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Letter to Editor

Bacterial Aerosol in Silesian Hospitals: Preliminary Results

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Abstract

The concentration levels of airborne bacteria were measured in clinical/hospital rooms in Upper Silesia, Poland, in buildings of varying conditions. It was found that the typical level of bacterial aerosol concentration is about 10^3 CFUm^{-3} in clinical outpatient rooms and ranges from 10^2 CFUm^{-3} to 10^3 CFUm^{-3} in hospitals, depending on the number of occupants and physical quality of the building. The increased level of the airborne bacteria in patient rooms resulting from bed-making was noticed. The *Staphylococcus/Micrococcus* group was a dominating part of the bacteria in studied hospitals/clinic air, contributing together 58-78% of the total bacteria concentration, confirming that detected airborne bacteria mainly originated from human organisms. The size distributions of bacterial aerosol in naturally ventilated rooms have peaks in the size range between 1.1 and 3.3 μm while in the mechanically ventilated hospital rooms with HVAC the peak appears in the diameter range from 3.3 μm to 4.7 μm.

Keywords: bioaerosol, bacteria, indoor air, hospital environment

Introduction

Certain human pathogens seem to be significant causes of infection from indoor air, infecting otherwise healthy individuals [1,2]. Exposure to bioaerosols may be especially hazardous in clinics and hospitals where they may be a major factor in increasing morbidity from respiratory diseases. Some bacteria such as *Streptococcus pyogenes, Neisseria meningitidis, Corynebacterium diphtheriae* and *Mycobacterium tuberculosis* are known to be transmitted predominantly by airborne droplets from infected people, and they may cause nosocomial infection [3]. It is interesting that many opportunistic bacteria found in the indoor environment pose a potential threat only to immunocompromised patients in hospitals.

In the absence of infection, exposure to endotoxic or immunogenic substances (derived from non-viable bacteria remnants) can lead to pulmonary irritation and allergic reactions associated with breathing contaminated air in buildings [4, 5]. This might be related to the sick building phenomenon. The limited introduction of "fresh" air and constant re-circulation create a fertile breeding ground for bacteria, once air-conditioning systems, humidifiers or water towers become contaminated [2].

Generally, there are various sources of airborne bacteria and fungi including dust introduced into the hospital

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and, resuspended in the air [3]. However, in many hospital buildings interchange between outdoor and indoor air is limited and the primary source of interchange is through the filtration system of the air-conditioner. Simmons et al. [6] has described the fungal colonization of air filter media used in HVAC systems of seven hospitals in the eastern United States. They reported the colonization of filter media by fungi more commonly associated with the incursion of outdoor air. The results obtained recently by Maus et al. [7] showed that bacterial and mold spores collected in air filter media are able to survive over a prolonged period of time and thus pose the potential of microbial growth. In the latter case abundant spore production and their release into the clean air stream of the filter is likely. On the other hand, Favero et al. have demonstrated that in hospital operating rooms bioaerosol contaminants were mainly associated with emissions from human hair, skin and respiratory tract [8]. Also, other published results of the identification of bacteria species indicate that bacteria in indoor air are mainly Gram-positive cocci which arise from occupants [9, 10]. However, although Gram-negative bacteria are not usually abundant, occasionally *Acinetobacter, Aeromonas, Flavobacterium* or, especially, *Pseudomonas* species may be prominent [10]. Their presence usually indicates growth in the sites where there is an abundant source of water. Therefore, it is necessary to assess the composition and concentration of airborne microorganisms in clinics and hospital buildings. In particular, bioaerosol monitoring in hospitals can provide information for epidemiological investigation of nosocomial infectious diseases, research into airborne microorganism spread and control, monitoring biohazard procedures, and can be used as a quality control measure.

In this study it was our intention to evaluate the level of airborne bacteria in a confined environment in some hospitals in Upper Silesia, Poland, characterized by the presence of subjects at risk, those in the internal medicine, alergology, cardio-surgery departments, and by a heavily populated outpatient clinic, as well as in the office room located in the hospital area. The physical quality of the studied indoor environments was significantly differentiated. One hospital is located in a 100-year-old, non-modernized building containing crowded patient rooms without any ventilation system (the study was carried out before the renovation), while other hospitals are very modern.

Methods

Measurements were carried out in 2001-02 in 3 hospitals and one medical clinic located in Upper Silesia, Poland, in the following towns: Katowice, Zabrze and Sosnowiec. The basic environmental parameters of the studied hospital/clinic rooms are briefly described in Table 1.

Samples were taken using the 6-stage Andersen impactor (with aerodynamic cut-size diameters of 7.0, 4.7, 3.3, 2.1, 1.1 and 0.65 μm) located, as a rule, in the center of the studied rooms at a height of 1.0-1.5 m above the floor. Unfortunately, the number of collected samples was rather low: between 2 and 4 in every studied room. Therefore, our results can be treated as preliminary data only.

Sampling time, following Nevalainen et al. [11,12], was 5-10 minutes. Microorganisms were collected on

Table 1. Hospital/clinic room characteristics.

Micro-Environment	Physical characteristics of the building	Number of persons in the room	Temp. [°C]	Humidity [%]
1. Office room, the hospital of occupational diseases, Sosnowiec	30-year-old building in very good condition. Natural ventilation only, 33 m^3 volume.	2	23-27	54-75
2. Outpatient room, clinic of occupational diseases, Sosnowiec	30-year-old building in very good condition. Natural ventilation only, 77 m^3 volume.	7-16	19-23	58-75
3. Patient room in hospital nr 5 in Sosnowiec, Department of Internal Medicine	New, modern hospital. Small patient rooms, mechanically ventilated, 30 m^3 volume.	3-5	24-25	65
4. Patient room in the hospital in Zabrze, Department of Internal Medicine & Allergology	100-year-old building in bad condition. Crowded patient rooms, naturally ventilated only. Windows are frequently open, 110 m^3 volume.	8	24	80
5. Operating room in the Cardio-Surgery Department, Hospital in Katowice-Ochojec	New, modern hospital. Operating rooms are located in restricted, sterilized area; mechanically ventilated (HVAC), 144 m^3 volume. (Measurement were made after a cardiological operation).	(1-3)*	26	70

*during sampling of airborne bacteria, i. e. after operation

trypcase soy agar -TSA (with cycloheximide added to inhibit fungal growth) in Petri-dishes located on all impactor stages.

The selection of the appropriate incubating temperature should be dependent on the main purpose of the microbiological study. Although TSA plates are often incubated at 37°C for 48 hours, incubation at lower temperatures may recover a greater number of species and give improved resuscitation of stressed bacteria [13]. Hence, according to the Report No. 12 of the European Collaborative Action [14], all our samples were incubated for 7 days at 22°C. Concentrations were calculated as colony forming units per cubic meter of air ($CFUm^{-3}$) using positive-hole correction. During sampling, the temperature and relative humidity of indoor air were recorded.

The bacterial aerosol samples were identified according to Gram staining and morphology. Next, visible colonies were subcultured onto either Chapman agar or onto MacConkey agar. Gram-positive and Gram-negative bacteria were finally identified by the biochemical API test.

Results and Discussion

Table 2 shows the mean concentration of airborne bacteria in the studied rooms. It can be seen that bacterial aerosol mean concentration was about 10^3 $CFUm^{-3}$ in the clinical outpatient room in Sosnowiec and varied from 10^2 $CFUm^{-3}$ to 10^3 $CFUm^{-3}$ in the hospitals. Generally, our results agree well with the data obtained by others. For example, Halcatova et al. [15] found in the operating theatre and intensive-care unit of the hospital in Prague, Czech Republic, where the average concentration of bacterial aerosol was about 10^2 $CFUm^{-3}$. In other European hospitals airborne bacteria levels usually have been found from less than 10 to 10^2 $CFUm^{-3}$ [16,17]. Similar data have been published by Brenniman and Allen [18] for a hospital in Illinois, USA. Li and Hou have reported that in the hospital operating rooms in Taipei, Taiwan, the concentration of airborne bacteria also varied from 10 to 10^2 $CFUm^{-3}$ [19], but in the bone marrow transplantation rooms the concentration was much lower, changing from 0 to 2 $CFUm^{-3}$ only. On the other hand, Yousefi and Rama [20] found in Johannesburg, South Africa, in the hospital located close to industry and residential area, that concentrations of bacterial aerosol ranged from 10^2 to 10^3 $CFUm^{-3}$.

Our values, presented in Table 2, are also comparable with the typical level of bacterial aerosol in Upper Silesia indoors, estimated at about 10^3 $CFUm^{-3}$ in homes and 10^2 $CFUm^{-3}$ in offices [21].

From the analysis of Table 2 it can be seen that the respirable bacterial particles (with the aerodynamic diameter less than 5 μm) varied from about 40% to more than 80% of the total bacteria concentration.

In one patient room in the hospital in Zabrze the measurements of airborne bacteria concentration was carried out immediately after bed-making. It can be seen that after bed-making the concentration level was almost twice higher than in the same room without bed-making. On the other hand, the ratio of respirable bacterial particles to total bacteria decreased from 86% to 48%, indicating that the resuspension phenomenon really occurred. This result agrees well with the previously published data. It is known that bed making generates dust and airborne microorganisms [22]. Increased bacterial counts in the indoor air resulting from bed-making were first noticed in the early 1940s [23]. Recently, Shiomori et al. found that methicillin-resistant *Staphylococcus aureus*,

Table 2. Concentration levels of the airborne bacteria in the studied hospital/clinic rooms.

Sampling location	Bioaerosol concentration [$CFUm^{-3}$]	
	Bacteria total	Ratio of respirable bacteria to total concentration
1. Office room	380 (221)*	0.73 (0.08)
2. Out-patient room, clinic of occupational diseases, Sosnowiec	1155 (406)	0.69 (0.09)
3. Patient room in the hospital in Sosnowiec	168 (0.04)	0.41 (35)
4. Patient room in the hospital in Zabrze - after bed making	820 (106) 1533	0.86 (0.02) 0.48
5. Operating room, Cardio-Surgery Department, Hospital in Katowice-Ochojec - after I operation, during room cleaning - after II operation	 236 (111) 136 (1.3)	 0.70 (017) 0.65 (0.09)

*Standard deviations (in parentheses)

which has frequently been reported as a major hospital-acquired pathogen in community hospitals, may be spread via respirable-sized aerosols generated by bed-making [24].

Further studies should also concern the generation of fungal aerosol by bed-making. Recently, Chew et al. [25] documented that dustborne and airborne fungal propagules represent a different spectrum of fungi. Therefore, it may be essential to collect both air and dust samples as indicators for fungal exposure.

It is well known that the aerodynamic size of a particle determines its behavior in an environment and in the human respiratory tract. Therefore, not only the concentration of bioaerosol is important but also the size distribution of the bioaerosol particles. The particles' size distributions of the collected airborne bacteria are presented in Figure 1. As can be seen, the indoor patterns of size distribution vary, depending mainly on the type of ventilation and number of persons in the room. Trying to explain the size distribution shape of the bacterial aerosol indoors, it should be noted that the coarse particles remain airborne only for a short time and they are removed from the air by gravity and resuspended again when disturbed mechanically. Therefore, ventilation is not a crucial factor in the removal of these particles. However, the behavior of fine particles is different. For example, a 1 μm particle does not settle down during 2 hours [9]. Hence, it can be concluded that the mechanical system of ventilation removes this fraction more efficiently than natural ventilation based on gravity [9]. The analysis of Figure 1 seems to confirm this relation. The size distributions of airborne bacteria in the outpatient clinic, as well as in the hospital in Zabrze, where there is only natural ventilation, have the peaks in the size range between 1.1 and 3.3 μm while in the mechanically ventilated hospital rooms (in Sosnowiec and in Katowice) the peak appears in the size range 3.3-4.7 μm, i. e. is shifted into larger particles. In the office, where only two people were present during the bioaerosol sampling, the size distribution curve was rather flat, without any particularly significant peak.

The detailed analysis of the bacterial genera and species in the studied hospitals is presented in Table 3. Analyzing the concentrations of bacterial aerosol in Table 3, it can be found that the total values are slightly different from those presented in Table 2. These differences appeared because the data in Table 2 are averaged while Table 3 presents the results of the identification analysis made only for the selected samples.

It is known that Gram-positive bacteria are often present in higher concentrations than Gram-negative bacteria in indoor air. However, due to the variety of bacteria present, speciation is complex and rarely exhaustive [26]. Considerable diversity is reported with Gram-positive cocci (including staphylococci and micrococci), pleomorphic organisms (including diphtheroids) and rods (including bacilli), all being common [26-30]. Our

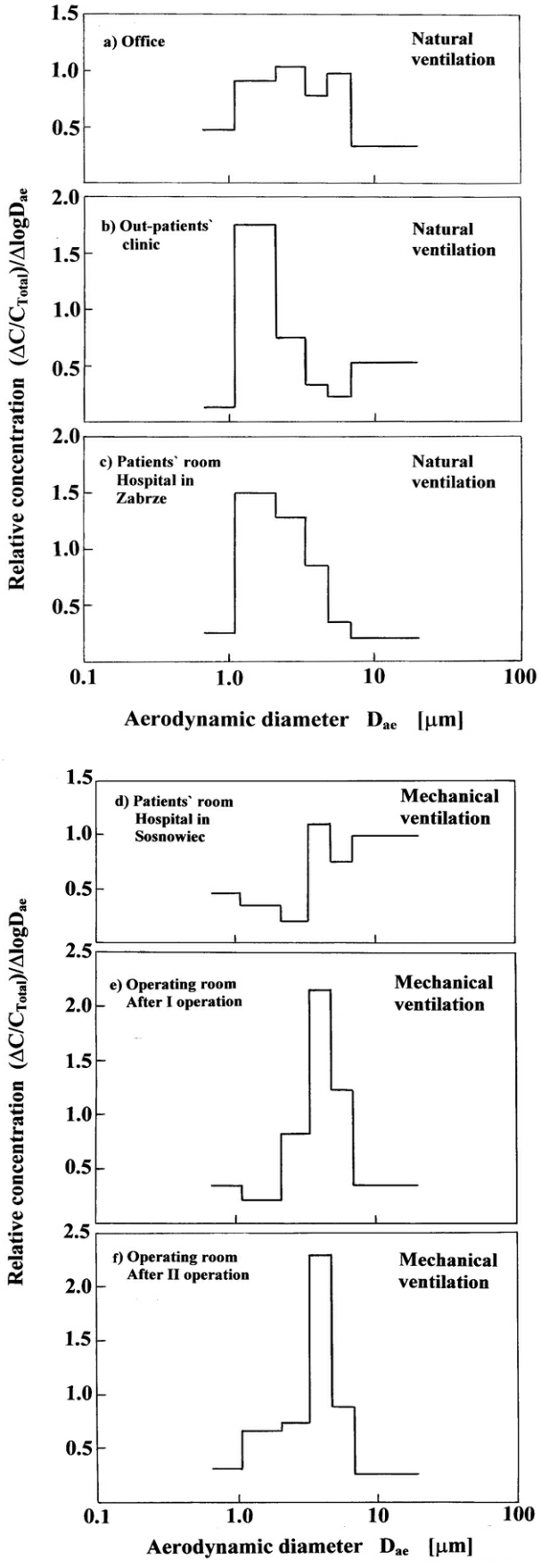

Fig. 1. Size distribution of airborne bacteria in the hospital/clinic rooms.

results, summarized in Table 3, show that the *Staphylococcus/Micrococcus* group was a dominating part of the bacteria in the studied hospital/clinic air. The main source of these bacteria indoors is human organism, especially human skin. It can be seen that *Staphylococcus* had the highest count, constituting 50-71% of the total bacterial genera. The second most common airborne bacteria was *Micrococcus*, but constituting only 1.5-33% of the total. It is interesting that *Corynebacterium* also was present at a high frequency, as well as at a high relative concentration, between 9 and 18%. It should be noted that in homes this relation is a little different. For example, in Upper Silesia only *Micrococcus* was present in all studied homes, constituting 36% of the bacterial genera. The second most common was *Staphylococcus epidermidis*, present in 76% of homes and constituting 14% of the total [21]. Similar relative counts of *Micrococcus* and *Staphylococcus*, equal to 30% and 10% respectively, have been reported by Nevalainen for Finnish homes [9].

It should be noted that in the patient room in the hospital in Zabrze some actinomecytes were found. Their occurrence evidently indicates a moisture problem in this building. This relation is well documented in the literature (for example [9]).

More advanced analysis of differences between airborne bacteria resulting from different buildings and different hygiene conditions is very limited because the number of persons present in the room during sampling varied from case to case. This aspect is emphasized by the fact that most bacteria were typical skin bacteria. Hence, this problem requires further investigation. Fu-

Table 3. Viable bacterial genera identified in the hospital/clinic rooms.

Sampling location	Bacteria	Concentration [CFUm^{-3}]	% of the total bacterial genera
Office room	Gram-positive cocci, including:	112	68
	Staphylococcus simulans	75	
	Staphylococcus xylosus	4	
	Staphylococcus cohnii	4	
	Micrococcus spp.	18	
	Micrococcus luteus	11	
	Nonsporing Gram-positive rods, including:	50	30
	Arthrobacter spp.	14	
	Brevibacterium spp.	11	
	Brevibacterium epidermidis	7	
	Corynebacterium propinquum	18	
	Gram-positive rods, family *Bacillacae,*	4	2
	including: *Bacillus spp.*	4	
	TOTAL	166	100
Patient room in the hospital in Sosnowiec	Gram-positive cocci, including:	110	58
	Staphylococcus lugdunensis	57	
	Staphylococcus haemolyticus	42	
	Staphylococcus hominis	7	
	Micrococcus spp.	4	
	Nonsporing Gram-positive rods, including:	77	40
	Brevibacterium spp.	35	
	Aureobacterium spp./Corynebacterium aquaticum	21	
	Others	21	
	Gram-positive rods, family *Bacillacae,*	4	2
	including: *Bacillus spp.*	4	
	TOTAL	198	100

Table 3. continued

Patient room in the hospital in Zabrze	Gram-positive cocci, including:	552	77
	Staphylococcus epidermidis	198	
	Staphylococcus saprophyticus	74	
	Staphylococcus hominis	11	
	Staphylococcus xylosus	85	
	Staphylococcus cohnii cohnii	11	
	Micrococcus spp.	166	
	Kocuria	7	
	Nonsporing Gram-positive rods, including:	140	19
	Corynebacterium striatum/amycolatum	75	
	Corynebacterium aquaticum	25	
	Corynebacterium propinquum	25	
	Corynebacterium pseudodiphtheriticum	4	
	Microbacterium spp.	11	
	Actinomycetes, including:	32	4
	Rhodococcus spp.	21	
	Others	11	
	TOTAL	724	100
Operating room, Cardio-surgery hospital in Katowice-Ochojec, after I operation, during room cleaning	Gram-positive cocci, including:	143	73
	Staphylococcus xylosus	86	
	Staphylococcus haemolyticus	23	
	Staphylococcus epidermidis	13	
	Staphylococcus hominis	15	
	Staphylococcus saprophyticus	3	
	Micrococcus spp.	3	
	Nonsporing Gram-positive rods, including:	54	27
	Corynebacterium *striatum/amycolatum*	18	
	Brevibacterium spp.	18	
	Others	18	
	TOTAL	197	100
Operating room, Cardio-surgery hospital in Katowice-Ochojec, after II operation	Gram-positive cocci, including:	110	81
	Staphylococcus spp.	10	
	Staphylococcus cohnii	50	
	Micrococcus spp.	20	
	Micrococcus luteus	25	
	Kocuria rosea	5	
	Nonsporing Gram-positive rods, including:	25	19
	Corynebacterium *striatum/amycolatum*	15	
	Corynebacterium aquaticum	10	
	TOTAL	135	100

ture studies should also give information about the air velocity in the mechanically ventilated rooms, which is necessary for discussing the concentration data in more detail. Anyway, the obtained results indicate that the microbial air quality in the Silesian hospitals and clinics is quite similar to the indoor air in hospitals in other countries, although in the future the concentration of airborne bacteria in the clinic in Sosnowiec and in the hospital in Zabrze should be reduced below the level of some hundred $CFUm^{-3}$.

Finally, exposure to airborne bacteria in the studied buildings in Upper Silesia (concerning both aspects: concentration levels as well as identified genera and species) can be estimated, generally, as low and safe.

Summary and Conclusions

Bioaerosol characteristics were evaluated in different types of rooms in the clinic and in three hospitals in Upper Silesia. It was found that bacterial mean concentration is about 10^3 $CFUm^{-3}$ in the clinical outpatient room and ranges from 10^2 $CFUm^{-3}$ to10^3 $CFUm^{-3}$ in hospitals, depending on the number of occupants, and on the physical quality of the building. In patient rooms, bedmaking increases the concentration level twice. Respirable bacteria represent 41-86% of the total airborne bacteria.

The size distributions of airborne bacteria in naturally ventilated rooms have peaks in the size range between 1.1 and 3.3 µm. Mechanical supply and exhaust ventilation shift the peak into larger particles.

Staphylococcus was the most frequently occurring bacteria in the studied clinic/hospital rooms followed by *Micrococcus,* which together contributed 58-78% of total bacteria concentration.

The exposure to airborne bacteria in the studied buildings can be estimated, generally, as low and safe.

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