

VARIABILITY OF AIRBORNE MICROFLORA IN A HOSPITAL WARD WITHIN A PERIOD OF ONE YEAR

Marta Augustowska¹, Jacek Dutkiewicz²

¹State Railway Hospital, Lublin, Poland

²Department of Occupational Biohazards, Institute of Agricultural Medicine, Lublin, Poland

Augustowska M, Dutkiewicz J: Variability of airborne microflora in a hospital ward within a period of one year. *Ann Agric Environ Med* 2006, **13**, 99–106.

Abstract: The aim of the study was to determine the seasonal variability of the airborne microflora in a hospital ward of the pneumonological department, with regard to potential impact on respiratory status of asthmatic patients hospitalized in the ward. Microbiological air sampling was carried out for a period of 1 year from June–May, during work-days, 16–21 days per month. Each day, the air samples were collected twice: in the morning at 09:00 and in the afternoon at 13:00. Air samples were taken with a custom-designed particle-sizing slit sampler enabling estimations of both total and respirable fractions of the microbial aerosol. Air samples for determination of bacteria were taken on blood agar and air samples for determination of fungi were taken on Sabouraud agar. Mean monthly concentrations of total microorganisms (bacteria + fungi) in the air of the examined hospital ward were between 296.1–529.9 cfu/m³. Mean monthly concentrations of airborne bacteria ranged from 257.1–436.3 cfu/m³, with peak values in November and May and the lowest values from December to February. Mean monthly concentrations of airborne fungi showed much greater variation than bacteria and ranged from 9.9–96.1 cfu/m³ with the very distinct peak in November and the lowest value in May. The variations in monthly concentrations of total microorganisms, bacteria and fungi in the air of hospital ward were statistically significant ($p < 0.001$). The concentrations of total airborne microorganisms, bacteria and fungi recorded in the hospital in the morning were significantly greater compared to those recorded in the afternoon ($p < 0.01$). The mean monthly values of respirable fraction for total microorganisms were within a range of 17.3–44.4%, for bacteria within a range of 17.2–44.8%, and for fungi within a range of 2.2–39.1%. The most common microorganisms in the air of the examined ward were Gram-positive cocci which accounted for 31.4–46.4% of the total count. Gram-negative bacteria and corynebacteria were less numerous, forming respectively 11.8–27.5% and 9.6–20.0% of the total count. Endospore-forming bacilli and actinomycetes occurred in small proportions, respectively 0.3–3.2% and 0–2.0% of the total count. Fungi formed 7.6–42.5% of the total count. The prevailing species was *Aspergillus fumigatus* which constituted on average 77.0% of total fungal strains isolated from the air of the hospital ward. A significant decrease of spirometric indices (VC, FEV₁) in asthmatic patients hospitalized in the ward, at increase of the concentration of airborne bacteria and/or fungi, was found in 9 out of 24 examined patients (37.5%) and in 19 out of 192 analysed single relationships (9.9%). In conclusion, although bacteria and fungi occurred in the air of the examined hospital ward in relatively low numbers (of the order 10² cfu/m³ and 10¹ cfu/m³ respectively), they should be considered as a possible cause of asthma exacerbations in some patients because of the presence of *Aspergillus fumigatus* and other potentially pathogenic species.

Address for correspondence: Prof. Jacek Dutkiewicz, PhD, Head of Department of Occupational Biohazards, Institute of Agricultural Medicine, Jaczewskiego 2, 20-090 Lublin, Poland. E-mail: dutkiewi@galen.imw.lublin.pl

Key words: hospital, pneumonology department, airborne microflora, seasonal variability, asthma exacerbations, bioaerosols, bacteria, fungi, *Aspergillus fumigatus*.

INTRODUCTION

Airborne microflora in hospital rooms was the subject of numerous studies as a potential cause of hospital

infections [13, 14, 18, 28, 41]. Most of the studies were performed in intensive care units, surgical units, haematological wards, maternity wards and other departments where the risk of infections is greatest [2, 13, 19, 28, 38,

40]. The levels of microorganisms (bacteria and fungi) found in most rooms were of the order 10^1 - 10^3 cfu/m³ [8, 16, 18, 19, 20, 30, 36, 37, 39, 40], except for rooms of high cleanness such as operating theatres or transplant units where they were of the order 10^{-1} - 10^1 cfu/m³ [13, 28, 38].

So far, little is known about microflora of air in the pneumonological departments of hospitals. Doleżal *et al.* [8] found the concentrations of microorganisms in the air of a pneumonological clinic within the range of 998-1311 cfu/m³. To the best of our knowledge, no long-term studies on airborne microflora have been performed in pneumonological departments.

The aim of the present work was to study the variability of the airborne microflora of a hospital ward of the pneumonological department within a period of 1 year, with regard to potential impact on respiratory status of asthmatic patients hospitalized in the ward.

MATERIAL AND METHODS

Study area. Long-term microbiological examinations of the air were carried out in one of the wards of the pneumonological department of the State Railway Hospital in Lublin (eastern Poland). The pneumonological department is located in a separate, two-storied building, surrounded with deciduous trees. The ward selected for study has an area of 16 m², the cubic capacity of 48 m³, and the gravity ventilation. Between 2-5 patients were hospitalized simultaneously in the ward during the study period.

Time and scope of the study. Microbiological air sampling was carried out for a period of 1 year from 9

June 1981 to 28 May 1982, during work-days, 16-21 days per month. Each day, the air samples were collected twice: in the morning at 09:00 and in the afternoon at 13:00. During the whole study period, the following microclimatic parameters were measured every day: temperature with a week thermograph TŻ-18, relative humidity with a hygrograph TŻ-18, and atmospheric pressure with a mercury barometer.

Microbiological examination of the air. Air samples were taken with a custom-designed particle-sizing slit sampler [9] enabling 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 m³; 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 [46].

Every day, at each sampling time (at 09:00 and 13:00) double samples were taken on the following agar media: blood agar for determination of total mesophilic bacteria, and Sabouraud agar for determination of fungi. The samples were taken at the height of 150 cm. The blood agar plates were incubated for 3 days at 37°C, and the

Table 1. Concentrations of total microorganisms (bacteria + fungi) in air of a hospital ward during the year (June 1981–May 1982).

Months	VI (N=16)	VII (N=21)	VIII (N=19)	IX (N=21)	X (N=21)	XI (N=21)	XII (N=19)	I (N=21)	II (N=20)	III (N=21)	IV (N=21)	V (N=20)
Sampling time												
Morning (09:00)	404.4 ±	441.7 ±	385.7 ±	390.6 ±	480.4 ±	608.2 ±	343.0 ±	311.4 ±	336.7 ±	381.7 ±	455.1 ±	430.5 ±
x ± S.D.	200.6	218.8	117.3	110.9	216.5	247.2	141.0	94.0	96.0	96.6	176.0	253.6
Respirable fraction (%)	25.8	47.5	29.6	16.2	31.5	27.5	24.9	21.9	24.5	21.2	19.2	20.1
Afternoon (13:00)	319.1 ±	353.3 ±	337.8 ±	284.0 ±	394.8 ±	451.6 ±	249.2 ±	300.0 ±	288.4 ±	278.5 ±	337.0 ±	420.4 ±
x ± S.D.	164.0	226.0	137.8	118.3	183.7	121.6	96.5	92.5	127.8	114.9	156.7	184.1
Respirable fraction (%)	30.7	41.0	30.2	18.3	31.4	28.0	32.1	18.1	15.9	16.2	20.8	23.3
Average												
x ± S.D.	361.7 ±	397.5 ±	362.2 ±	337.3 ±	437.6 ±	529.9 ±	296.1 ±	305.7 ±	312.5 ±	330.1 ±	396.0 ±	425.4 ±
	274.0	259.0	173.8	265.0	348.6	460.7	257.1	85.1	151.9	253.5	311.6	197.0
Respirable fraction (%)	28.3	44.3	29.9	17.3	31.5	27.8	28.5	20.0	20.2	18.7	20.0	21.7
Microclimatic parameters (average)												
Temperature (°C)	20	21	23	19	20	20	21	18	18	20	21	20
Relative humidity (%)	70	66	68	70	65	57	55	49	53	50	50	62
Atmospheric pressure (mm Hg)	754	754	758	759	753	756	751	758	764	759	756	760

N – number of days in which samples were collected.

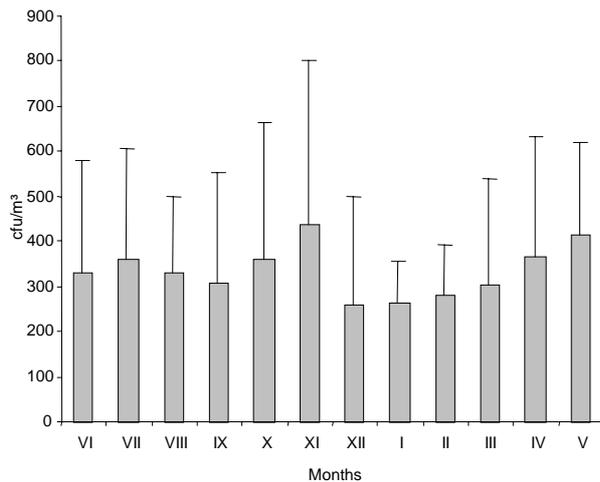


Figure 1. Monthly fluctuations in concentration of bacteria in air of a hospital ward (mean \pm S.D.).

Sabouraud agar plates were incubated for 7 days at 28°C. The grown colonies were counted and differentiated and the data reported as cfu per 1 cubic metre of air (cfu/m³). The total concentration of microorganisms in the air was obtained by the addition of the concentrations of total mesophilic bacteria and fungi. The percent composition of the total microflora of the air was then determined.

Bacterial isolates were identified with microscopic and biochemical methods, as recommended by Bergey's Manual [5] and Cowan & Steel [6]. Fungi were classified with microscopic methods, according to Barron [4], Litvinov [29], and Raper & Fennell [42].

Spirographic examinations of patients. For determination of the potential impact of microbial pollution of air on the respiratory status of asthmatic patients in the examined ward, spirographic examinations of patients were conducted during a period from 4 January 1982 to 28 May 1982. A total of 24 patients (17 males and 7 females, age range 23-80 years) with the diagnosis of bronchial asthma were examined. The impairment of lung function was severe in 10 patients, moderate in 5 patients, mild in 4 patients, and absent in 5 others. The patients were hospitalized in the examined ward for a period of 1-8 weeks.

The spirographic examinations were carried out with the use of a "Vitalograph" twice a day, in parallel with air sampling: in the morning at 09:00 and in the afternoon at 13:00. Forced expiratory volume in 1 second (FEV₁) and vital capacity (VC) were determined.

Statistical analysis. The data were analysed by Shapiro-Wilk test for distribution, chi² test, Wilcoxon matched-pairs test, Spearman's correlation test and Pearson's test for correlation, using STATISTICA for Windows v. 5.0 package (Statsoft Inc., Tulsa, Oklahoma, USA).

The maintenance conditions in the examined pneumological department did not substantially change since the study period. The results of this study have not been published until now, except for a short communication in

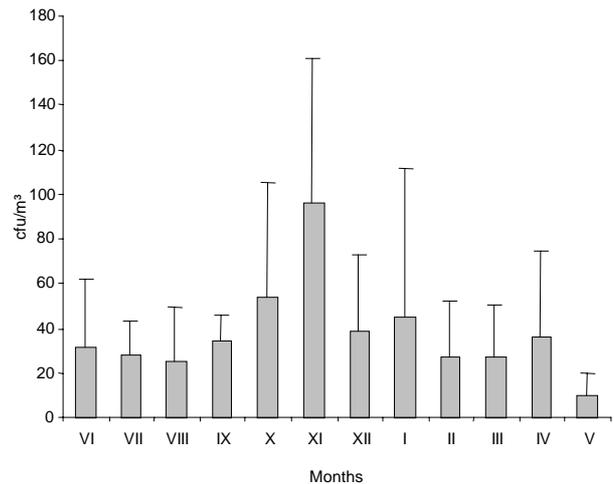


Figure 2. Monthly fluctuations in concentration of fungi in air of a hospital ward (mean \pm S.D.).

which the results for the month of January 1982 were presented [3].

RESULTS

Seasonal fluctuations of airborne microflora. Mean monthly concentrations of total microorganisms (bacteria + fungi) in the air of the examined hospital ward were between 296.1–529.9 cfu/m³, with the highest values in November (529.9 cfu/m³), October (437.6 cfu/m³) and May (425.4 cfu/m³), and the lowest values in winter months: December (296.1 cfu/m³), January (305.7 cfu/m³), and February (312.5 cfu/m³) (Tab. 1).

The fluctuations in the monthly concentrations of bacteria in the air of the hospital ward are presented in Figure 1. The concentrations of airborne bacteria ranged from 257.1–436.3 cfu/m³, with peak values in November and May and the lowest values from December to February.

The fluctuations in the monthly concentrations of fungi in the air of the hospital ward (Fig. 2) were much greater compared to bacteria. The numbers of airborne fungi ranged from 9.9–96.1 cfu/m³ with the very distinct peak in November and the lowest value in May.

The variations in monthly concentrations of total microorganisms, bacteria and fungi in the air of the hospital ward were statistically significant ($p < 0.001$). No significant relationship was found between the microclimatic parameters of the hospital ward (temperature, relative humidity, atmospheric pressure) and the concentrations of total microorganisms, bacteria and fungi in the air.

Daily fluctuations of airborne microflora. The concentrations of total airborne microorganisms, bacteria and fungi recorded in the hospital in the morning (respectively 311.4–608.2 cfu/m³, 267.1–505.6 cfu/m³, and 11.3–107.4 cfu/m³) were significantly greater compared to those recorded in the afternoon (respectively 249.2–451.6 cfu/m³, 213.6–410.9 cfu/m³, and 8.5–84.8 cfu/m³) ($p < 0.01$) (Tab. 1).

Respirable fraction. The mean monthly values of respirable fraction for total microorganisms were within the range of 17.3-44.4% (Tab. 1), for bacteria within the range of 17.2-44.8%, and for fungi within the range of 2.2-39.1%.

Composition of airborne microflora. The composition of microflora isolated from the air of the hospital ward is presented in Figure 3 (total count) and Figure 4 (respirable fraction). As seen from these Figures, the most common microorganisms were Gram-positive cocci which accounted for 31.4-46.4% of the total count and 37.2-49.6% of the respirable fraction. Prevailing isolates were the coagulase-negative white staphylococci, classified as *Staphylococcus epidermidis*, while the remaining part of this group belonged to the genera *Micrococcus* and *Streptococcus*. Coagulase-positive strains of *Staphylococcus aureus* were not found.

Gram-negative bacteria constituted 11.8-27.5% of the total count and 5.6-30.2% of the respirable fraction. The most common were strains of *Flavobacterium* spp., followed by *Acinetobacter calcoaceticus* and *Pantoea agglomerans*. The other isolates were identified as *Escherichia coli*, *Enterobacter* spp., *Klebsiella oxytoca*, *Pseudomonas aeruginosa*, *Branhamella catarrhalis* and *Neisseria flavescens*.

Corynebacteria formed 9.6-20.0% of the total count and 4.5-22.1% of the respirable fraction. The most common were strains of *Arthrobacter* spp. followed by *Brevibacterium* spp. Less numerous were strains of *Corynebacterium* spp. and *Rhodococcus* spp. Endospore-forming bacilli (*Bacillus* spp.) were relatively rare and constituted 0.3-3.2% of the total count and 0-4.5% of the respirable fraction. They comprised the species *Bacillus subtilis*, *Bacillus cereus* and *Bacillus megaterium*. Actinomycetes (*Streptomyces albus*, *Streptomyces* spp.)

Table 2. Correlation between concentration of microorganisms in air of hospital ward and spirometric indices in asthmatic patients hospitalized in the ward.

No.	Initials	Number of compared measurements	Compared correlation							
			Concentration of bacteria versus spirometric indices				Concentration of fungi versus spirometric indices			
			Total bacteria		Respirable bacteria		Total fungi		Respirable fungi	
			VC	FEV ₁	VC	FEV ₁	VC	FEV ₁	VC	FEV ₁
1	B. S.	21	*	**	*	*	(-)	(-)	*	(-)
2	C. A.	18	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
3	C. R.	30	(-)	(-)	*	(-)	(-)	(-)	(-)	(-)
4	F. A.	20	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
5	G. B.	12	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
6	K. H. 1	9	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
7	O. F.	57	(-)	*	(-)	**	(-)	(-)	(-)	(-)
8	P. F. 1	25	(-)	(-)	(-)	(-)	(-)	*	(-)	(-)
9	S. C.	27	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
10	T. W.	70	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
11	W. A.	12	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
12	G. S.	32	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
13	K. A. 1	27	(-)	*	(-)	(-)	(-)	*	(-)	(-)
14	K. H. 2	11	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
15	Ł. S.	34	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
16	T. M.	12	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
17	N. S.	18	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
18	O. M.	35	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
19	W. M.	30	**	(-)	*	(-)	(-)	(-)	(-)	*
20	K. A. 2	20	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
21	P. F. 2	42	(-)	(-)	(-)	(-)	**	(-)	(-)	(-)
22	R. J.	40	(-)	(-)	(-)	(-)	(-)	(-)	(-)	*
23	B. B.	24	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
24	P. T.	38	(-)	(-)	*	(-)	**	*	(-)	(-)

*-** statistically significant negative correlation: *p<0.05, **p<0.01. (-): no significant correlation.

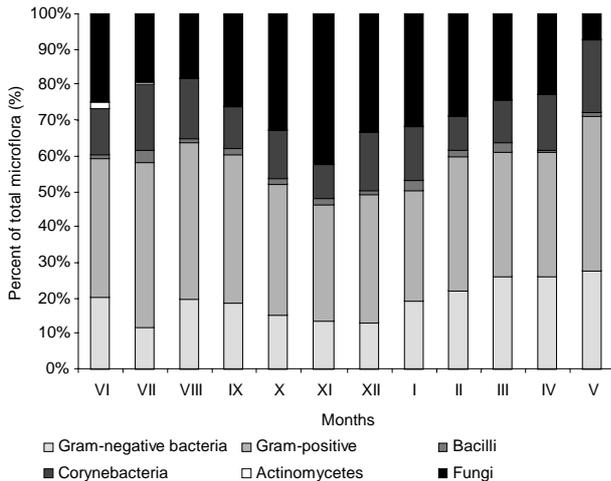


Figure 3. Composition of airborne microflora in a hospital ward: total count.

occurred in very small numbers and formed only 0-2.0% of the total count and 0-2.0% of the respirable fraction.

Fungi constituted 7.6-42.5% of the total count and 0.9-41.0% of the respirable fraction. The prevailing species was *Aspergillus fumigatus* which formed on average 77.0% of all fungal strains isolated from the air of the hospital ward. Other fungal isolates comprised *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus* spp., *Penicillium* spp., *Geotrichum candidum*, *Trichoderma album*, *Mucor* spp., and *Rhizopus nigricans*. As seen in Figure 5, the concentration of *Aspergillus fumigatus* in the air was similar to other fungi during the summer months between June and September, and starting from October rapidly raised with a distinct peak in November (96.1 cfu/m³, 91.1% of all fungi). The numbers of *Aspergillus fumigatus* remained relatively high during winter and spring months and rapidly dropped in May (Fig. 5).

Correlation between concentration of microorganisms in the air of the hospital ward and lung function tests in asthmatic patients hospitalized in the ward.

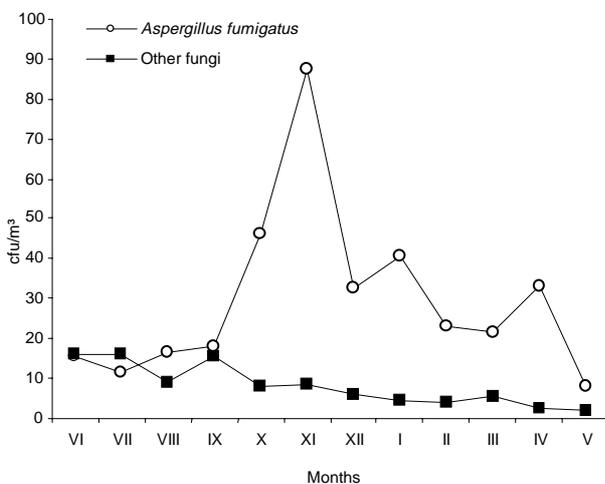


Figure 5. Monthly concentrations of *Aspergillus fumigatus* versus other fungi in the air of a hospital ward.

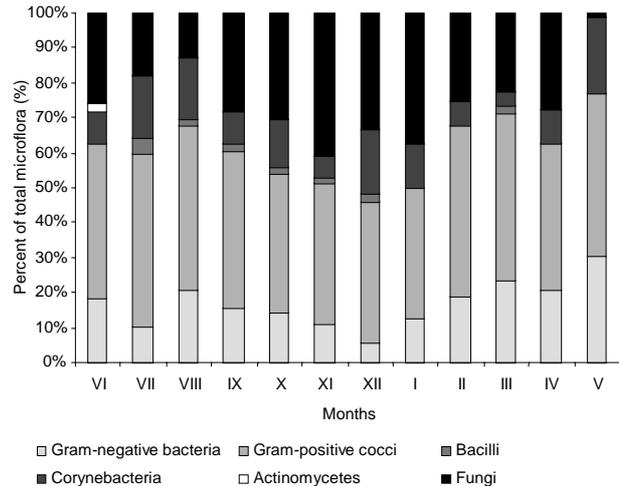


Figure 4. Composition of airborne microflora in a hospital ward: respirable fraction.

The significant decrease of spirometric indices (VC, FEV₁) in asthmatic patients at increase of the concentration of bacteria and/or fungi in the air of the hospital ward was found in 9 out of 24 examined patients (37.5%) and in 19 out of 192 analysed single relationships (9.9%). The decrease of vital capacity (VC) was correlated in 2 cases with the increase of the concentration of total bacteria, in 4 cases – of respirable bacteria, in 2 cases – of total fungi, and in 1 case – of respirable fungi. The decrease of forced expiratory volume in 1 second (FEV₁) was correlated in 3 cases with an increase of the concentration of total bacteria in 2 cases – of respirable bacteria, in 3 cases – of total fungi, and in 2 cases – of respirable fungi (Tab. 2). Altogether, out of 19 significant single relationships, in 11 cases the decrease of spirometric indices was due to an increase of bacteria, and in 8 cases due to an increase of fungi.

DISCUSSION

The levels of airborne bacteria measured in the hospital ward of the pneumonological department within a period of 1 year were of the order 10² cfu/m³. They showed a significant seasonal variation but without distinctly directed trends. The recorded levels of airborne bacteria were lower compared to those reported by Doležal *et al.* [8] from a pneumonological department, by Kelsen & McGuckin [18] from a cardiothoracic surgical unit, by Kotlarek-Haus *et al.* [19] from a haematological department, and by Panecka *et al.* [39] from a surgical unit; - similar to those reported by Nunes *et al.* [37] from general hospitals; - and greater compared to those reported by Li & Hou [28] from operating theatres.

Staphylococci prevailed among bacteria isolated from the air of the examined hospital ward, similar to the studies of earlier authors who examined microflora of air in hospitals [8, 13, 16, 19, 39]. The risk of exposure to staphylococci was diminished by the fact that the isolated strains were coagulase-negative and unlikely to cause infections. Gram-negative bacteria found in the air of the

hospital ward could be a source of adverse endotoxin [10, 11, 24] and *Acinetobacter* strains may be a potential cause of hospital infections transmitted by air [1]. Some of the Gram-positive isolates belonging to corynebacteria and actinomycetes (*Arthrobacter* spp., *Brevibacterium* spp., *Streptomyces albus*) reveal allergenic properties [10, 23, 31].

The levels of airborne fungi measured in the hospital ward of the pneumonological department within a period of 1 year were of the order 10^1 cfu/m³. The recorded levels of airborne fungi were lower compared to those reported by Nowicka *et al.* [36] from a haematological department, by Krajewska *et al.* [20] from an obstetrics department, by Rainer *et al.* [41] from a bone-marrow transplantation unit, and by Martins-Diniz *et al.* [30] from a surgical centre and intensive care units; - similar to those reported by Pini *et al.* [40] from haematology wards, and by Nunes *et al.* [37] and Herman [14] from general hospitals; - and greater compared to those reported by Panagopoulou *et al.* [38] from high-risk departments of hospitals, and by Li & Hou [28] from operating theatres.

Fungal flora of the air of the examined hospital ward was dominated by the species *Aspergillus fumigatus* which formed 77% of total isolates. *Aspergillus fumigatus* is a known hazardous agent which may cause allergic alveolitis, asthma, pulmonary aspergillosis and possibly mycotoxicoses [7, 10, 14, 21, 23, 24, 26]. Some other fungi isolated from the air of the ward (*Aspergillus niger*, *Aspergillus flavus*, *Penicillium* spp., *Rhizopus nigricans*) also pose a respiratory risk as potential sources of allergens and toxins [10, 12, 23, 24, 32]. Holmberg [15] has shown that the concentrations of *Aspergillus* spores above 50 cfu/m³ in Swedish dwellings were associated with a higher prevalence of the sick building syndrome - symptoms in exposed people. The prevalence of *Aspergillus* spp. spores in the air of hospital wards was found also by Noble & Clayton [35], by Thirumalaikolundusubramanian *et al.* [45], by Lentino *et al.* [27], and by Panagopoulou *et al.* [38]. In contrast, Nowicka *et al.* [36], Pini *et al.* [40], and Martins-Diniz *et al.* [30] found the prevalence of *Cladosporium* (*Cladophialophora*) spp. and Krajewska *et al.* [20] the prevalence of *Candida* spp. in the air of examined hospital rooms.

In this study, fungi showed a highly significant seasonal variation which was distinctly greater compared to bacteria. It was characterized by a very distinct peak in November and with the lowest value in May. Keeping in mind that this peak was due to the increase of *Aspergillus fumigatus* spores, our results correspond to those reported by Noble & Clayton [35] and Mullins *et al.* [34], who also found the seasonal peaks of *Aspergillus fumigatus* in the air of European hospitals in the autumn and winter months. The present results are also in agreement with those of Herman [14] who noted the highest counts of airborne fungi outside and inside the hospital from late September through to early December, and with those

obtained by Laham *et al.* [25], who reported the peak values of the concentration of *Aspergillus* spp. in the air of hospital rooms during autumn and winter. In contrast, Solomon *et al.* [43] and Hospenthal *et al.* [17] have not found a seasonal variability of airborne *Aspergillus fumigatus* spores in hospitals located in the USA. The concentrations of *Aspergillus fumigatus* spores found in the air of the hospital ward in the present study are lower compared to those reported by Noble & Clayton [35], but greater compared to those reported by Solomon *et al.* [43], Arnow *et al.* [2], and Hospenthal *et al.* [17]. Generally, *Aspergillus fumigatus* is recovered commonly from the hospital environment, posing a particular risk of invasive aspergillosis in immunocompromised patients [2, 14, 17, 33, 40].

Staib *et al.* [44] reported the increase of *Aspergillus fumigatus* in the open air of West Berlin (Germany) between October and March, which is in accordance with the results of the present work. Mullins *et al.* [34] explain the "winter" increase of airborne *Aspergillus fumigatus* spores by the availability of nutrient-rich decaying plant debris originating from fallen leaves, which promotes growth of the fungus. The possible impact of fallen leaves on increase of the outdoor and indoor *Aspergillus* counts is pointed out also by Herman [14]. The above-mentioned suggestions could be considered in the case of our study, as the hospital building is surrounded with deciduous trees shedding leaves in autumn.

Different patterns of variability of airborne fungi in the air of hospitals were reported from countries characterized by warm or hot climates. Panagopoulou *et al.* [38] found the highest levels of airborne fungi in examined hospitals in Greece in summer and autumn. Martins-Diniz *et al.* [30], who found the prevalence of *Cladosporium* in the air of examined hospital wards in Brazil, noted the peak value of airborne fungi in July.

As there are no generally accepted threshold limit values concerning concentrations of bacteria and fungi in the air of hospitals, the obtained results could be compared only with the values recommended by various authors or institutions. The mean levels of airborne fungi noted in the present work were below the value proposed by Krzysztofik [22] for hospital wards, equal to 200 cfu/m³, whereas the mean levels of airborne bacteria were below the Polish state recommendation for class III hospitals equal to 700 cfu/m³ and slightly above the Polish state recommendation for class II hospitals, equal to 300 cfu/m³, and the European recommendation for class III hospitals, equal to 200-300 cfu/m³ [20, 22].

In 9 out of 24 asthmatic patients (37.5%) hospitalized in the examined ward during the study period, significant correlations were found between increase of the concentrations of bacteria and/or fungi in the air of the ward and decrease of the lung function parameters. Numbers of single correlations associated with the increase of bacteria or fungi were similar (respectively 11 and 8) although the numbers of airborne bacteria in the ward were circa 10 times greater than fungi. This seems

to indicate the substantial role of fungi in causing decrease of lung function parameters and asthma exacerbations in prone patients, which could be caused either by allergen-specific reactions, or nonspecific, immunotoxic reactions. It was evidenced that sensitization and exposure to airborne moulds is associated with exacerbations of asthma [47], and that low numbers of *Aspergillus* spores in the air (50 cfu/m³), which were exceeded in the course of this study, could evoke symptoms of sick building syndrome [15]. Nevertheless, the obtained statistical results could be taken with caution, as only 10% of the analysed single relationships proved to be significant. Another limitation of the study is that the patients could not be tested with bacterial and fungal allergens to confirm the observed relationships.

CONCLUSIONS

1. Monitoring of airborne microflora of a hospital ward of the pneumonological department during 1 year revealed levels of bacteria of the order 10² cfu/m³ and levels of fungi of the order of 10¹ cfu/m³.

2. The concentrations of airborne bacteria and fungi showed a significant monthly variation. It was much more expressed in fungi than in bacteria with a distinct peak in November and with the lowest value in May.

3. *Aspergillus fumigatus* was predominant among airborne fungi and staphylococci prevailed among bacteria.

4. Results of statistical analysis suggest that in some asthmatic patients the increase in numbers of airborne microbes in the hospital ward might be associated with decrease of lung function parameters.

REFERENCES

- Allen KD, Green HT: Hospital outbreak of multi-resistant *Acinetobacter anitratus*: an airborne mode of spread? *J Hosp Infect* 1987, **9**, 110-119.
- Arnou PM, Sadigh M, Costas C, Weil D, Chudy R: Endemic and epidemic aspergillosis associated with in-hospital replication of *Aspergillus* organisms. *J Infect Dis* 1991, **164**, 998-1002.
- Augustowska M, Durda M, Dutkiewicz J: Badania nad mikroflorą powietrza sal szpitalnych w aspekcie możliwego oddziaływania na stan chorych z astmą oskrzelową [Microflora of the air of hospital wards as a possible factor influencing the clinical condition of asthmatic patients]. In: III Konferencja Naukowa nt. „Biozanieczyszczenia w Budynkach a Zdrowie”, Referaty - cz. II. *Zeszyty Naukowe AWF w Krakowie* 1983, **35**, 13-20 (in Polish).
- Barron GL: *The Genera of Hyphomycetes from Soil*. Williams & Wilkins, Baltimore 1968.
- Buchanan RE, Gibbons NE (Eds): *Bergey's Manual of Determinative Bacteriology*. Williams and Wilkins, Baltimore 1974.
- Cowan ST, Steel KJ: *Manual for the Identification of Medical Bacteria*. University Press, Cambridge 1965.
- DiSalvo AF (Ed): *Occupational Mycoses*. Lee and Febiger, Philadelphia 1983.
- Doleżał M, Doleżał M, Pawliszyn W, Śliwa A, Owsiański J, Hartwich A: Mikroflora powietrza pomieszczeń szpitalnych a codzienna praca kliniki [Atmospheric microflora in hospital wards and everyday hospital routine]. *Pol Tyg Lek* 1978, **33**, 613-616 (in Polish).
- Dutkiewicz J, Kwapiszewski C: Nowy aparat do badania mikrobiologicznego zanieczyszczenia powietrza [New sampler for microbiological examination of the air]. *Ochrona Powietrza* 1975, **9(2)**, 37-42 (in Polish).
- Dutkiewicz J, Jabłoński L: *Biologiczne Szkodliwości Zawodowe [Occupational Biohazards]*. PZWL, Warsaw 1989 (in Polish).
- Dutkiewicz J: Bacteria and fungi in organic dust as potential health hazard. In: Midtgård U, Poulsen OM (Eds): Waste Collection and Recycling – Bioaerosol Exposure and Health Problems. Proceedings of an International Meeting held in Køge, Denmark, 13-14 September 1996. *Ann Agric Environ Med* 1997, **4**, 11-16.
- Eduard W, Sandven P, Levy F: Serum IgG antibodies to mold spores in two Norwegian sawmill populations: relationship to respiratory and other work-related symptoms. *Am J Ind Med* 1993, **24**, 207-222.
- Gould JC: Airborne pathogenic bacteria in a tissue transplant unit. In: Silver IH (Ed): *Aerobiology*. Academic Press, London 1970, 62-76.
- Herman LG: *Aspergillus* in patient care areas. *Ann NY Acad Sci* 1980, **353**, 140-146.
- Holmberg K: Indoor mould exposure and health effects. In: Seifert B et al. (Eds): *Indoor Air' 87. Vol. 1: Volatile Organic Compounds, Combustion Gases, Particles and Fibers, Microbiological Agents*, 637-642. Institute for Water, Soil and Air Hygiene, Berlin 1987.
- Horoszewicz J: Badania nad mikroflorą powietrza pomieszczeń Centralnego Wojskowego Szpitala Klinicznego w Łodzi [Studies on microflora of the air of Central Military Hospital in Łódź]. *Biul WAM* 1958, **1**, 86-93 (in Polish).
- Hospenthal DR, Kwon-Chung KJ, Bennett JE: Concentrations of airborne *Aspergillus* compared to the incidence of invasive aspergillosis: lack of correlation. *Med Mycol* 1998, **36**, 165-168.
- Kelsen SG, McGuckin M: The role of airborne bacteria in the contamination of fine particle nebulizers and the development of nosocomial pneumonia. *Ann NY Acad Sci* 1980, **353**, 218-229.
- Kotlarek-Haus S, Nowicka I, Juźwiakowa J, Siemek R: Analiza zanieczyszczenia bakteryjnego powietrza w salach oddziału hematologicznego [Analysis of bacterial pollution of the air in wards of haematological department]. *Pol Tyg Lek* 1980, **35**, 1149-1152 (in Polish).
- Krajewska K, Krajewska-Kułak E, Łukaszuk C, Rolka H, Lach J, Karczewski J: Analiza występowania patogenów grzybiczych w powietrzu sal oddziału położniczego [Analysis of the incidence of fungal pathogens in the air of rooms of obstetrics department]. *Gin Pol* 2004, **75**, 451-456 (in Polish).
- Krysińska-Traczyk E: Pleśnie *Aspergillus fumigatus* jako przyczyna schorzeń płuc o charakterze zawodowym [Molds *Aspergillus fumigatus* as a cause of respiratory diseases of occupational origin]. *Med Wiejska* 1973, **8**, 275-284 (in Polish).
- Krzysztofik B: *Mikrobiologia Powietrza [Microbiology of the Atmosphere]*. Wydawnictwo Politechniki Warszawskiej, Warsaw 1992 (in Polish).
- Lacey J, Crook B: Review: Fungal and actinomycete spores as pollutants of the workplace and occupational allergens. *Ann Occup Hyg* 1988, **32**, 515-533.
- Lacey J, Dutkiewicz J: Bioaerosols and occupational lung disease. *J Aerosol Sci* 1994, **25**, 1371-1404.
- Laham MN, Jefery B, Carpenter JL: Frequency of clinical isolation and winter prevalence of different *Aspergillus* species at a large southwestern army medical center. *Ann Allergy* 1982, **48**, 215-219.
- Land CJ, Hult K, Fuchs R, Hagelberg S, Lundström H: Tremorgenic mycotoxins from *Aspergillus fumigatus* as a possible occupational health problem in sawmills. *Appl Environ Microbiol* 1987, **53**, 787-790.
- Lentino JR, Rosenkranz MA, Michaels JA, Kurup VP, Rose HD: Nosocomial aspergillosis: a retrospective review of airborne disease secondary to road construction and contaminated air conditioners. *Am J Epidemiol* 1982, **116**, 430-437.
- Li CS, Hou PA: Bioaerosol characteristics in hospital clean rooms. *Sci Total Environ* 2003, **305**, 169-176.
- Litvinov MA: *Opredelitel' Mikroskopicheskikh Pochvennykh Gribov [Guide for Determination of the Microscopic Soil Fungi]*. Izd. Nauka, Leningrad 1967 (in Russian).
- Martins-Diniz JN, da Silva RA, Miranda ET, Mendes-Giannini MJ: Monitoring of airborne fungus and yeast species in a hospital unit. *Rev Saude Publica* 2005, **39**, 398-405.

31. Milanowski J, Dutkiewicz J, Potoczna H, Kuś L, Urbanowicz B: Allergic alveolitis among agricultural workers in eastern Poland: A study of twenty cases. *Ann Agric Environ Med* 1998, **5**, 31-43.
32. Miller JD, Laflamme AM, Sobol Y, Lafontaine P, Greenhalgh R: Fungi and fungal products in some Canadian houses. *Int Biodeter* 1988, **24**, 103-120.
33. Morris G, Kokki MH, Anderson K, Richardson MD: Sampling of *Aspergillus* spores in air. *J Hosp Infect* 2000, **44**, 81-92.
34. Mullins J, Harvey R, Seaton A: Sources and incidence of airborne *Aspergillus fumigatus* (Fres). *Clin Allergy* 1976, **6**, 209-217.
35. Noble WC, Clayton YM: Fungi in the air of hospital wards. *J Gen Microbiol* 1963, **32**, 397-402.
36. Nowicka I, Kotlarek-Haus S, Walów B, Baran E: Flora grzybicza powietrza różnych pomieszczeń oddziału hematologicznego [Fungal flora of the air in various rooms of haematological department]. *Przeg Lek* 1982, **39**, 305-308 (in Polish).
37. Nunes ZG, Martins AS, Altoe AL, Nishikawa MM, Leite MO, Aguiar PF, Fracalanza SE: Indoor air microbiological evaluation of offices, hospitals, industries and shopping centers. *Mem Inst Oswaldo Cruz, Rio de Janeiro* 2005, **100**, 351-357.
38. Panagopoulou P, Filoti J, Petrikkos G, Giakouppi P, Anatoliotaki M, Farmaki E, Kanta A, Apostolou H, Avlami A, Samonis G, Roilides E: Environmental surveillance of filamentous fungi in three tertiary care hospitals in Greece. *J Hosp Infect* 2002, **52**, 185-191.
39. Panecka A, Rękawek M, Hencner Z, Dutkiewicz J: Oddział intensywnej opieki pooperacyjnej w świetle porównawczych badań bakteriologicznych [Intensive postoperative care unit in the light of comparative bacteriological investigations]. *Szpital Pol* 1981, **25**, 107-110 (in Polish).
40. Pini G, Donato R, Faggi E, Fanci R: Two years of a fungal aerobiocontamination survey in a Florentine haematology ward. *Eur J Epidemiol* 2004, **19**, 693-698.
41. Rainer J, Peintner U, Poder R: Biodiversity and concentration of airborne fungi in a hospital environment. *Mycopathologia* 2001, **149**, 87-97.
42. Raper KB, Fennell DI: *The Genus Aspergillus*. Williams & Wilkins, Baltimore 1965.
43. Solomon WR, Burge HP, Boise JR: Airborne *Aspergillus fumigatus* levels outside and within a large clinical center. *J Allergy Clin Immunol* 1978, **62**, 56-60.
44. Staib F, Abel T, Mischura SK, Grosse G, Focking SK, Blisse A: Zum Vorkommen von *Aspergillus fumigatus* in Berlin West – Ein Beitrag zur Epidemiologie der Aspergillose des Menschen [Occurrence of *Aspergillus fumigatus* in West Berlin – a contribution to epidemiology of aspergillosis in man]. *Zbl Bakt Hyg I Abt Orig A* 1978, **241**, 337-357 (in German).
45. Thirumalaikolundusubramanian P, Uma A, Chandramohan M, Thiagarajan R, Muthu AK, Johnson ES: Yeast and mould flora in hospital air and hospital patients in India. In: Preusser HJ (Ed): *Medical Mycology*, 115-122. Zbl Bact Supl 8, Gustav Fischer, Stuttgart 1980.
46. *Threshold Limit Values for Chemical Substances and Physical Agents and Biological Exposure Indices*, 1993-1994. American Conference of Governmental Industrial Hygienists, Cincinnati 1993.
47. Zureik M, Neukirch C, Leynaert B, Liard R, Bousquet J, Neukirch F; European Community Respiratory Health Survey: Sensitisation to airborne moulds and severity of asthma: cross sectional study from European Community respiratory health survey. *BMJ* 2002, **325**, 411-414.