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EXPOSURE TO BIOAEROSOLS IN A MUNICIPAL SEWAGE TREATMENT PLANT

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Abstract: Microbiological air sampling was performed in a medium-size sewage treatment plant processing municipal wastewater from a city located in eastern Poland. Air samples for determination of the concentrations of viable mesophilic bacteria, Gram-negative bacteria, thermophilic actinomycetes, fungi and endotoxin were collected at 12 sites associated with various phases of sewage treatment process. The concentrations of total mesophilic bacteria (both Gram-positive and Gram-negative) were within a range of 2.4-70.7 × 10^2 cfu/m^3. Gram-positive coryneform bacteria and cocci were dominant, forming respectively 56.6% and 24.0% of the total count. The concentrations of Gram-negative bacteria, thermophilic actinomycetes, and fungi were respectively within ranges of 0.2-5.7 × 10^2 cfu/m^3, 0-0.5 × 10^2 cfu/m^3, and 0.24-1.4 × 10^2 cfu/m^3. Among Gram-negative bacteria, commonly occurred *Enterobacter cloacae* (17.3% of the total count), followed by *Acinetobacter calcoaceticus* (16.2%), *Pseudomonas* spp. (14.0%) and *Stenotrophomonas maltophilia* (11.1%). Among thermophilic actinomycetes prevailed *Thermoactinomyces thalpophilus* (47.2%) and *Thermoactinomyces vulgaris* (22.2%), while among fungi, *Geotrichum candidum* (32.2%), *Penicillium* spp. (20%), *Cladosporium lignicola* (12.2%), and *Alternaria alternata* (10.4%). Altogether, 20 potentially pathogenic species or genera of bacteria and fungi were identified in the air samples taken in the examined plant. The values of the respirable fraction of airborne microflora varied within a fairly wide range and were between 24.1-100%. The concentrations of airborne endotoxin were in the range of 0.104-5.2 ng/m^3. In conclusion, the concentrations of microorganisms and endotoxin in the examined municipal sewage treatment plant were low and did not exceed proposed occupational exposure limit values. A moderate risk for the workers may be associated with the presence of potentially pathogenic microbial species having allergenic and/or immunotoxic properties.

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Key words: sewage treatment, occupational exposure, bioaerosols, bacteria, fungi, endotoxin.

INTRODUCTION

The sewer workers are exposed to droplet aerosols that may contain a large variety of infectious, immunotoxic and allergenic biohazards [7, 43, 50, 56]. The infectious agents include viruses (hepatitis A virus, polioviruses, coxsackieviruses, echoviruses, rotaviruses, adenoviruses, Norwalk virus), bacteria (*Salmonella* spp., *Shigella* spp., *Campylobacter jejuni*, *Yersinia enterocolitica*, *Legionella pneumophila*, *Helicobacter pylori*, *Listeria monocytogenes*, *Mycobacterium xenopi*), parasitic protozoa (*Giardia lamblia*, *Entamoeba histolytica*) and helminths [4, 7, 8, 13, 26, 55].

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A significant risk is also posed by microbial allergens and toxins, of which endotoxin, a lipopolysaccharide produced by Gram-negative bacteria is considered as a most important hazard [32, 38, 40, 41, 43, 50, 57]. Accoding to Rylander [50], endotoxin could be a cause of airways and intestinal inflammation and work-related symptoms (diarrhoea, fatigue, nose irritation) in sewer workers. A causal relationship between exposure to non-infectious airborne biohazards (endotoxin, (1→3)-β-D glucans, bacteria, fungi) and occurrence of gastrointestinal symptoms, fever, respiratory symptoms, skin disorders, eye irritation, headache, fatigue and nausea in the workers of sewage treatment plants has also been considered by other authors [4, 38, 40, 41, 56, 57]. Melbostad *et al.* [41] demonstrated a significant relationship between exposure to rod-shaped bacteria and the occurrence of fatigue and headache in sewage treatment workers.

It has been demonstrated that the workers of sewage treatment plants could be exposed to changeable quantities of airborne bacteria, fungi and endotoxin which may vary depending upon type and capacity of the facility, performed activities and weather conditions [7, 32, 33, 51, 56, 57, 61]. The aim of the present work was to determine the levels of microorganisms and endotoxin in the air of a medium-size municipal sewage treatment plant and to study the species composition of airborne microflora. The examined plant was a modern, open air facility, typical for many Polish towns.

MATERIALS AND METHODS

Examined facility. Air sampling was performed in a medium-size municipal sewage treatment plant that collects wastewater from a city located in eastern Poland, inhabited by 160,000 people. The facility has a capacity of 36,000-45,000 m^3 per day and a total of 55 workers is employed in the plant.

Influent sewage is cleared on grids in a grid basin and then by separation of sand in a sanding basin. Cleared raw sewage is pumped to primary sedimentation basins and then distributed to 3 aeration basins. Thereafter, aerated sewage is pumped to secondary sedimentation basins and finally refined wastewater is discharged into a river while deposited sludge sediment is dewatered by using a belt press and disposed for composting and agricultural use. Most of the processes take place outdoors, except for grid clearing and sludge dewatering.

Air samples for microorganisms and endotoxin were collected at 9 sites in the sewage treatment plant throughout the whole production process, and at 3 sites beyond the area of this plant. The outside sampling sites were located in the centre of the city (2–3 km from the plant) at the installations forming a part of the municipal sewage collection system: two local pump stations and an inlet to a sewer duct. Air sampling was conducted in May 1999 at the air temperature 15–16ºC. The samples were taken upwind, 145 cm above the ground, close to the sewage treatment installations (0.5–1.0 m).

Altogether, air samples were taken at the following 12 sites: 1) Grid basin; 2) Sanding basin; 3) Pumping raw sewage into the sedimentation basins; 4) Primary sedimentation basin; 5) Aeration basin “A”; 6) Aeration basin “B”; 7) Aeration basin “C”; 8) Secondary sedimentation basin; 9) Sludge dewatering area; 10) Pump station “A” (city); 11) Pump station “B” (city); 12) Inlet to a sewer duct (city).

Table 1. Microorganisms in the air of a sewage treatment plant: concentrations and respirable fractions (Rf).

Sampling site (Number, name)	Total mesophilic bacteria (Blood agar)		Gram-negative bacteria (EMB agar)		Thermophilic actinomycetes (Tryptic soya agar)		Fungi (Malt agar)		Total microorganisms	
	Concentration (mean ± SD, cfu/m^3)	Rf (%)	Concentration (mean ± SD, cfu/m^3)	Rf (%)	Concentration (mean ± SD, cfu/m^3)	Rf (%)	Concentration (mean ± SD, cfu/m^3)	Rf (%)	Concentration (mean ± SD, cfu/m^3)	Rf (%)
1. Grid basin	2350 ± 1630	23.4	190 ± 241	36.0	40 ± 55	25.0	20 ± 45	100	2410 ± 1632	24.1
2. Sanding basin	7070 ± 3897	37.3	540 ± 319	37.0	10 ± 22	100	50 ± 70	100	7130 ± 3950	37.8
3. Pumping raw sewage	1870 ± 767	94.6	190 ± 160	57.9	20 ± 45	100	20 ± 45	100	1910 ± 819	94.7
4. Primary sedimentation basin	1730 ± 1272	100	570 ± 334	40.4	20 ± 45	0	30 ± 67	66.7	1780 ± 1226	98.3
5. Aeration basin “A”	1460 ± 769	48.6	100 ± 100	100	0	0	10 ± 22	0	1470 ± 761	48.3
6. Aeration basin “B”	2180 ± 2296	94.9	120 ± 130	83.3	0	0	150 ± 141	40.0	2330 ± 2269	91.4
7. Aeration basin “C”	1340 ± 605	82.8	70 ± 45	85.7	20 ± 45	100	50 ± 50	40.0	1410 ± 590	81.3
8. Secondary sedimentation basin	610 ± 275	100	20 ± 45	100	20 ± 45	100	10 ± 22	100	640 ± 270	100
9. Sludge dewatering area	770 ± 342	83.1	60 ± 55	33.3	50 ± 86	80.0	80 ± 83	100	900 ± 332	84.4
10. Pump station “A” (city)	240 ± 277	100	130 ± 67	23.1	40 ± 89	100	70 ± 45	100	350 ± 320	100
11. Pump station “B” (city)	500 ± 291	28.0	70 ± 97	71.4	20 ± 45	100	140 ± 108	57.1	660 ± 319	36.4
12. Inlet to a sewer duct (city)	3340 ± 1011	43.4	110 ± 124	100	10 ± 22	100	70 ± 45	71.4	3420 ± 1053	44.1
Mean	1955 ± 2243	58.2	181 ± 235	50.6	21 ± 47	75.4	58 ± 78	68.9	2034 ± 2241	58.7
Median	1200		100		0		100		1300	

Table 2. List of microbial species and genera identified in the samples of air from a sewage treatment plant.

Gram-negative bacteria: ***Acinetobacter calcoaceticus****+ (1-4, 6, 11, 12), *Aeromonas* spp. (6), *Chryseomonas luteola* (5, 6), ***Citrobacter freundii***+ (2-4), ***Citrobacter*** spp.+ (6, 7), ***Enterobacter cloacae***+ (1-3, 5, 10, 12), ***Escherichia coli***+ (2, 4), ***Hafnia alvei*** + (4), ***Klebsiella*** spp.+ (2), *Pseudomonas* spp. (1, 2, 4, 5, 8, 9, 11), *Serratia* spp. (4), *Stenotrophomonas maltophilia* (2, 4, 5, 11).

Bacilli: *Bacillus cereus* (1-5, 12), *Bacillus megaterium* (1-12), ***Bacillus subtilis**** (1-12), *Bacillus* spp. (1-12).

Corynebacteria: ***Arthrobacter globiformis**** (1-12), *Arthrobacter* spp. (1-12), ***Brevibacterium linens**** (1-12), *Brevibacterium helvolum* (1), *Corynebacterium xerosis* (2, 12), *Corynebacterium* spp. (1-12).

Gram-positive cocci: *Micrococcus luteus* (1-12), *Micrococcus roseus* (1-5, 7-10, 12), *Staphylococcus epidermidis* (1-12), *Staphylococcus saprophyticus* (1-12), *Staphylococcus* spp. (1-12), *Streptococcus* spp. (10).

Mesophilic actinomycetes: ***Streptomyces albus**** (2, 4, 6-8).

Thermophilic actinomycetes: ***Saccharomonospora viridis**** (2), ***Saccharopolyspora rectivirgula**** (3, 7, 11), ***Thermoactinomyces thalpophilus**** (1, 4, 8, 9, 12), ***Thermoactinomyces vulgaris**** (1, 3, 10, 12), ***Thermomonospora fusca**** (2, 9, 10).

Fungi: ***Alternaria alternata****+ (3, 4, 6, 7, 11), *Aspergillus chevalieri* (4), ***Candida* spp.*** (1, 2), *Citromyces* spp. (1, 3, 6, 8), *Cladosporium lignicola* (6, 9-12), *Geotrichum candidum* (2, 6, 7, 9-12), *Heterocephalum aurantiacum* (8), *Monilia sitophila* (10), *Monosporium silvaticum* (1, 11), ***Mucor racemosus**** (2), *Oidiodendron truncatum* (3, 5), *Papularia sphaerospermum* (1), ***Penicillium* spp.***+ (1, 2, 6, 7, 9-12), *Prophytroma tubularis* (8).

Sites of isolation are given in parentheses. Names of the species reported as having allergenic and/or immunotoxic properties (see text) are in bold and marked as follows: * allergenic species; + immunotoxic species.

Microbiological examination of the air. Air samples were taken in the sewage treatment plant with a custom-designed particle-sizing slit sampler [11] which enabled estimations of both total and respirable fractions of the microbial aerosol (Polish Patent 87612 assigned on 6 June 1977). Each air sample was in duplicate, taken at a flow rate of 20 l/min. It consisted of two parallelly exposed agar plates: one, "a" sampled directly for all organisms and used for the estimation of the total concentration of cfu per m^3; and the other, "b" sampled through a pre-selector (consisting of a system of glass tubes and regulated deposition disks covered with sticky substance) for the respirable fraction. The value of respirable fraction was expressed as a percent (%) of the total count, calculated by division of the number(s) of cfu on plate(s) "b" through the number(s) of cfu on plate(s) "a" and multiplication by 100. The median cut-off point for the respirable fraction was 3.0 µm, approximating the recommendations of the American Conference of Governmental Industrial Hygienists [58].

At each sampling site, a series of 5 double samples were taken on each of the following agar media: blood agar for total mesophilic Gram-negative and Gram-positive bacteria, eosin methylene blue (EMB) agar for Gram-negative bacteria, half-strength tryptic soya agar for thermophilic actinomycetes, and malt agar for fungi. The blood agar plates were subsequently incubated for 1 day at 37ºC, then 3 days at 22ºC and finally 3 days at 4ºC. The malt agar plates were subsequently incubated for 4 days at 30ºC and 4 days at 22ºC [12]. The prolonged incubation at lower temperatures aimed to isolate as wide a spectrum of bacteria and fungi as possible. The EMB agar plates were incubated in the same way as the blood agar plates and the tryptic soya agar plates were incubated for 5 days at 55ºC. The grown colonies were counted and differentiated and the data reported as cfu per 1 cubic meter of air (cfu/m^3). The total concentration of micro-organisms in the air was obtained by the addition of the concentrations of total mesophilic bacteria (grown on blood agar medium), thermophilic actinomycetes and fungi.

Bacterial isolates were identified with microscopic and biochemical methods, as recommended by Bergey's Manuals [22, 28, 54, 60] and Cowan & Steel [5]. Additionally, the selected isolates were identified with microtests, using API Systems 20E and NE (bioMérieux, Marcy l'Etoile, France). Fungi were classified with microscopic methods, according to Barron [2], Larone [35], Litvinov [37], Ramirez [46], and Raper & Fennell [47].

For determination of the endotoxin concentrations, the air samples were collected on the polyvinyl chloride filters by use of an AS-50 one-stage sampler (TWOMET, Zgierz, Poland), at the flow rate of 50 l/min. Two samples were taken at each sampling site. The concentration of bacterial endotoxin in the air was determined by the *Limulus* amebocyte lysate gel tube test (LAL) [36]. The filters were extracted for 1 hour in 10 ml of pyrogen-free water at room temperature, heated to 100ºC in a Koch apparatus for 15 min (for better dissolving of endotoxin and inactivation of interfering substances), and after cooling, serial dilutions were prepared. The 0.1 ml dilutions were mixed equally with the "Pyrotell" *Limulus* reagent (Associations of Cape Code, Palmouth, MA, USA). The test was incubated for 1 hour in a water bath at 37ºC, using pyrogen-free water as a negative control and the standard lipopolysaccharide (endotoxin) of *Escherichia coli* 0113:H10 as positive control. The formation of a stable clot was regarded as a positive result. The results were reported as nanograms of the equivalents of the *E. coli* 0113:H10 endotoxin per 1 m^3 of air. To convert to Endotoxin Units (EU), the value in nanograms was multiplied by 10.

Statistical analysis. The results were analysed by chi-square test for assessment of differences between the sampling sites, and by Spearman rank order test for assessment of correlation between bioaerosol components, using STATISTICA for Windows v. 5.0 package (Statsoft©, Inc., Tulsa, Oklahoma. USA).

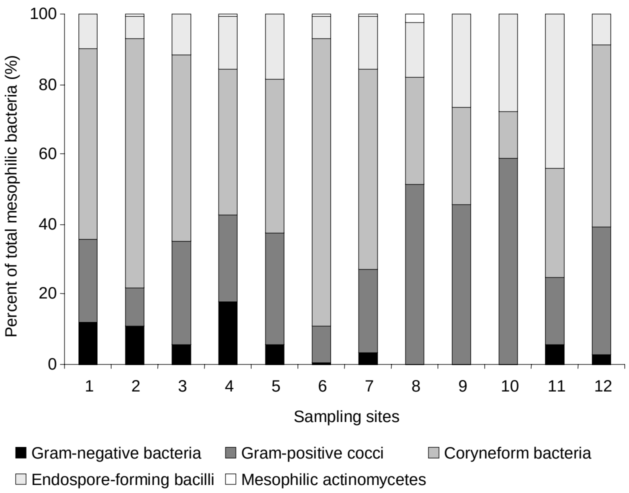

Figure 1. Composition of mesophilic bacteria isolated from the air of a sewage treatment plant (total count).

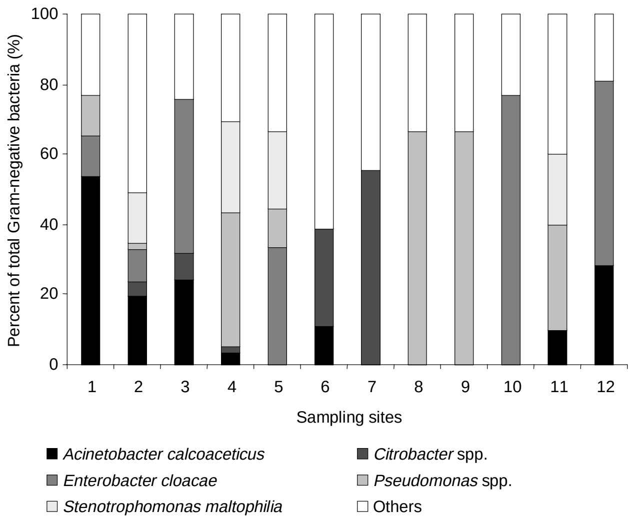

Figure 2. Composition of Gram-negative bacteria isolated from the air of a sewage treatment plant (total count).

RESULTS

The concentrations of total mesophilic bacteria (both Gram-positive and Gram-negative), which formed a vast majority of airborne microflora (over 95%), were within a range of 240–7,070 ($2.4–70.7 \times 10^2$) cfu/m^3 (median 1,200 cfu/m^3). The concentrations of Gram-negative bacteria, thermophilic actinomycetes, and fungi were respectively within ranges of $0.2–5.7 \times 10^2$ cfu/m^3, $0–0.5 \times 10^2$ cfu/m^3, and $0.24–1.4 \times 10^2$ cfu/m^3 (Tab. 1). No significant correlation could be found between the concentrations of mesophilic bacteria, thermophilic actinomycetes and fungi ($p > 0.05$).

The quantities of airborne microorganisms showed a significant variation depending on the sampling site ($p < 0.000001$). The concentrations of total microorganisms noted at the initial and intermediate phases of sewage treatment (clearing, primary sedimentation, aeration) were on the average 2–3 times higher compared to those found at the final phases (secondary sedimentation, sludge dewatering) (Tab. 1).

Table 3. Concentration of bacterial endotoxin in the air of a sewage treatment plant.

Sampling site (number, name)	Concentration of endotoxin (ng/m^3)
1. Grid basin	5.02
2. Sanding basin	0.21
3. Pumping raw sewage	5.20
4. Primary sedimentation basin	0.21
5. Aeration basin “A”	5.20
6. Aeration basin “B”	2.10
7. Aeration basin “C”	1.04
8. Secondary sedimentation basin	1.04
9. Sludge dewatering area	2.10
10. Pump station “A” (city)	0.21
11. Pump station “B” (city)	0.104
12. Inlet to a sewer duct (city)	1.6
Mean ± S.D.	2.00 ± 2.02
Median	1.32

The values of the respirable fraction of airborne microflora varied within a fairly wide range and were between 24.1–100% (on average 58.7%) (Tab. 1).

Gram-positive bacteria distinctly prevailed among the airborne microorganisms in the examined sewage treatment plant. The most numerous were corynebacteria (irregular Gram-positive rods of the genera *Corynebacterium*, *Arthrobacter*, *Brevibacterium*) which dominated at 8 sites (Fig. 1) and formed on average 56.6% of the total count of mesophilic bacteria. Gram-positive cocci (*Staphylococcus*, *Micrococcus*) and endospore-forming bacilli (*Bacillus*) were respectively dominant at 3 and 1 sites, forming on average 24.0% and 11.7% of the total count of mesophilic bacteria. Only less than 1% of the total mesophilic bacteria recovered on blood agar showed hemolytic properties.

Four taxa of Gram-negative bacteria constituted over a half of the strains isolated from the air of a sewage treatment plant. They were: *Enterobacter cloaceae* (17.3% of the total count), *Acinetobacter calcoaceticus* (16.2%), *Pseudomonas* spp. (14.0%), and *Stenotrophomonas maltophilia* (11.1%) (Fig. 2). Altogether, non-enterobacteria outnumbered enterobacteria (60.5% *versus* 39.5% of the total isolates).

Thermoactinomyces thalpophilus strains constituted nearly a half of the isolated thermophilic actinomycetes (47.2%), followed by *Thermoactinomyces vulgaris* (22.2%), *Saccharopolyspora rectivirgula* (13.9%) and *Thermomonospora fusca* (13.9%) (Fig. 3).

Among fungi, 4 species or genera formed nearly 75% of the total count. They were: *Geotrichum candidum* (32.2%), *Penicillium* spp. (20.0%), *Cladosporium lignicola* (12.2%), and *Alternaria alternata* (10.4%) (Fig. 4).

In the air samples taken in the examined facility, altogether 34 species or genera of bacteria and 14 species

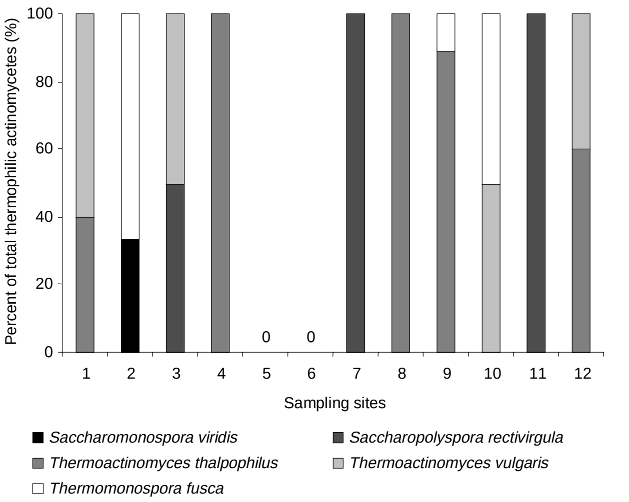

Figure 3. Composition of thermophilic actinomycetes isolated from the air of a sewage treatment plant (total count).

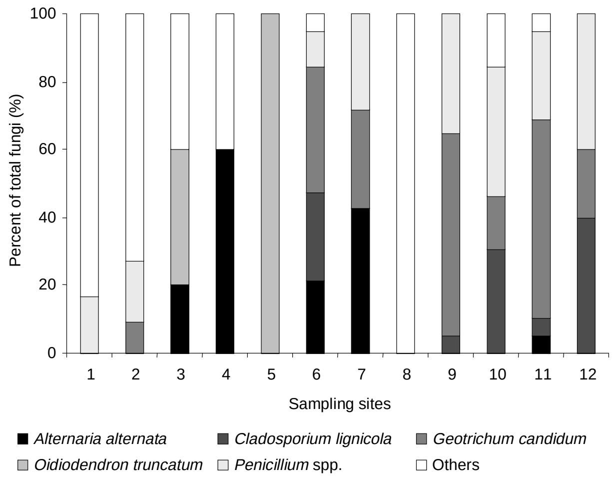

Figure 4. Composition of fungi isolated from the air of a sewage treatment plant (total count).

or genera of fungi were identified, of these, 16 and 4 species or genera respectively were reported as having allergenic and/or immunotoxic properties [13, 17, 23, 24, 29, 30, 42] (Tab. 2). These figures are certainly an underestimation, as a part of bacterial and fungal strains could be identified only to the generic level.

The concentrations of airborne endotoxin in the examined plant were low, within the range of 0.104–5.2 ng/m^3. The endotoxin levels varied depending on the sampling site, but to a lesser extent than microorganisms ($0.01 < p < 0.05$). No significant correlation could be found between the concentrations of viable microorganisms and endotoxin ($p > 0.05$).

DISCUSSION

The levels of total microorganisms and endotoxin found in the air of the examined sewage treatment plant were respectively of the order 10^2–10^3 cfu/m^3 and 10^{-1}-10^0 ng/m^3. They were higher compared to Polish dwellings and offices [19] but distinctly lower compared to other facilities contaminated with bioaerosols [6, 14, 15, 16, 18]. For example, the highest concentrations of airborne microorganisms recorded in the examined sewer plant were respectively 88 and 13 times lower compared to those found in herb- and potato processing plants [15, 16] while the highest concentrations of airborne endotoxin were respectively 516,000 and 364,000 times lower [15, 16]. The Occupational Exposure Limit (OEL) values proposed by various authors for airborne microorganisms [13, 39], mesophilic bacteria [19], Gram-negative bacteria [3, 13, 19, 39], thermophilic actinomycetes [13, 19] and fungi [13, 19] were not exceeded anywhere. At no site did the concentrations of airborne endotoxin exceed the OEL value of 0.1 µg/m^3 proposed by Clark [3], Rylander [49] and Malmros *et al.* [39] and the OEL value of 25 ng/m^3 suggested by Laitinen *et al.* [34]. At 3 out of 12 sampling sites the concentrations of endotoxin in the air bordered the restrictive OEL value of 5 ng/m^3 (50 EU/m^3), proposed by Dutch Expert Committee on Occupational Standards (DECOS) [10].

The concentrations of microorganisms found in the air of the examined sewage treatment plant were similar to the values of the order 10^1–10^3 cfu/m^3 reported by Kocwa-Haluch [25, 26] and Wlazło *et al.* [61] from Polish plants and by Haas *et al.* [20] from Austrian plants, but lower compared to the values of the order 10^3–10^5 cfu/m^3 reported by Wanner [59] and Schira *et al.* [51] from Swiss plants. Concentrations of airborne endotoxin were similar to the values of the order 10^{-1}-10^0 ng/m^3 reported by Douwes *et al.* [9] from the Netherlands and to the values 0–10^1 ng/m^3 reported by Schira *et al.* [51] from Switzerland. They were slightly lower compared to those reported recently by Krajewski *et al.* [27] who found in a large Polish sewage treatment plant a geometric mean concentration of endotoxin equal to 20.3 ng/m^3, and a mean concentration of (1→3)-β-D glucans equal to 7.76 ng/m^3. On average, the levels of airborne microorganisms and endotoxin found in the present work are lower compared to the values recorded in Scandinavian sewage treatment plants by Andersson *et al.* [1], Mattsby and Rylander [40], Rylander *et al.* [48, 50], Lundholm and Rylander [38], Melbostad *et al.* [41], and Laitinen *et al.* [31, 32, 33]. The quoted authors reported the concentrations of microorganisms of the order 10^1–10^7 cfu/m^3 [1, 31, 32, 33, 38, 40, 48] and the concentrations of endotoxin of the order 10^{-2}–10^4 ng/m^3 [31, 32, 33, 38, 41, 48, 50]. The differences may be due mainly to the technology and kind of sewage processed. While in Poland most of treatment process is performed outdoors, in Scandinavia indoor sewage treatment is more common and is associated with greater exposure to bioaerosols [32, 57]. Laitinen *et al.* [32, 33] found high levels of airborne microorganisms in plants treating nutrient-rich wastewater from the food industry while in those treating municipal sewage microbial levels proved to be lower by order of magnitude

[33]. Processing of heat-dried sewage sludge, as in the plant examined by Mattsby and Rylander [40], favours the dispersal of bioaerosols. It seems that the use of medium-size open air sewage treatment plants producing sludge by belt press decreases the risk of exposure to biohazards, though such risk could not be ruled out.

The majority of bioaerosol particles in the examined plant were within the respirable fraction (aerodynamic diameter of particles equal to or less than 3 µm), which increases the risk of penetration into alveoli of exposed workers. This result is in accordance with those obtained by Laitinen *et al.* [32] and Wlazło *et al.* [61]. Laitinen *et al.* [31] found a significant correlation between the concentrations of bacteria and endotoxin in the sewage treatment plant which was not confirmed by Melbostad *et al.* [41]. This relationship was also not found in the present work, which could be partly due to the relatively small number of samples collected and partly to much lower durability of Gram-negative bacteria in the air compared to endotoxin [21, 25, 49].

The airborne microflora of the examined sewage treatment plant was clearly dominated by Gram-positive bacteria, mostly by corynebacteria, which made up over half of all microbial isolates at the majority of sampling sites. These bacteria are commonly associated with organic dusts [42] and occupational bioaerosols [12, 13, 14] but so far little is known about their pathogenic properties. Cases of allergic alveolitis caused by *Arthrobacter globiformis* and *Brevibacterium linens* have been reported [42] and the involvement of peptidoglycan produced by these bacteria in causing organic dust toxic syndrome (ODTS) cannot be ruled out.

The species composition of Gram-negative bacteria isolated from the air of the examined plant was similar to that reported by other authors [32, 33, 38, 51, 61]. A number of the isolated enterobacterial species (*Enterobacter cloacae*, *Escherichia coli* and others) are known endotoxin producers. These bacteria are recognized indicators of faecal contamination [25] and their presence in the air proves the risk of exposure to faecal particles by the workers of the examined plant. Of non-enterobacterial isolates, *Acinetobacter calcoaceticus* was proved to possess allergenic and endotoxic properties [52, 53] and the presence of *Aeromonas* strains might pose a health risk, as *Aeromonas hydrophila* was suggested as a producer of protein enterotoxin causing gastrointestinal symptoms in sewage workers [48]. No potentially infectious strains of *Salmonella*, *Shigella*, and *Yersinia* were isolated. Although in the earlier studies by Prażmo [44] and Prażmo *et al.* [45] numerous *Salmonella* strains belonging to 22 serotypes were isolated from the samples of Polish sewage and sewage sludge, the probability of *Salmonella* isolation from air samples is small because of its low concentration in sewage and rapid death during aerosolization [21, 25].

A small proportion of hemolytic bacteria in the air of the examined plant decreases potential hazard, as these bacteria were proposed by Kocwa-Haluch [25] as an indicator of health risk for workers of sewage treatment plants and residents of neighbouring areas.

The presence of 5 species of thermophilic actinomycetes known as causative agents of allergic alveolitis (farmer's lung, mushroom worker's lung) [29, 30] poses a potential risk of pulmonary allergy for the workers of the examined plant in spite of a small concentration of these organisms in the atmosphere. Until recently, a potential risk of thermophilic actinomycetes in sewage has been considered in relatively few papers [20].

Among airborne fungi in the examined sewage treatment plant the greatest risk of evoking allergic disease pose *Penicillium* spp. and *Alternaria alternata*. A potential risk to fungi was diminished by their very low concentration and the absence of pathogenic *Aspergillus* strains, in particular *Aspergillus fumigatus*, which is regarded as a notable risk factor for sewage and compost workers [4].

Generally, the obtained results confirm the view on the variability of exposure to bioaerosols among sewer workers and seem to indicate that in the modern, open air sewage treatment plants in Central Europe the risk may be relatively low. This does not rule out a much greater exposure at different climatic and technological conditions and does not change an opinion expressed by many authors [7, 43, 50, 56] that the workers of sewage treatment plants are among occupational groups seriously endangered by biohazards.

CONCLUSION

The concentrations of microorganisms and endotoxin in the examined municipal, open air sewage treatment plant were low and did not exceed proposed occupational exposure limit values. A moderate risk for the workers may be associated with the presence of potentially pathogenic microbial species having allergenic and/or immunotoxic properties.

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