ORIGINAL ARTICLES

BACTERIAL CONTAMINATION OF INDOOR AIR, SURFACES, AND SETTLED DUST, AND RELATED DUST ENDOTOXIN CONCENTRATIONS IN HEALTHY OFFICE BUILDINGS

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Abstract: Endotoxin, a characteristic external fraction of the outer membrane from Gram-negative bacteria, continuously shed into the environment, is considered as an important risk factor for human health. Our purpose was to study the bacterial species contaminating healthy working environments. Airborne, working surfaces and carpet dust samples were collected from 25 offices. Bacterial species were identified with biochemical ApiSystem® strips. Endotoxin concentrations in settled dust were measured with the kinetic chromogenic Limulus assay. The airborne bacterial level varied from 44-2,511, with a median of 277 cfu/m3. Bacterial contamination on surfaces ranged from 1-1000, with 33 cfu/25 cm² as median value. On carpets, bacterial concentration ranged from 0.73–185 \times 10⁵ cfu/g, with 7.28 \times 10⁵ cfu/g as median value. Endotoxin concentration varied from 4.6-116.2 EU/mg, with a median of 20.3 EU/mg. Altogether, 501 bacterial strains were isolated. The species variability was greater in Gram-negative bacteria than in Gram-positive cocci with 41 versus 34 various species. In conclusion, people working in healthy offices can be exposed to large concentrations of airborne and dust bacteria and related endotoxin concentrations, giving a risk of work-related diseases.

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Key words: healthy office buildings, occupational exposure, settled dust, bioaerosols, bacteria, endotoxin.

INTRODUCTION

During occupational activities or in home environments many individuals are exposed to dust from vegetable, animal or microbial origin. Inhalation of endotoxins, major component of the outer membrane of Gramnegative bacteria (GNB), carried by airborne dust, leads to some adverse effects on human health. Endotoxins initiate a cascade of biochemical and cellular events giving rise to multiple dysfunction associated to the sick building syndrome (SBS), or to acute chronic lung diseases [15, 27]. SBS, defined by the World Health Organisation as symptoms involving unpaired performance, such as nasal and pharyngeal mucous membrane irritations, skin dryness, itchy eyes, headache, shortness of breath, wheeze or asthma, is an emerging problem in many countries [17, 20, 21, 23]. On the other hand, several studies suggest that endotoxins are also potent

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stimulators of the immune system, and that endotoxin exposure in early life minimizes the risk of developing atopic diseases [19, 30]. The aim of this work was to conduct a survey on qualitative and quantitative bacterial contamination of air, surfaces and settled carpet dust in healthy office buildings. Endotoxins were measured in dust from carpeted floors. The diversity of GNB, the source of endotoxin, was investigated in healthy working environments.

MATERIALS AND METHODS

Study location. A complex of 6 healthy office buildings located in the city of Luxemburg (The Grand Duchy of Luxemburg) were investigated - a total of 25 rooms, 15 offices with carpeted floors and 5 offices, 4 classrooms of a nursery school and a training room, also with synthetic floors. Basic information, such as the presence of green plants, smokers, and photocopiers were recorded. The clearing regimes were similar for each building. There were no air-conditioning systems or other ventilation installations in the buildings. All samples were taken on the same day in June 2001 during normal occupational activities.

Measurements of physical indoor air characteristics. Temperature and relative humidity were monitored with a portable combined thermo-hygrometer (PolyLabo, Belgium).

Airborne bacteria sampling. Field measurements were made in duplicate with a "Merck100 Air Sampler" or MAS (Merck, Germany). The sampling height, which approximated the breathing zone of the rooms' occupants, varied from 0.75–1.2 m above floor level. Bacteria were collected on TSA medium (Oxoid, England). The air volume sampled was 180 litres. Plates were incubated at $32\pm2.5^{\circ}$ C, during 2–5 days for the detection and the enumeration of aerosolised bacteria. Bacterial counts were expressed as colony forming units per cubic metre (cfu/m³).

Surfaces studies. For each room, 4 surfaces were monitored at different representative points of human activities with Bacto[®]letheen agar Rodac[®]plates (Difco laboratories, USA). In the offices, samples were made on desk blotters, conference tables, and computer tables, or near photocopiers. In the training room, plain areas of muscle development machines or bench seats were chosen and in classrooms samples were made on play tables. Plates were incubated at 32±2.5°C. Bacterial counts were expressed as colony forming units per 25 centimetre square area (cfu/25 cm²).

Settled dust sampling. Dust from the carpets was collected with cordless portable vacuum cleaners (HC300Dustbuster[®] Black&Decker[®], Belgium) by sampling a square metre during 5 min. The succion air flow rate was 650 l/min. The dust-exhaust vacuum filter was covered, after cleaning, with pure bleach water and

ethanol, with a sterile paper filter. Less than 24 hours after sampling, sterile normal saline dilutions of dust were inoculated in triplicate on TSA plates to obtain the total viable bacterial contamination. Plates were incubated at 32 ± 2.5 °C during 2–5 days, and bacterial counts were expressed as colony forming units per gram of dust (cfu/g). For endotoxin assays, the dusts were stored at -30°C until further analysis.

Bacterial identification. Each colony type, taking into account their macroscopic description, was cultured on TSA plates before identification. Gram-positive cocci (GPC) and GNB bacterial species were characterised, after a Gram staining, with biochemical strip tests "APISystem" interpreted with the data base "APILAB Plus Software" (bioMérieux, Marcy-l'Etoile, France).

Endotoxin assay in carpet dust. Dust samples were extracted in 5 ml pyrogen free water (BioWhittaker, Europe), rocked either vigorously for 1 minute and placed in an ultrasonic bath (Fisher Bioblock Scientific, Belgium) at 75°C during 30 minutes. Endotoxins were assayed immediately after the extraction procedure with a quantitative kinetic chromogenic Limulus Amebocytes lysate (LAL) method "Kinetic-QCLTM" (BioWhittaker, Europe). Analyses were performed with an automated microtitre plate (Falcon MicrotestTM, USA) reader Kinetic-QCL monitored by a «WinKQCL 1.2 software» (BioWhittaker, Europe). The control standard endotoxin Escherichia coli strain 055:B5 (BioWhittaker, Europe) was calibrated versus the United States Reference Standard EC-6. To avoid activators/inhibitors interference with the LAL, parallel dilutions were spiked with endotoxin at 0.5 endotoxin units (EU) per ml. The sensitivity of our assays was 0.005 EU/ml. Serial dilutions of each sample were run in duplicate. Endotoxin concentrations were expressed in EU per gram of dust (EU/g) and in EU per square metre (EU/ m^2).

Statistical analysis. The results were analysed with the tests of Kolmogorov-Smirnov and Anova to compare the median between the studied parameters.

RESULTS

Room's characteristics. During the field sampling day temperature ranged from 19–28°C with an arithmetic mean of 23.7°C. Relative humidity varied from 38–59% with an arithmetic mean of 48%.

Bacterial characteristics

In the air (n=25). Bacterial levels ranged from 44–450, with a median of 177 cfu/m^3 in rooms with a carpeted floor (n=15). In rooms with synthetic floor, contamination varied from 122–794, with a median of 189 cfu/m^3 for the offices (n=5) and from 428–2,511 with a median of 708 cfu/m^3 for the nursery schools (n=4). The

Table 1. List of microbial species identified in healthy offices.

Gram-positive cocci

Catalase positive: Dermacoccus nishinomiyaensis [A2], Kocuria kristinae [A3 S2], K. rosea [A10 C1], K. varians [A1], Kytococcus sedentarius [A2], Micrococcus luteus [A19 S17 C6], M. lylae [A1 S2 C3], Staphylococcus aureus [S4 C4], S. auricularis [A2 S2], S. capitis [A3 S15 C7], S. caprae [C5], S. carnosus [C7], S. chromogenes [A1 S1], S. cohnii [A5 S6 C4], S. epidermidis [A16 S14 C8], S. haemolyticus [A10 S6 C10], S. hominis [A15 S19 C4], S. hyicus [C1], S. lentus [A2], S. lugdunensis [A4 S3], S. saprophyticus [A5 S7 C7], S. sciuri [A4 S3 C2], S. simulans [S2 C11], S. warneri [A6 S10 C7], S. xylosus [A5 S5 C2], Stomatococcus mucilaginosus [A3 S3].

Catalase negative: Aerococcus viridans [C1], Enterococcus casseliflavus [C5], E. durans [C4], E. faecalis [C3], E. faecium [C13], E. gallinarum [C3], Gemella haemolysans [S1], Lactococcus lactis [C1].

Gram-negative bacteria

Oxidase positive: Alcaligenes denitrificans [S1 C2], Aeromonas salmonicida [A2 S2], Agrobacterium radiobacter [S1], Brevundimonas vesicularis [A2 S1], Chryseobacterium indologenes [A2 S1], Comamonas acidovorans [C1], C. testoteroni [A1 S2], Empedobacter brevis [A1], Methylobacterium mesophilicum [S1], Moraxella spp. [A1 S3], Ochrobactrum anthropi [A3 S10], Oligella ureolytica [S2], Pasteurella haemolytica [A2 S3], Pseudomonas alcaligenes [A1 S4], P. fluorescens [A2 S2 C2], P. stutzeri [A2], P. putida [C6], Psychrobacter phenylpyruvicus [S1], Ralstonia pickettii [A1 C1], Sphingomonas paucimobilis [A1 S3].

Oxidase negative: Acinetobacter baumannii [A1 S1], A. calcoaceticus [A1 C3], Citrobacter braackii [C1], Chryseomonas luteola [A1], Enterobacter amnigenus [C4], E. cloacae [S1 C4], E. sakazakii [A1 C1], Escherichia coli [C2], E. hermannii [C1], E. vulneris [A1 C3], Erwinia spp. [S2], Ewingella americana [S2], Hafnia alvei [C1], Flavimonas orizyhabitans [A2 S3], Pantoea spp. 1 [A1 C2], Pantoea spp. 2 [C2], Pantoea spp. 3 [A4 S5 C9], Pantoea spp. 4 [C4], Serratia ficaria [C1], S. rubidaea [C1], Stenotrophomonas maltophilia [A3 S2 C1].

Sites of isolation are given in brackets. The letter means that the species was isolated from the air [A], on surfaces [S], on carpets [C] with the number of sites where it was identified.

difference of airborne bacterial contamination between offices with carpeted and synthetic floors and nurseries was statistically significant (p=0.010). The gymnasium (n=1) showed a value of 1,572 cfu/m³. A total of 155 bacterial strains were isolated, 119 were GPC with 21 various species, and 36 were GNB with 22 different species. No catalase negative GPC were found from the airborne samples. Oxidase positive GNB species showed a greater variability than oxidase negative ones; 13 oxidase positive versus 9 oxidase negative species were identified. The airborne microorganisms isolated with the highest frequency were GPC from human skin, between which Micrococcus luteus (19/25), Staphylococcus and S. epidermidis (16/25)hominis (15/25).Staphylococcus sciuri (4/25) and S. lentus (2/25), species from veterinary sources, appeared punctually. Few GNB were present in the air. Pantoea spp. 3 (4/25), Stenotrophomonas maltophilia (3/25) and Ochrobactrum anthropi (3/25) were the species mostly encountered. The list of the airborne bacterial species identified, noted [A], is detailed in Table 1.

On surfaces (n=97). Bacterial contamination on surfaces in carpeted office floors (n=58) varied from 2–1,000 with 26 cfu/25 cm² as median value and from 1–86 with a median of 20 cfu/25 cm² for synthetic floor (n=20). On surfaces in nursery schools (n=16), bacterial levels ranged from 3–120 with a median of 20 cfu/25 cm². From the gymnasium, surface (n=3) contamination varied from 12–250 with 23 cfu/25 cm² as median value. On the 97 surface samples taken, a total of 175 bacterial identifications were made. 122 being composed of GPC with 18 catalase positive various species, and 1 catalase negative identified once on a carpeted office floor, *Gemella haemolysans* and 53 GNB with 22 various species. As in the air samples, oxidase positive species showed a greater

variability than oxidase negative species; 15 oxidase positive versus 7 oxidase negative species were identified. The dominant microorganisms on surfaces belonged, as in the air, to the *Staphylococcaeae* family such as *Staphylococcus hominis* (19/25), *Micrococcus luteus* (17/25) and *Staphylococcus capitis* (15/25). *Staphylococcus aureus* was found at 4 sites including 2 classrooms. Two veterinary species, *Staphylococcus chromogenes* and *S. sciuri*, appeared respectively in 1 and 3 rooms with carpeted floors. Among the GNB, *Ochrobactrum anthropi* (10/25) and *Pantoea* spp. 3 (5/25) prevailed. Detailed bacterial species identified on surfaces, noted [S], are presented in Table 1.

On settled dust (n=15). On carpets, bacterial levels ranged from $0.73-185 \times 10^5$ cfu/g, with 7.28×10^5 cfu/g as median value. Endotoxins were detected in all samples, concentrations expressed in EU/mg varied from 4.6-116.2 with a median of 20.3 EU/mg while, when expressed in EU/m², endotoxin levels ranged within 304.0–21,864.0, with 6,079.0 EU/m^2 as median value. A total of 171 identifications were made, among which were 119 GPC with 17 catalase positive and 7 catalase negative various species and 52 GNB with 21 various species. Oxidase negative GNB species showed a greater variety than oxidase positive bacilli; 16 oxidase negative versus 5 oxidase positive species were identified. Among the GPC, germs identified with the highest frequency belonged to the Streptococcaceae family. Enterococcus faecium was identified on 13/15 carpets. From the group of Micrococcaceae, Staphylococcus simulans (11/15) and S. haemolyticus (10/15) prevailed. Staphylococcus aureus was identified in 4 offices. Veterinary species, such as Staphylococcus caprae, was found only in settled dust in 5/15 offices. Staphylococcus sciuri appeared on 2/5 carpets. The dominant GNB species, belonging to the

	Air (N=25)			Surfaces (N=97)			Settled dust (N=15)		
-	n	%	n/N	n	%	n/N	n	%	n/N
Isolated strains									
GPC	119	76.8	4.76	122	69.7	1.25	119	69.6	7.93
GNB	36	23.2	1.44	53	30.3	0.55	52	30.4	3.46
Total	155	100	6.2	175	100	1.80	171	100	11.4
Identified species									
GPC catalase positive	21	100		18	94.7		17	70.8	
GPC catalase negative	0	0		1	5.3		7	29.2	
Total of GPC	21	100		19	100		24	100	
GNB oxidase positive	13	59.1		15	68.2		5	23.8	
GNB oxidase negative	9	40.9		7	31.8		16	76.2	
Total of GNB	22	100		22	100		21	100	
Total of species	43			41			45		

Table 2. Number of isolated strains, identified species, their ratio and percentage in the air, on surfaces and in settled dust.

Pantoea spp., was isolated in 12/15 offices. *Pseudomonas putida* contaminated 6 carpets. Detailed bacterial species identified in settled carpet dust, noted [C], are listed in Table 1. Table 2 summarises the numbers of isolated strains, the identified species, their ratio and their percentage in the air, on surfaces and in settled dust.

DISCUSSION

Measurements were carried out in offices to characterise the bacterial flora in the air, on surfaces and in settled dust with endotoxin related release from healthy working environments.

In the air. Our data obtained for bacterial contamination ranged from 44-450 cfu/m³ for carpeted office floors and from 122–794 cfu/m³ for synthetic office floors. Bacterial levels varied from 428-2,511 cfu/m³ in nursery schools, and the gymnasium showed a value of 1,572 cfu/m³. In classrooms or in the training room, where the number of people and their movements are important, the airborne bacterial levels were normally higher than in offices. To date, there are no internationally recognised Occupational Exposure Limit (OEL) values or Threshold Limit Values (TLV) for bioaerosols [7]. Moreover, comparisons are difficult due to the variability in air sampling methods used in the studies. Dutkiewicz [7] cited the OEL values of 10⁵ cfu/m³ or of 10⁴ cfu/m³ for total microorganisms proposed by Malmros et al. These limits, established in working environments where people are exposed to large quantities of organic dust, such as in agriculture or waste treatments, were hundred to thousand folds higher than our values. Dacarro et al. proposed a global index of microbial contamination per cubic metre (GIMC/m³) for the assessment of air quality in buildings based on results obtained from 226 offices. In this study, 95.5% of the offices had a GIMC/m³ value below the 1,000 proposed as a threshold limit for healthy offices [3]. The values recorded in our study for the offices were all below this value. We obtained similar results to others, such as Sessa et al. in Rome, who observed that in the presence of people the average airborne bacterial concentrations were higher than in their absence, respectively 493 and 126 cfu/m³ [28]. For 6 large office buildings in metropolitan areas in Iowa, Minnesota and Nebraska, USA, Reynolds et al. indicated a maximum value of 150 cfu/m³ [26]. Twelve of our 20 offices exceed this value. Bholah and Subratty found concentrations ranging between 3 and 1,110 cfu/m³ in 23 buildings in Mauritius [1]. A study conducted in Estonia by Indermitte and cited by Górny and Dutkiewicz pointed out an averaged airborne contamination of 384 cfu/m³ in 4 office buildings [13]. Airborne bacterial levels in offices were lower, anyway, than those found in other occupational environments. I.e. reviewed data, on industrial environments located in eastern Poland, reported total mesophilic bacterial ranges of 0.24–7.07 \times 10^3 cfu/m 3 in a municipal sewage treatment plant [25]; of 2.83–9.31 \times 10⁴ cfu/m³ in a potato processing plant [11]; of 0.19–2.75 \times 10⁴ cfu/m³ in a furniture factories [16]; of $0.72-9.12 \times 10^4$ cfu/m³ in a sawmills [9] and of $7.18-9.52 \times 10^4$ cfu/m³ in a fibreboard factory [10]. At least, bacterial airborne contaminations in offices were lower than in domestic environments. In Poland, Górny and Dutkiewicz reported concentrations of airborne bacteria in healthy dwellings between 88-4,297 cfu/m³ [13]. Moreover, it should be borne in mind that results obtained for airborne contamination give a somewhat incomplete picture of the total exposure assessment of airborne viable bacteria. The number of culturable microorganisms may underestimate the viable number because the method probably compromised bacterial viability by damage incurred during sampling. Nevertheless, the obtained data can be considered as contributing towards the identification of acceptable levels for bioaerosols in common healthy indoor environments. Several researchers have evaluated quantitative indoors bacterial composition in occupational environments; few investigators, however, have examined in detail the bacterial species found in office environments. In this study, 119 GPC strains were isolated representing

21 various species and 36 GNB strains representing 22 various species. All the isolated GPC belonged to the Micrococcaceae family and were closely related to humans or animals. In our study, Micrococcus luteus (19/25), Staphylococcus epidermidis (16/25) and S. hominis (13/25) were commonly identified in more than 60% of samples, whatever the nature of the sampled room. Several species occur frequently but exclusively in the air, such as Dermacoccus nishinomiyaensis, Kocuria varians and Kytococcus sedentarius. Our results are in accordance with those presented by Górny and Dutkiewicz, in the indoor air of 60 human dwellings situated in upper Silesia where Micrococcus/Kocuria species and Staphylococcus species occurred in 100% of the samples. However, the authors recorded the presence of Pseudomonaceae in 80% of the examined sites versus 20% in our study, and the presence of Aeromonas spp. in 40% versus 8% in our samples [13]. Prażmo et al. [25] found similar results about bacterial composition in a municipal sewage treatment plant located in eastern Poland. Gram-positive bacteria Micrococcus/Staphylococcus distinctly prevailed among the airborne microorganisms. Among the GNB, Enterobacter cloacae, followed by Acinetobacter calcoaceticus, Pseudomonas species and Stenotrophomonas maltophilia occurred commonly [25]. In a cattle feedlot pen, all the airborne bacteria collected with an Andersen biological cascade sampler were GPC [32], while in pig houses the airborne bacteria were dominated by Enterobacteriaceae in which the species Escherichia coli and Enterobacter agglomerans prevailed [33]. In our study, no faecal species, such as Escherichia coli or Enterococci were isolated from the air.

On surfaces. The main researches about bacterial contamination of surfaces were made in food processing environments or in special care units within a context of microbial quality control. Publications about offices or domestic environments were not found, therefore observations are original in this field. The values of bacterial contamination were statistically different (p=0.048), mostly between offices with and without carpets. The maximum values of bacterial levels reached 1,000 cfu/25 cm² for carpeted office floors; 86 cfu/25 cm² for offices with synthetic floor; 120 for the classrooms and 250 cfu/25 cm² for the training room. Surfaces of 7 carpeted floor offices showed a higher contamination level than those encountered in the other sites. Among the GPC, 3 species were mostly identified namely: Micrococcus luteus, Staphylococcus capitis and S. hominis, host of the human or animals skin. Gemella haemolysans, identified once on a carpeted office floor, is a parasite of mammals found in bronchial secretions from the respiratory tract. The GNB species isolated were all from environmental sources, such as soil, plants and water. More particularly, species such as Agrobacterium radiobacter and Methylobacterium mesophilicum were present only on working surfaces in offices with pot plants.

In settled dust. The bacterial contamination in carpet dust range from $0.73-185 \times 10^5$ with 7.28×10^5 cfu/g as median value. Data about bacterial levels in settled dust are less numerous than those concerning airborne contamination. In domestic environments, Horak et al. reported an average of 16×10^5 cfu/g of bed dusts from homes in Upper Silesia [14]. In industrial environments, particularly in agricultural concerns, settled dust contaminations were higher. Total bacterial plate counts from dust of 5 grain elevators along the lower Mississippi River ranged from $19-534 \times 10^5$ cfu/g [4]. In corn silage near Cooperstown in New York, Dutkiewicz et al. found bacterial contamination up to 10^9 cfu/g [6]. Near Shanghai, in factories processing rice and wheat straw, Shen *et al.* pointed out bacterial levels ranging from 10^7 – 10^9 cfu/g of dust [29]. Our results were closer to those found in domestic environments than with those found in other more specifics occupational contexts. In our samples, germs belonging to the Micrococcaceae and especially Sphaphylococcus species occured mostly. Several species such as Staphylococcus caprae, S. carnosus and S. hyicus were found only on carpets. Contrarily to germs identified on surfaces or in airborne dust, Streptococcaceae like Enterococcus faecium (13/15) or E. faecalis (3/15) were isolated only in carpet dust; some of them being of faecal origin. Among GNB occurring in settled dusts and conversely to the other samples, oxidase negative bacteria showed a greater variety than oxidase positive bacteria.

In summary, a total of 501 bacterial strains were isolated, including 26 GPC catalase positive, 8 GPC catalase negative, 21 GNB oxidase negative and 20 GNB oxidase positive species. The range of the bio-diversity was thus greater with the GNB than with the GPC. If we take into account the ratio of the sampling number to the related isolated strains, we obtain 11.4 for the settled dust, 6.2 for the air and only 1.8 for the surfaces. Settled dust therefore seems to be a better environmental support for the bacterial survival and could be considered as a reservoir of germs for surface or air contamination.

While it is generally accepted that variations in bacterial composition of airborne or settled dust are associated with environmental characteristics, the strength of this association remains complex. However, no standard sampling method or culture media have been adopted to ensure the validity of the studies and allowing comparisons [18]. The risk of exposure to microflora in offices is increased by the presence of species that may evoke atopic reactions. The obtained data can be considered as a step towards identifying which species were the best endotoxins producers in common indoor environments.

Endotoxin. In settled carpet dust, endotoxin concentration range from 4.6–116.2 EU/mg with a median of 20.3 EU/mg. Domestic environments were the most similar context to ours. Our findings were consistent with previous studies conducted in house dust from carpeted living rooms or on kitchen floors. In previous studies, we measured mean endotoxin concentrations of 17.8 EU/mg in dust from mattresses and 18.6 EU/mg in dust from floors [22]. Böttcher et al. found a median of 16.1 EU/mg with a range of 0.25-358.0 EU/mg from carpets in Estonian and Swedish homes [2], while Park et al. reported a geometric mean of 79.0 EU/mg, with a range from 2.0-713.0 EU/mg on family room floors in houses located in Boston, USA [24]. Von Mutius et al., in a study comparing farming and non-farming families, observed in rural areas in Germany, values of 143.0 EU/mg from farmers versus 39.0 EU/mg from non-farming families in kitchen floor dusts [30]. If expressed in EU/m², our endotoxin levels varied from 304.0-21,864.0 with 6,079.0 EU/m² as median value. Living room floor dusts showed a value of 1,569.0 EU/m² in houses located in Amsterdam and a range from $160.0-2,670,001.0 \text{ EU/m}^2$ from German houses [5, 12]. Geometric mean of endotoxin levels reported by Wickens et al. on carpets in New Zealand homes, were 30,544.0 EU/m² [31]. However, the exact threshold for adverse health effects due to exposure to endotoxin is not exactly known.

CONCLUSION

The present study has demonstrated that office workers are exposed significantly to microorganisms and endotoxin, suggesting an occupational hazard. The main source of Gram-negative bacteria is the settled dust. Future studies are necessary to relate both the bacterial contamination and the endotoxin levels in offices with the risk to develop symptoms and diseases.

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