^{-3}$  observed in the present study were higher than the arithmetic mean  $0.41 \times 10^5$   $\text{m}^{-3}$  of hyphal fragments observed in strawberry production (Tendal and Madsen, 2011), but lower than the  $30 \times 10^5$   $\text{m}^{-3}$  observed in grain farming (Halstensen *et al.*, 2007) and the  $86.9 \times 10^5$   $\text{m}^{-3}$  observed in seed handling where it was associated with organic dust toxic syndrome (Madsen *et al.*, 2012).

This study showed that although the GM exposure was relatively low for some of the analyzed components, the highest exposures for each individual component were observed in different departments, which suggest that for future epidemiological studies a broad exposure characterization is required including sample collection

in a wide range of departments. The microbial exposure was highest in workers working with green timber, where microorganisms growing on logs may be released into the air upon handling and cutting of the logs and green timber. High levels of thoracic fungal spores and inhalable endotoxins were also found in workers involved in sorting of dry timber. This is most likely due to microbial colonization of the timber before kiln drying, followed by aerosolization upon handling the dry timber during sorting. As expected, the exposure to mono- and sesquiterpenes were also highest in the green part of the sawmill (i.e. volatile terpenes are released before the timber is dried). Relatively high levels were also observed in the planing department, which may be explained by terpenes residing inside the wood that are released from freshly planed timber. The high exposure level of maintenance workers is probably due to maintenance work in areas of the sawmills where green timber is processed. Dust and resin acid exposure were both highest in the dry areas of the sawmills. Dust and resin acids are probably more easily released into the air in dry departments compared to green departments, due in part to the nature of fresh and dry dust, and the different processes in the different departments. For example, in the green departments, the dust is fresh and damp, and may be more likely to stick to the timber and therefore less likely to become airborne. Sawing will also generate large wood dust particles that quickly settle when aerosolized. Dry wood particles are more likely to become airborne due to less adhesion and higher electrostatic charge, and the sorting and planing processes are likely to generate more dust with smaller particle sizes. The high concentration of resin acids associated with sorting of dry timber cannot directly be explained by corresponding high dust loads, since the correlation between resin acids and dust were only moderate. However, it is possible that resin acids are resident mostly on small dust particles that may not fully represent gravimetric measurements.

The weak to moderate correlations between the exposure level in thoracic and inhalable samples of dust, endotoxin, and fungal spores suggest that future studies may benefit from separate exposure assessment for each fraction as each fractions may differentially affect respiratory health. The same may apply to resin acids, although the concentrations between the size fractions were highly correlated. The present paper showed that the sum of the individual components of resin acids, monoterpenes, sesquiterpenes, and fungal fragments were strongly correlated suggesting that they are representative for these groups of exposure, and could be used in epidemiological analyses instead of the

individual compounds. However, the individual composition and correlations may be different for different wood types and environments. The weak or moderate correlation found between the different groups of exposures strongly suggest that they may each be used in future analyses of exposure determinants and associations with respiratory health. Several of the components analyzed have not previously been investigated (either individually or in combination) in epidemiological exposure-response studies. It will therefore be of considerable interest to assess such associations in the future.

### Conclusions

Although mean exposures were relatively low for both the thoracic and inhalable fractions, the variance between measurements and departments was large, and levels regularly exceeded the relevant OELs for at least some of the exposures. Although the exposure to spores and endotoxin was relatively high in workers sorting dry timber, the exposures to microbial components and mono- and sesquiterpenes were in general highest in the green part of the sawmills, whereas dust and resin acid exposure were highest in the dry areas of the sawmills. As the highest concentration of each analyzed exposure component were observed in different departments, a comprehensive exposure characterization may be required for a valid risk assessment. Furthermore, low to moderate correlation between components suggest that assessments of exposure-response associations with individual components is feasible in future epidemiological studies.

### Funding and conflicts of interest declaration

Financial contribution to the study was received from the Research council of Norway, project no. 218232/H20. The authors declare no conflicts of interest.

### Acknowledgements

We thank all saw-, sorting-, and planning companies for participating in the study. Kristin Halgard, Ragnhild Martinsen Ånestad, Lene Madsø, Grete Friisk, and Ine Pedersen are acknowledged for excellent assistance in field and lab work.

### References

- Afanou AKJ, Eduard W, Johnsen HL *et al.* (2018) Fungal fragments and fungal aerosol composition in sawmills. *Ann Work Expo Health*; 62: 559–570.
- Arbeidstilsynet. (2016). AT 704. *Forskrift om tiltaks- og grenseverdier*. Trondheim, Norge: Direktoratet for arbeidstilsynet.
- Ayars GH, Altman LC, Frazier CE *et al.* (1989) The toxicity of constituents of cedar and pine woods to pulmonary epithelium. *J Allergy Clin Immunol*; 83: 610–8.
- Barcenas CH, Delclos GL, El-Zein R *et al.* (2005) Wood dust exposure and the association with lung cancer risk. *Am J Ind Med*; 47: 349–57.
- Dahlqvist M, Ulfvarson U. (1994) Acute effects on forced expiratory volume in one second and longitudinal change in pulmonary function among wood trimmers. *Am J Ind Med*; 25: 551–8.
- Demers PA, Teschke K, Davies HW *et al.* (2000) Exposure to dust, resin acids, and monoterpenes in softwood lumber mills. *AIHAJ*; 61: 521–8.
- Demers PA, Teschke K, Kennedy SM. (1997) What to do about softwood? A review of respiratory effects and recommendations regarding exposure limits. *Am J Ind Med*; 31: 385–98.
- Dickinson DJ, Levy JF. (1990) A review of methods used to study the microbial ecology of timber and forest products. *Method Microbiol*; 22: 479–96.
- Douwes J, McLean D, van der Maarl E *et al.* (2000) Worker exposures to airborne dust, endotoxin and beta(1,3)-glucan in two New Zealand sawmills. *Am J Ind Med*; 38: 426–30.
- Douwes J, McLean D, Slater T *et al.* (2006) Pine dust, atopy and lung function: a cross-sectional study in sawmill workers. *Eur Respir J*; 28: 791–8.
- Douwes J, Versloot P, Hollander A *et al.* (1995) Influence of various dust sampling and extraction methods on the measurement of airborne endotoxin. *Appl Environ Microbiol*; 61: 1763–9.
- Dutkiewicz J, Krysińska-Traczyk E, Prazmo Z *et al.* (2001) Exposure to airborne microorganisms in polish sawmills. *Ann Agric Environ Med*; 8: 71–80.
- Eduard W. (2009) Fungal spores: a critical review of the toxicological and epidemiological evidence as a basis for occupational exposure limit setting. *Crit Rev Toxicol*; 39: 799–864.
- Eduard W, Sandven P, Levy F. (1992) Relationships between exposure to spores from *rhizopus microsporus* and *paecilomyces variotii* and serum IgG antibodies in wood trimmers. *Int Arch Allergy Immunol*; 97: 274–82.
- Eduard W, Sandven P, Levy F. (1993) Serum ige antibodies to mold spores in 2 Norwegian sawmill populations - relationship to respiratory and other work-related symptoms. *Am J Ind Med*; 24: 207–22.
- Eriksson KA, Stjernberg NL, Levin JO *et al.* (1996) Terpene exposure and respiratory effects among sawmill workers. *Scand J Work Environ Health*; 22: 182–90.
- Færden K, Lund MB, Mogens Aaløkken T *et al.* (2014) Hypersensitivity pneumonitis in a cluster of sawmill workers: a 10-year follow-up of exposure, symptoms, and lung function. *Int J Occup Environ Health*; 20: 167–73.
- Fengel D, Wegener G. (2003) *Wood: chemistry, ultrastructure, reactions*. Remagen, Germany: Verlag Kessel.
- Granström KM. (2010) Underestimation of terpene exposure in the Nordic wood industry. *J Occup Environ Hyg*; 7: 144–51.

- Halpin DM, Graneek BJ, Lacey J *et al.* (1994) Respiratory symptoms, immunological responses, and aeroallergen concentrations at a sawmill. *Occup Environ Med*; **51**: 165–72.
- Halpin DM, Graneek BJ, Turner-Warwick M *et al.* (1994) Extrinsic allergic alveolitis and asthma in a sawmill worker: case report and review of the literature. *Occup Environ Med*; **51**: 160–4.
- Halstensen AS, Nordby KC, Wouters IM *et al.* (2007) Determinants of microbial exposure in grain farming. *Ann Occup Hyg*; **51**: 581–92.
- Health Council of the Netherlands. (2010) *Endotoxins. Health-based recommended occupational exposure limit*. The Hague: Health Council of the Netherlands; publication no. 2010/04OSH. ISBN 978-90-5549-804-8.
- Hedenstierna G, Alexandersson R, Wimander K *et al.* (1983) Exposure to terpenes: effects on pulmonary function. *Int Arch Occup Environ Health*; **51**: 191–8.
- Hessel PA, Herbert FA, Melenka LS *et al.* (1995) Lung health in sawmill workers exposed to pine and spruce. *Chest*; **108**: 642–6.
- HSE. (2011). *EH40/2005 workplace exposure limits*. EH40 2nd ed. London: Health and Safety Executive.
- IARC. (2012). *Arsenic, metals, fibers, and dusts. Volume 100 C. A review of human carcinogens. IARC Monographs on the evaluation of the carcinogenic risk of chemicals to humans*. Lyon, France: International Agency for the Research on Cancer.
- Jayaprakash V, Natarajan KK, Moysich KB *et al.* (2008) Wood dust exposure and the risk of upper aero-digestive and respiratory cancers in males. *Occup Environ Med*; **65**: 647–54.
- Johard U, Larsson K, Löf A *et al.* (1993) Controlled short-time terpene exposure induces an increase of the macrophages and the mast cells in bronchoalveolar lavage fluid. *Am J Ind Med*; **23**: 793–9.
- Kalliny MI, Brisolara JA, Glindmeyer H *et al.* (2008) A survey of size-fractionated dust levels in the U.S. wood processing industry. *J Occup Environ Hyg*; **5**: 501–10.
- Liljelind IE, Rappaport SM, Levin JO *et al.* (2001) Comparison of self-assessment and expert assessment of occupational exposure to chemicals. *Scand J Work Environ Health*; **27**: 311–7.
- Madsen AM, Tendal K, Schlünssen V *et al.* (2012) Organic dust toxic syndrome at a grass seed plant caused by exposure to high concentrations of bioaerosols. *Ann Occup Hyg*; **56**: 776–88.
- Rosenberg C, Liukkonen T, Kallas-Tarpila T *et al.* (2002) Monoterpene and wood dust exposures: work-related symptoms among Finnish sawmill workers. *Am J Ind Med*; **41**: 38–53.
- Rossell SE, Abbot EGM, Levy JF. (1973) Bacteria and wood - review of literature relating to presence, action and interaction of bacteria in wood. *J Inst Wood Sci*; **28**–35.
- Strong N, Webber J, Eaton R. (2005) Factors affecting the fungal colonization of pine lumber. *For Pathol*; **35**: 195–203.
- Tendal K, Madsen AM. (2011) Exposure to airborne microorganisms, hyphal fragments, and pollen in a field of organically grown strawberries. *Aerobiologia*; **27**: 13–23.
- Teschke K, Demers PA, Davies HW *et al.* (1999) Determinants of exposure to inhalable particulate, wood dust, resin acids, and monoterpenes in a lumber mill environment. *Ann Occup Hyg*; **43**: 247–55.
- Wimander K, Belin L. (1980) Recognition of allergic alveolitis in the trimming department of a Swedish sawmill. *Eur J Respir Dis Suppl*; **107**: 163–7.
- Yamanaka MW, Guidotti TL, Koehncke N *et al.* (2009) Wood dust levels in Alberta sawmills. *Arch Environ Occup Health*; **64**: 270–7.