Airborne Microorganisms, Endotoxin, and Dust Concentration in Wood Factories in Italy

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Exposure to biological agents and dusts occurs in homes and occupational environments and it is known to cause adverse health effects. There is limited information concerning the occupational exposure levels of airborne biohazard during wood processing, but this exposure is associated with a range of adverse health effects. Control of exposure to microbiological hazards and dust in woodworking is not easy. In fact, various types of wood are commonly used and they generate complex mixtures of dusts and biological agents with various health risks. The aim of this study was to investigate the concentration of dust, bacteria, and endotoxins encountered in six different wood factories. These people were exposed to between 0.05 and 12.00 mg inhalable dust m$^{-3}$ and between 0.40 and 6.93 ng inhalable endotoxins m$^{-3}$. Total bacteria concentrations in the air of the factories examined were within a range of 130–2000 CFU m$^{-3}$, the value of Gram negative was within a 0–164 CFU m$^{-3}$, and the concentration of Gram positive was within 1–104 CFU m$^{-3}$. In conclusion, people working in wood factories may be exposed to high levels of inhalable dust and endotoxins.

Keywords: airborne microorganisms; bioaerosols; dust; endotoxin; wood factories

INTRODUCTION

Wood is among the most important natural resources in the world and annually at least 1700 million m$^3$ are collected for industrial use. Dust generated from wood processing is a heterogeneous mixture of inorganic and organic particles: wood fragments, viable and non-viable microorganisms, endotoxins, glucans, allergens, or mycotoxins, which all represent health hazards upon inhalation (Lacey and Crook, 1988; Bohn and BeMiller, 1995; Ulmer et al., 1997; Duchaine, et al., 2000; Rylander, 2002; Douwes, 2005; Young and Castranova, 2005).

In addition, wood dust is classified as a known human carcinogen on the basis of studies linking exposure and cancers of the nasal cavity and sinuses. In 2000–2003, ~3.6 million workers (2.0% of the employed population) were occupationally exposed to inhalable wood dust in 25 member states of the European Union (Kauppinen et al., 2006).

The International Agency for Research on Cancer (IARC) has classified wood dust as carcinogenic to humans on the basis of epidemiological evidence (IARC, 1995). The European Union Directive (1999/38) and more recently in Italy by legislative decree 66/2000 (Italy, 2000) has also classified hardwood dusts as carcinogenic and has set the occupational exposure limit (OEL) for hardwood dust to 5 mg of inhalable dust in cubic metre of workroom air. In Italian regulations, in the legislative decree 81/2008, attachment XLIII (Italy, 2008), an OEL of 5.0 mg m$^{-3}$ is applied for total hardwood dust (inhalable fraction over an 8-h exposure period). Whereas the American Conference of Governmental Industrial Hygienists (ACGIH, 2010) lists two different threshold limit values (TLV$^\text{®}$s): 0.5 mg m$^{-3}$
for the western red cedar (a sensitizing agent) and 1.0 mg m\(^{-3}\) for all other species (inhaleable fraction).

But the risks can be searched also in exposure to biological factors. Biological agents are represented by bacteria, moulds, endotoxins, and mycotoxins. These agents can spread in the work environment mixed with wood dust and be inhaled. Interest in bioaerosol exposure has increased over the last few decades. This is largely because it is now appropriately recognized that exposures to biological agents both in the occupational and in the residential indoor environment are associated with a wide range of adverse health effects with major public health impact, including contagious infectious diseases, acute toxic effects, and allergies (Douwes et al., 2003). In the last few years, it has emerged that in several industrial activities, exposures to biological agents can be abundant. Bacteria and fungi may abundantly develop in timber logs or on wood chips stored in piles in factory yards (Jääppinen et al., 1987; Dutkiewicz, 1989; Dutkiewicz et al., 1992). When dispersed into air during wood processing, they may be inhaled by workers and evoke airways inflammation either by specific allergic reactions or by non-specific immunostimulation (Rylander, 1987; Lacey and Dutkiewicz, 1994; Wintemeyer et al., 1997; Alwis et al., 1999; Mandryk et al., 1999, 2000). Moulds, abundantly developing on wood chip piles, are regarded as a main risk factor for chip handling workers (Jääppinen et al., 1987; Kolmodin-Hedman et al., 1987; Wintemeyer et al., 1997). Among bacteria, a potential hazard is posed by endotoxin-producing Gram-negative bacteria that may abundantly develop in the softwood of stored timber of pine, beech, and other trees (Dutkiewicz, 1989; Dutkiewicz et al., 1992).

It is difficult to assess the microbiological quality of indoor air because there are no international standards, even if there are reference limits in many states. In particular, endotoxin from Gram-negative bacteria has been recognized as an important factor in the aetiology of occupational lung diseases, including asthma (Douwes et al., 2003; Williams et al., 2005; Madsen et al., 2008) and organic dust toxic syndrome (Smit et al., 2005, 2006). Inhaled endotoxins and organic dust particles have been shown to have synergistic pro-inflammatory effects in organic dust-induced asthma (Pirie et al., 2003). Although the danger of endotoxin is recognized, there is no internationally accepted threshold OEL, only the DECOS proposed in 1998 a OEL of 50 endotoxin unit (EU) m\(^{-3}\) for inhalable endotoxins but this OEL has been increased to 200 EU m\(^{-3}\) in 2000, and definitively abolished in 2003 from the Netherlands regulation, in 2010 the DECOS proposed a OEL of 9.0 ng m\(^{-3}\) (DECOS, 2010).

However, many studies have suggested different values of TLV\(^{®}\) both for the inhalable fraction and for the total endotoxins between 3 and 80 ng m\(^{-3}\) (Haglind and Rylander, 1984; Rylander et al., 1985; Castellan et al., 1987; Kennedy et al., 1987; Smid et al., 1992, 1993; Donham and Cumro, 1999; Donham et al., 2000; Swan et al., 2003).

Many authors have previously evaluated the presence of microorganisms, endotoxin, and wood dust in carpentry workshops in different parts of the world. In all these studies, they used different methods of analysis, and there is great variability in both the concentration of microorganisms and the endotoxins. The difficulty of these measures is the lack of a common standardized method. For example, Oppliger et al. (2005) have monitored the presence of wood dust and bioaerosol in some sawmills in Switzerland. This research showed that there was a high bacterial and fungal contamination in all the sawmills analysed, while the amount of wood dust and endotoxin was low on average. Other authors have tried to evaluate the occupational exposure of workers in Polish sawmills considering various aspects, air quality, but also to determine the reactivity of sawmill workers to biological allergens associated with wood dust (Dutkiewicz et al., 2001a, c). Others have tried to optimize the method for sampling and analysis of endotoxin in sawmills (Duchaine et al., 2001).

Because it is a very complex study, the aim of this paper was, therefore, to characterize personal exposure to airborne dust, microorganisms, and endotoxins during wood processing in two sawmills and four carpentries in South Italy, in order to contribute to find common lines and comparable methods for the assessment of occupational exposure to bioaerosols and dust.

METHODS

Wood factories

The investigation was carried out on six wood factories located in South Italy: two sawmills and four carpentries. Measurements were performed for each factory in the different workstations and each time over two successive working days (Table 1). In all factories, wood was produced from different wood types but only hardwood.

In sawmills ‘A’ and ‘B’, air samples for determination of bacterial concentration, dust, and endotoxins were taken at the following two sites, marked A1–A2 and B1–B2: sawmill station (A1 and B1) and barker station (A2–B2). In factories ‘C’, ‘D’, ‘E’, and ‘F’, air samples were taken at the following four
sites, marked 1–4: sanding station (C1–D1–E1–F1), cut station (C2–D2–E2–F2), flanging station (C3–D3–E3–F3), and assemblage station (C4–D4–E4–F4). Temperature and relative humidity (RH) % have been found by mean of portable detector RS-1360 (RS Components Ltd. Corby, Northants, UK).

Microbiological examination of the air

Air samples for microbiological analysis were taken by an impactor Microflow (Aquaria, Milan, Italy). Each air sample was in duplicate, taken at a flow rate of 20 l min⁻¹, as described by Prazmo et al. (2003). The choice of using a stream so low is based on our previous studies, which indicated the best use of a low flow (data not shown). In addition, there are no indications about the choice of the flow even in the UNI EN 13098, which we used. Air volumes of 200 l were sampled for bacteria and 300 l for moulds. Total cultivable bacteria were impacted onto Tryptone soja agar plates (TSA) and Plate count agar (PCA), Gram-negative bacteria onto C-EC and MacConkey (McC), Gram-positive bacteria onto Mannitole Salt agar (MSA), and Bile esculin agar (BEA), fungi onto Bengan Red agar (RB), and Candida onto Candida agar (all from Biolife Italiana Srl, Milan, Italy).

The plates were subsequently incubated for 1 day at 37°C (TSA, MSA, C-EC, McC, and BEA), for 2 days at 30°C (PCA) and for 3 days at 22°C (Candida agar and RB). Grown colonies were counted, differentiated, and the data reported as CFU per 1 m³ of air (CFU m⁻³). Bacterial isolates were identified with microscopic and biochemical methods by API tests (Biomerieux, France), as recommended by Bergey’s Manuals (Krieg and Holt, 1984; Holt et al., 1994). Identification of mould genus was accomplished via microscopic and macroscopic examination.

**Personal sampling of inhalable aerosols**

For determination of dust and endotoxins concentration, air samples were sampled continuously for 4 h, on polyvinyl chloride filters (PVC) and IOM samplers, by use of an AirChek 2000 personal sampler (SKC Inc., Eighty Four, PA, USA), set at a flow rate of 2.0 l min⁻¹, as described by Harper and Andrew, (2006). Two samples were taken in each sampling site. PVC filters were chosen because of their high collection efficiency, low toxicity, and low pressure drop.

**Gravimetric determination of dust on PVC filters**

Inhalable dust was sampled on PVC filters and IOM sampler (SKC Inc.). Filters were weighed on a AND balance HM-202 (A&D Engineering, Inc., San Jose, CA, USA) (precision 0.01 mg) after conditioning for at least 24 h at constant temperature (20 ± 0.5°C) and RH (50 ± 1%). Each filter was weighed both before and after dust sampling, after the filter had been removed from the IOM sampler. For these measures, the official method NIOSH 0500 for dust was used (NIOSH, 1994).

**Extraction of dust for endotoxin analysis**

Dust on the PVC filters was extracted in 5.0 ml pyrogen-free water with 0.05% Tween 20 by orbital shaking (300 r.p.m.) at room temperature for 60 min and centrifuging (1000 g) for 15 min. The supernatant was stored at −80°C for 12–24 h to await endotoxin measurement. (CEN EN 14031:2003).

**Determination of endotoxin by the Limulus method**

Supernatant was analysed (in duplicate) for endotoxin using the kinetic Limulus Amoebocyte Lysate test (Kinetic-QCL endotoxin kit; Biowhittaker, Lonza Bio Science, Verviers, Belgium). A standard curve obtained from an Escherichia coli O55:B5 (Biowhittaker) reference endotoxin was used to determine the concentrations in terms of EUs (10.0 EU = 1.0 ng). The limit of detection was 0.005 EU ml⁻¹. The data are presented as nanograms per cubic metre of the air sampling.

**RESULTS**

The concentration of airborne bacteria in the sawmills and carpentries examined is shown in Table 2. The total value of microbial contamination is high for all sampling sites, except at the sites of C1–C4 and D3 where the concentration of total bacteria was lower. Table 2 was also shown Gram negative.

<table>
<thead>
<tr>
<th>Day 1</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>D</th>
<th>E</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Np</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>WS</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>21.7</td>
<td>11.9</td>
<td>22.0</td>
<td>14.2</td>
<td>17.3</td>
<td>9.3</td>
</tr>
<tr>
<td>RH (%)</td>
<td>54.6</td>
<td>53.5</td>
<td>57.2</td>
<td>76.0</td>
<td>49.7</td>
<td>67.9</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Day 2</th>
<th>A</th>
<th>B</th>
<th>C</th>
<th>D</th>
<th>E</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Np</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
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<tr>
<td>WS</td>
<td>2</td>
<td>2</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>22.0</td>
<td>13.3</td>
<td>23.0</td>
<td>15.7</td>
<td>18.0</td>
<td>13.6</td>
</tr>
<tr>
<td>RH (%)</td>
<td>60.0</td>
<td>52.2</td>
<td>65.6</td>
<td>65.2</td>
<td>51.2</td>
<td>63.3</td>
</tr>
</tbody>
</table>

Np, numbers of personal monitoring; WS, workstation monitoring; temperature and RH are mean values measured in two or four locations.
and Gram positive, and the concentration was within a range of 0–164 and 1–104 CFU m\(^{-3}\), respectively.

In air samples taken in the wood factories examined, 19 species of Gram-negative bacteria, 14 species of Gram-positive bacteria, and 18 species of moulds were identified, many of these species were reported as having allergenic, immunotoxic, and/or infectious properties (Johnson et al., 1980; Kagen et al., 1981; Lacey and Crook, 1988; Lacey and Dutkiewicz, 1994; Milanowski et al., 1998; Rylander, 1999; Laitinen et al., 2001) (Table 3).

The samplings carried out by selective media showed in general an evident presence of *Staphylococcus* spp. in all sampling sites and in all wood factories (Fig. 1).

### Table 2. Microbial flora in the air of wood factories: mean concentrations of two consecutive samples, in each workstation. Total cultivable bacteria (TCB at 30 and 37°C) and concentrations of dust and bacterial endotoxins in the wood factories with only personal samplers.

<table>
<thead>
<tr>
<th>Sampling site</th>
<th>(Mean cfu m(^{-3}))</th>
<th>(n)</th>
<th>Concentration of dust (mean, mg m(^{-3}))</th>
<th>Concentration of endotoxin (mean, mg m(^{-3}))</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TCB 37°C</td>
<td>TCB 30°C</td>
<td>Gram-negative</td>
<td>Gram-positive</td>
</tr>
<tr>
<td>Sawmill ‘A’</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A1 sawmill station</td>
<td>1143</td>
<td>1107</td>
<td>164</td>
<td>18</td>
</tr>
<tr>
<td>A2 barker station</td>
<td>842</td>
<td>2000</td>
<td>32</td>
<td>20</td>
</tr>
<tr>
<td>Sawmill ‘B’</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B1 sawmill station</td>
<td>&gt;2000</td>
<td>1104</td>
<td>25</td>
<td>40</td>
</tr>
<tr>
<td>B2 barker station</td>
<td>&gt;2000</td>
<td>1900</td>
<td>15</td>
<td>31</td>
</tr>
<tr>
<td>Carpentry ‘C’</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C1 sanding station</td>
<td>211</td>
<td>167</td>
<td>2</td>
<td>15</td>
</tr>
<tr>
<td>C2 cut station</td>
<td>231</td>
<td>329</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>C3 flanging station</td>
<td>131</td>
<td>852</td>
<td>14</td>
<td>13</td>
</tr>
<tr>
<td>C4 assemblage station</td>
<td>164</td>
<td>345</td>
<td>13</td>
<td>11</td>
</tr>
<tr>
<td>Carpentry ‘D’</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1 sanding station</td>
<td>&gt;2000</td>
<td>297</td>
<td>2</td>
<td>52</td>
</tr>
<tr>
<td>D2 cut station</td>
<td>350</td>
<td>162</td>
<td>0</td>
<td>28</td>
</tr>
<tr>
<td>D3 flanging station</td>
<td>173</td>
<td>180</td>
<td>4</td>
<td>27</td>
</tr>
<tr>
<td>D4 assemblage station</td>
<td>&gt;2000</td>
<td>142</td>
<td>3</td>
<td>34</td>
</tr>
<tr>
<td>Carpentry ‘E’</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>E1 sanding station</td>
<td>&gt;2000</td>
<td>&gt;2000</td>
<td>16</td>
<td>79</td>
</tr>
<tr>
<td>E2 cut station</td>
<td>&gt;2000</td>
<td>&gt;2000</td>
<td>5</td>
<td>33</td>
</tr>
<tr>
<td>E3 flanging station</td>
<td>&gt;2000</td>
<td>&gt;2000</td>
<td>2</td>
<td>104</td>
</tr>
<tr>
<td>E4 assemblage station</td>
<td>&gt;2000</td>
<td>&gt;2000</td>
<td>4</td>
<td>22</td>
</tr>
<tr>
<td>Carpentry ‘F’</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F1 sanding station</td>
<td>929</td>
<td>266</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>F2 cut station</td>
<td>226</td>
<td>160</td>
<td>9</td>
<td>1</td>
</tr>
<tr>
<td>F3 flanging station</td>
<td>628</td>
<td>252</td>
<td>1</td>
<td>6</td>
</tr>
<tr>
<td>F4 assemblage station</td>
<td>&gt;2000</td>
<td>&gt;2000</td>
<td>2</td>
<td>9</td>
</tr>
</tbody>
</table>

### Table 3. List of species and genera of bacteria and moulds identified in air samples from wood factories.

**Gram-negative:** *Klebsiella pneumoniae*, *Pantoea* spp., *Enterobacter cloacae*, *Aeromonas hydrophila/caviae*, *Chryseomonas luteola*, *Pseudomonas luteola*, *Shigella sonni*, *Rhahnella aquatilis*, *Pseudomonas fluorescens*, *Brevundimonas vesicularis*, *Escherichia vulneris*, *Burkolderia cepacia*, *Pseudomonas stutzeri*, *Moraxella* spp., *Stenotrophomonas maltophilia*, *Bordetella* spp., *Flavimonas oryzae*, *Agrobacterium radiobacter*

**Gram-positive:** *Staphylococcus capitis*, *Staphylococcus sciuri*, *Staphylococcus xylosus*, *Micrococcus* spp., *Staphylococcus cohnii cohnii*, *Staphylococcus lentus*, *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Staphylococcus warneri*, *Staphylococcus hominis*, *Staphylococcus haemolyticus*, *Bacillus circulans*, *Bacillus megaterium*, *Bacillus lentus*, *Streptococcus equinus*.

As for the carpentries ‘A’, only in the A1, the sawmill station, the percentage of Staphylococcus spp., was smaller than that Coliforms. In other carpentries examined, the only station where the percentage of Staphylococcus spp. was decreased as to the Enterobacteriaceae was in the cutting station (F2) of the Carpentry ‘F’.

The sampling by the use of personal samplers for the detection of the inhalable fraction of endotoxin and wood dust shows the presence of bacterial endotoxin at all stages and in all the carpentry work but at lower levels than the limit proposed by DECOS, with regard to the levels of OEL of 5 ng m$^{-3}$, except in some cases, that is in A1 during the cut, C4 in the assemblage station, and here again F2 during cutting. It is evident that the highest concentration of endotoxin in these two workstations (A1, C4, and F2) corresponds with the presence of Gram-negative bacteria (Fig. 1). But the concentration of endotoxin also depends on the temperature and RH; in fact, after conducting a statistical analysis on the possible correlation between temperature, RH, and endotoxin data show that there is correlation very significant (Spearman test: $r_s = 0.847$ $P < 0.001$ for temperature vs endotoxin; $r_s = 0.766$ $P < 0.001$ for RH vs endotoxin, $n = 20$).

With regard to wood dust, only in some cases, they exceed the exposure limits allowed by law (D.Lgs. 81/08 attachment XLIII), which imposes a limit of 5 mg m$^{-3}$ for inalable dust. In all cases examined, the values were within the limits of law, only the E1 shows values of wood dust of 11.28 mg m$^{-3}$.

**DISCUSSION AND CONCLUSIONS**

The first aspect to consider is the concentration of bacteria in all the sawmills and carpentries examined. Total bacteria concentrations at 30 and 37°C in the air were of the order of $10^2 - 10^3$ CFU m$^{-3}$. In addition, the concentrations of Gram-negative bacteria in wood factories examined were of the order of $10^1 - 10^2$ CFU m$^{-3}$; values found were smaller than those found in other working environments where there were greater amounts of organic dust, such as animal farms or grain industry facilities (Dutkiewicz, 1978; Dutkiewicz et al., 2000). The concentration of Gram negative was higher in these workplaces because there is a high level of contamination of the materials (hay, grain, etc.). On the other hand, these values are very similar to those found in various wood processing factories, for example in a Polish pine processing sawmill, but lower compared with levels observed in other joineries in different States (Alwis et al., 1999; Mandryk et al., 1999, 2000; Dutkiewicz et al., 2001a). In some cases, the concentration of Gram-negative bacteria may depend on the type of wood that is processed as described in the paper of Prazmo et al. (2000). In fact, the concentration of Gram-negative bacteria in pine sawmills was lower than that found in beech sawmills. Another factor that may affect the concentration of Gram-negative bacteria, as well as with endotoxin, is the climate (temperature and humidity).
It has been shown that the greatest risk of exposure to Gram-negative bacteria occurs in the production cycle of the sawmill A. In the other stages of wood processing, the concentration of Gram-negative bacteria in the air is ~10 times lower. Although the type of work is the same as in A1 and B1, the presence of Gram-negative bacteria may be influenced by the temperature difference measured at two sites, as shown in Table 1. In fact, in the A1 site, the temperature was 21.8°C and in B1 was 12.6°C in average in the 2 days of measurement. In addition to the number of microorganisms found in the workplaces examined, we have identified several species of microorganisms, as shown in Table 3. It has been shown that many of these microorganisms as Enterobacter spp., Pantoea spp., Rahnella spp., and Klebsiella spp. are producers of endotoxin and exhibit a high degree of biological activity (Dutkiewicz et al., 1987, Hølt et al., 1994) and often show strong allergenic properties, increasing then the risk of diseases in workers exposed (Skórska et al., 1996; Milanowski et al., 1998; Prazmo et al., 2000; Dutkiewicz et al., 2001a,b). In the wood factories examined, potentially pathogenic non-enterobacterial Gram-negative species were also found: Pseudomonas fluorescens implicated in the aetiology of allergic disease among machine industry workers exposed to metalworking fluids (Bernstein et al., 1995), and Aeromonas hydrophila suspected to produce exotoxin causing gastrointestinal symptoms in workers of a sewage treatment plant (Rylander et al., 1982).

With regard to Gram-positive bacteria identified, most belonging to Staphylococcus genus, as shown in Table 3. Different papers reported that strains belonging to the species Staphylococcus hominis, Staphylococcus capitis, Staphylococcus lugdunensis, Staphylococcus warneri, and Staphylococcus saccharolyticus can occasionally cause bacteremia and infective endocarditis (Kloos and Bannerman, 1994; Schnitzler et al., 1997; Weinstein et al., 1998; Wieser and Busse, 2000). Among airborne fungi identified, Penicillium spp. and Alternaria alternata pose the greatest risk of evoking allergic disease. A potential risk to fungi was diminished by their very low concentration and by the absence of pathogenic Aspergillus strains, in particular Aspergillus fumigatus.

The assessment of microbial contamination according to the guidelines given by the European Commission (CEN 1993) (Table 4) shows that in all the companies examined, a level of contamination is between middle and very high, and in any case, the contamination of the stations from 500 to >2000 CFU m⁻³. The carpentry ‘E’ had the most polluted air, showing high levels of contamination in all workstations. The lower concentration of microorganisms in the air of the joinery ‘C’ was in agreement with the presence of a ventilation system more modern than the other joiners. However, these results should be interpreted taking into account that the document refers to non-industrial workplaces. In fact, in the sawmills located in various parts of the world, the concentrations of airborne microorganisms were of the order 10⁹–10¹⁰ cfu m⁻³, as in sawmills and carpentries examined in this paper (Alwis et al., 1999; Mandryk et al., 1999, 2000; Duchaine, et al., 2000). Although these concentrations were measured frequently, however, they can represent a hazard to the health of workers exposed because the risk may depend not only on the concentration of microorganisms but also on their type, as mentioned previously.

Because there are no legal regulations on microbial contamination in industrial workplaces, both in Italian and in international law, the only reference are tables of CEN (1993), although for industrial areas should be used more wisely.

In fact, as shown in Table 4, the monitored workstations in the different carpentries, compared with the levels of the Classification of indoor total microbial contamination, were between a middle and a very high level, although this level of contamination is easily measurable in industrial workplaces. Then, the total microbial contamination was classified by the tables recommended by Commission of European Communities for the quality of indoor air in non-industrial workplaces (CEN, 1993), but remembering what was said previously.

Table 4. Classification of indoor air quality in each workstation in wood factories examined. Tables recommended by CEN (1993) for the quality of indoor air in non-industrial workplace.

<table>
<thead>
<tr>
<th>Classification of indoor total microbial concentration</th>
<th>Concentration CFU m⁻³</th>
<th>Workstation of carpentries</th>
</tr>
</thead>
<tbody>
<tr>
<td>Very low</td>
<td>&lt;50</td>
<td>C1, C2, C3, C4, D2, D3, F2, F3</td>
</tr>
<tr>
<td>Low</td>
<td>50–100</td>
<td>A1, A2, B1, B2, D1, D4, F1</td>
</tr>
<tr>
<td>Middle</td>
<td>101–500</td>
<td>E1, E2, E3, E4, F4</td>
</tr>
<tr>
<td>High</td>
<td>501–2000</td>
<td></td>
</tr>
<tr>
<td>Very high</td>
<td>&gt;2000</td>
<td></td>
</tr>
</tbody>
</table>
Some countries, like Poland, have proposed limits for bioaerosols in industrial workplaces. These values are much higher (~1000-fold) than those proposed for non-industrial workplaces. But, no international organization (AIHA, ACGIH, NIOSH, and OSHA) has managed to define limit values for bioaerosols in the workplace. The lack of standards and limit values for biological agents are due to several reasons. Firstly, dose–response relationships are still indeterminate and controversial in many aspects. Furthermore, many species of microorganisms and products of microbiological origin cause similar health effects. In addition, the susceptibility to biohazards is an individual feature of each person. Finally, there is a lack of measurement data of concentrations of biological agents in different working environments, which is mainly due to a lack of resources available to perform the measurements and not only to a lack of standardized sampling methods and experimental procedures.

In all carpentries, different selective culture media were used for many bacterial types in order to have a complete picture of air quality. As can be seen from Fig. 1, the percentage composition of microbial flora changes in the different companies and from point to point, although you may notice a massive presence of Staphylococcus spp. in all the sampled points. In some areas, the percentage of Staphylococci decreased in favour of the presence of Gram-negative bacteria (A1, C3, C4, and F2).

The concentration of dust in the air of the facilities examined was below the Italian OEL level of 5.0 mg m\(^{-3}\) (Italy, 2008), in almost all workplaces, except in one case, the station of sanding the carpentry ‘E’ (E1), where the concentration of wood dust is 11.28 mg m\(^{-3}\).

Bacterial endotoxin concentrations in the air of the wood factories examined was variable, of the order of 0.14–6.93 ng m\(^{-3}\). Even within the same joinery, there are sites where the presence of endotoxin is greater; this shows a different exposure to endotoxin in workers performing different tasks, creating a distinct health hazard for exposed workers. The average concentration of endotoxin in the four carpentries examined in this study was 1.76 ng m\(^{-3}\).

About the PVC filter, these have been used by many authors to monitor endotoxin in different workplaces. For example, the PVC filter was utilized in many papers for the standardization of methods for assessing the presence of endotoxins (Laitinen, 1999; Duchaine et al., 2001; Astrakianakis et al., 2006). In 2006, Harper assessed the presence of endotoxin in woodworking shops (Harper and Andrew, 2006), by using the filters in PVC. In this paper, the results were then compared with those obtained in carpentries in Australia and Tanzania for wood dust and endotoxin.

Comparing our results with those presented in the work of Harper, it is clear that the concentration of endotoxin is similar to what he described but significantly lower than that shown by Alwis et al. (1999) in Australia and Rogo Rongo et al. (2004) in Tanzania, where the concentration of endotoxin was 2.4 and 9.1 ng m\(^{-3}\), respectively. This difference for endotoxin may be due to the type of wood, or to the climate, but may also be due to differences in drying practice. In fact, in sawmills, the average concentration of endotoxin (2.66 ng m\(^{-3}\)) from fresh wood is greater than the carpentries, as was shown by Alwis, which contains a concentration of endotoxins in the sawmills of 4.3 ng m\(^{-3}\).

The concentrations of endotoxin in the air in all cases do not exceed the value of 0.2 \(\mu\)g m\(^{-3}\), which has been shown to cause a decrease of lung function during work shift, or the values of 1–2 \(\mu\)g m\(^{-3}\) which are supposed ODTS to evoke symptoms (Rylander, 1994).

It is noteworthy that the largest value of endotoxin concentration in the air equal to 6.93 ng m\(^{-3}\) was recorded at the cut site in the carpentry ‘F’ (F2). This rise of airborne endotoxin could be the result of a higher concentration of Gram-negative bacteria in the cut station (Fig. 1). In the other cases, this increase may be caused not only by the concentration of Gram-negative but also could be the result of elevated temperature and humidity in the wood factories as it is known that heating might enhance the biological activity of the endotoxin by changing its physical structure (Rylander, 1987). In conclusion, although bacteria, endotoxins, and dust occur in the air of wood factories in relatively low concentrations, they may exert adverse effects on exposed workers, as evidenced by the presence of numerous potentially pathogenic species. Thus, these microorganisms pose a potential risk of respiratory disease for the workers, in particular for those engaged in sawmill and sanding of wood. The results of this study emphasize both the level of microbial contamination of the air related to woodworking and the chemical risks linked to the presence of wood dust. The presence of endotoxins and toxic or allergenic microorganisms may affect the health of exposed workers, although at present, there are no limits accepted by the scientific community to assess the risk of exposure. For these reasons, it is necessary a more careful monitoring of these work environments in order to identify the common threshold of risk with regard to biological agents and it is especially important to contain the presence of the wood dust.
No less important information and training of workers for proper use of personal protective equipment. In fact, the control of dust exposure is not an easy task, especially in small workplaces or in particular types of work, in which it is difficult to ensure proper ventilation. Furthermore, we must remember that these kinds of work activities, involving the simultaneous handling of different types of wood, produce complex mixtures of powders with various health risks.

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**REFERENCES**

ACGIH. (2010) TLVs and BEIs based on the documentation of the threshold limit values for chemical substances and physical agents & biological exposure indexes. Cincinnati, OH: American Conference of Governmental Industrial Hygienists.


Airborne microorganisms, endotoxin, and dust concentration in wood factories in Italy


