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## OCCURRENCE OF FUNGAL AEROSOL IN OVERGROUND AND UNDERGROUND HEALTH RESORTS

### BADANIA AEROZOLU GRZYBOWEGO W SANATORIUM NADZIEMNYM I PODZIEMNYM

**Abstract:** For many years sanatorium therapeutics has been very popular in Poland as well as abroad. Detailed pulmonological research showed the appropriateness of such treatment in respiratory diseases. According to this, specific environmental conditions, which occur in the underground subterranean therapy chambers and rooms of overground sanatoriums become more and more important. The main aim of the research was to define and divide the particles of biological aerosol into fractions (depending on their aerodynamical diameters) into: over 7.0  $\mu\text{m}$ , 7.0–4.7  $\mu\text{m}$ , 4.7–3.3  $\mu\text{m}$ , 3.3–2.1  $\mu\text{m}$ , 2.1–1.1  $\mu\text{m}$  and 1.1–0.65  $\mu\text{m}$  which occur in the underground subterranean therapy chambers in Salt Mine in Bochnia and in overground sanatorium chambers in “Health Resort Szczawnica”. The microbiological analyses of the air were carried out during the winter and spring of 2008, in 2 underground sanatorium chambers in Bochnia and in sanatorium chambers in Szczawnica. The measurements were carried out by means of the six-step Graseby-Anderson impactor from the human’s respiratory zone (oral and nasal cavity position) in the state of so called “original sterility” which is before introducing sick people and the personnel into the chambers and during the period of visitors stay. As a result of the analyses, significant differences between the amount of fungal aerosol in different places of chambers were showed. It has been ascertained that the biological aerosol occurred in subterranean therapy chambers in definitely higher concentrations during the treatment activity than during the period of the break between the turns. The highest concentrations of fungi in Salt Mine in Bochnia ascertain in Koldras Chamber and gymnasium in Wazyń Chamber; in Szczawnica the highest concentration was in the pump room.

**Keywords:** health resorts, fungi, air, salt mine

The degree of air pollution has great influence on human organism. Dust and gas pollutants play the main role in atmospheric air contamination, but the recent research has shown, that microbiological contamination is equally, and sometimes even more important [1–7].

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The occurring air contamination is the reason why immunological barrier in humans weakens, which results in potentially pathogenic and sometimes even saprophytic microorganisms becoming the cause of diseases. The degree of microbiological contamination of air in closed rooms, especially these, in which people of weak immunological barrier and ill people dwell becomes particularly important [8–10].

The above-mentioned phenomena influence the popularity of health resorts as well in Poland as abroad. The detailed pulmonological research shows the purposefulness of undertaking such cures in treatment of different types of respiratory ways diseases. In order to enable the effective therapy, specific environmental conditions, which occur in the underground subterranean therapy chambers and in overground health resorts, become essential [5, 10–13].

The main aim of the research was to characterize the fungal aerosol occurring in sanatorium chambers in Salt Mine in Bochnia and in rooms of overground health resort in Szczawnica, regarding the part of overground sanatorium in Szczawnica, taking the share of respirable fraction into consideration.

## Materials and methods

The research concerning the occurrence of fungal bioaerosol was carried out in the underground subterranean therapy chambers in Salt Mine in Bochnia and in sanatorium rooms in Health Resort Szczawnica. The first one, as well as the second sanatorium specializes itself in treatment of respiratory ways diseases, including lingering nose and throat inflammations, voice apparatus diseases, allergic diseases of respiratory ways and asthma. The healing factors are: the specific microclimate in both of the health resorts and saline aerosol floating in all underground chambers in Salt Mine in Bochnia [5, 11].

The measurements were carried out during the periods of presence of bathers, but the part of the measurements was performed in so called state of “primary cleanliness”, which is before entering of bathers and personnel into the sanatorium chambers (marked in the Tables as BK) and during the presence of bathers and performing therapeutic operations (marked as ZK).

Table 1 compares measuring points, in which measurements were carried out in underground and in overground sanatorium.

The air samples were taken using six-step Graseby-Anderson impactor, which was placed in the tested room on the height of 1.0–1.5 m over the floor to take the sample of bioaerosol from the respiratory zone (oral and nasal cavity position) of people. The microbiological analyses included evaluation of the general amount of fungi on the MEA medium (*Malt Extract Agar*, OXOID, Basingstoke, Great Britain).

The samples were incubated in thermostats in 30 °C for 4 days. The method was compatible with the Polish Standard PN-89/Z-04111/03 [14].

Table 1

## The description of measuring points

No.	Measuring points	Point description
Salt Mine in Bochnia		
1	B1	The Koldras Chamber
2	B2	The ramp between levels IV and VI
3	B3	The Wazyn Chamber – the gym
4	B4	The Wazyn Chamber – the bedroom
5	B5	The gangway behind the Wazyn Chamber (in the Sutoris shaft direction)
6	B6	Inside control (background – IC) – the gangway leading the air to the levels IV and VI of mine from the Trinitatis shaft
7	B7	Outside control (background – OC) – in the open air, next to the Trinitatis shaft
Sanatorium chambers in Szczawnica Health Resort		
8	S1	Nature Treatment Institute building – inside the room of mineral baths
9	S2	Nature Treatment Institute building – inside the room of the whirl massage of limbs
10	S3	Pump Room building – inside
11	S4	Inhalatorium building – inside the room of chamber inhalations
12	S5	Inhalatorium building – inside the room of individual inhalations
13	S6	Inside control (background – IC) Inhalatorium building – in the corridor leading to the rooms of the sanatorium bathers
14	S7	Outside control (background – OC) – in the open air, nearby the building of the Management of Przedsiębiorstwo “Uzdrowisko Szczawnica” SA

Legend: IC – inside control; OC – outside control.

The concentration of the tested aerosol was expressed as an amount of spores or mycelium hyphae fragments able to develop in the form of the colony – as colony forming units (cfu) present in one cubic meter [ $m^3$ ] of the taken air.

## Results and discussion

Fungal aerosol gets into the rooms mainly from the outside environment (soil, water, plants etc.), during the whole year its migration intensity varies, but in winter it is the lowest. In winter the soil is frozen and covered with snow – such conditions do not promote the natural emission of these microorganisms from soil. As a result, the concentration of fungal aerosol in the outer environment is usually lower than in flats. Penetration of fungal aerosols into interiors is the main reason for biological contamination of the interior environments [2, 4, 12].

The results, gained from the measurements of the fungal aerosol performed in two different types of sanatoriums – on as well as under the Earth surface, are presented in the Tables 2 and 3. In the tested sanatorium chambers in Salt Mine in Bochnia, the fungal aerosol occurred in values from 7 to 614 cfu in  $1 m^3$ , whereas in the sanatorium rooms in Szczawnica – from 0 to 706 cfu in  $1 m^3$  (Tables 2 and 3). The fungal aerosol

concentration in the external environment (outside background level) was from 282 to 3010 cfu in 1 m<sup>3</sup> and from 14 to 290 cfu in 1 m<sup>3</sup>, respectively.

Table 2

The number of airborne fungi in the underground chambers in Salt Mine in Bochnia in winter and spring 2008

Examination point	Winter 2008		Spring 2008	
	[number of colony forming units (cfu) in 1 m <sup>3</sup> of the air]			
	Patients absent – PA (17.02.08)	Patients present – PP (16.01.08)	Patients absent – PA (27.04.08)	Patients present – PP (20.05.08)
B1	7	56	77	565
B2	70	197	91	134
B3	99	133	105	566
B4	70	155	49	7
B5	35	126	56	614
B6 – IC	21	148	63	120
B7 – OC	282	346	311	3010

Legend: the same as for the Table 1.

The fungal aerosol concentration in sanatorium chambers in Bochnia was definitely lower (even 400 times lower) than in the external environment, and it was irrespective of the presence or absence of the bathers (Table 2). The situation was more complex in Szczawnica – during the two winter measurement sessions the amount of fungi in the sanatorium rooms was higher than in the examination point which was the outside background. Similar relationship was found during the spring measurements with present bathers. Different results were found during the spring measurements without the bathers, when the amounts in the sanatorium were a few times higher than outside (Table 3).

Table 3

The number of airborne fungi in rooms of overground sanatorium health resort Szczawnica in winter and spring 2008

Examination point	Winter 2008		Spring 2008	
	[number of colony forming units (cfu) in 1 m <sup>3</sup> of the air]			
	Patients absent – PA (23.01.08)	Patients present – PP (03.02.08)	Patients absent – PA (13.04.08)	Patients present – PP (14.04.08)
S1	154	233	120	63
S2	368	367	91	28
S3	310	165	71	706
S4	0	240	28	21
S5	14	452	28	21
S6 – IC	63	1575	35	226
S7 – OC	56	92	290	14

Legend: the same as for the Table 1.

In the sanatorium chambers in Salt Mine in Bochnia, there was found visibly inhibiting influence of the specific environment saturated with sodium chloride on the fungal spores. The air is pumped into the mine by the Trinitatis shaft, next to which the examination point – outside background (B7) is located. The air then flows to the B6 point, covering the distance of about 1000 m. In this part the fungal spores' concentration decreases even 25 times. Then, the amount of fungi spores increases along the way of the flowing air. This is undoubtedly caused by the dust collected on the floor of the chambers and corridors, which then is floating in the air. The influence of the bathers' presence on the amount of fungal spores in the air is ambiguous – in winter no changes in the abundance were observed, whereas in spring the amount of fungi in the chambers was a few times higher than in the inside control point.

On the basis of the results concerning the amount of fungal aerosol, it may be also concluded, that its highest values occurred in the underground sanatorium in Bochnia in Wazyn Chamber – in the gym (from 99 to 566 cfu in 1 m<sup>3</sup>) and in the gangway which leads the air from the Wazyn Chamber (from 35 to 614 cfu in 1 m<sup>3</sup>). The smallest amounts of the bioaerosol (7 cfu in 1 m<sup>3</sup>) were found in the state of so called “original sterility”, which is before entering the bathers into the sanatorium chambers in Koldras Chamber and in the bedroom part of the Wazyn Chamber – during the bathers' presence (also 7 cfu in 1 m<sup>3</sup>).

In the sanatorium rooms in Szczawnica, the highest amounts of fungi were found in the outside control point (Inhalatorium building – the corridor – from 35 to 1575 cfu in 1 m<sup>3</sup>) and inside the Pump Room building – from 71 to 706 cfu in 1 m<sup>3</sup>. The smallest amounts of the tested bioaerosol (7 cfu in 1 m<sup>3</sup>) were found in the Inhalatorium building, in the room of chamber inhalations (0 cfu in 1 m<sup>3</sup> – without the bathers and 21 cfu in 1 m<sup>3</sup> – with the bathers).

The comparison of the gained results with literature data is difficult because of the minute amount of the available publications as well as the differences in methods.

In normal conditions, the fungal aerosol concentration in rooms, in which people are present, is within the range of 10 to 1.000 cfu in 1 m<sup>3</sup> [15, 16], but according to the other microbiological analyses, the proposed highest admissible fungal aerosol concentration in flats and offices is defined as 500 cfu in 1 m<sup>3</sup> [17] or only in offices as 200 cfu in 1 m<sup>3</sup> [18].

It should be emphasized, that although many bathers were present in the sanatorium rooms in both of the health resorts, who moreover were intensively moving, the concentrations of fungal aerosol are generally lower than in the habitable rooms.

The analysis of the grain size distribution shows, that the share of the respirable fraction of the fungal aerosol in the sanatorium chambers in the salt Mine in Bochnia in the period between turns is significantly higher and is from 62.5 to 100 % (average 88.5 %) than during the treatment turn, when it ranges between 0 and 96.6 % (average 73.7 %) (Table 3). This means, that the concentration of the respirable fraction of the fungal aerosol does not depend on the amount of the patients who are present in the healing rooms. Average concentration of the fungal bioaerosol in sanatorium chambers in Bochnia amounts 81 % and is very close to the amount given by Pastuszka [4]. In the sanatorium rooms in Szczawnica, the share of the respirable fraction of the fungal

aerosol is related to the presence of the patients. When they are present, this fraction is on average 92.9 %, and when they are absent, it decreases to 79.5 % (Table 4).

Table 4

The proportion of respirable fraction of general number of fungi in the chambers in Salt Mine in Bochnia in winter and in spring 2008

Examination point	Winter 2008		Spring 2008	
	[%]			
	Patients absent – PA (17.02.08)	Patients present – PP (16.01.08)	Patients absent – PA (27.04.08)	Patients present – PP (20.05.08)
B1	100.0	87.5	100.0	82.7
B2	80.0	85.8	100.0	89.6
B3	100.0	73.7	66.7	52.5
B4	90.0	91.0	85.7	0.0
B5	100.0	77.8	62.5	96.6
B6 – IC	66.7	100.0	88.9	82.5
B7 – OC	70.2	91.9	54.3	67.6

Legend: the same as for the Table 1.

Table 5

The proportion of respirable fraction of general number of airborne fungi in the chambers in Szczawnica Health Resort Mine in winter and in spring 2008

Examination point	Winter 2008		Spring 2008	
	[%]			
	Patients absent – PA (17.02.08)	Patients present – PP (16.01.08)	Patients absent – PA (27.04.08)	Patients present – PP (20.05.08)
S1	72.7	97.0	94.2	88.9
S2	76.9	88.6	100.0	100.0
S3	86.5	95.8	90.1	98.0
S4	0.0	94.2	75.0	100.0
S5	100.0	100.0	100.0	66.7
S6 – IC	88.9	97.8	100.0	90.7
S7 – OC	100.0	92.4	100.0	50.0

Legend: the same as for the Table 1.

According to the same author, for the external environment, the share of the respirable fraction is usually 60 % in winter. In the research to the present paper, for the external environment in winter, higher share of the respirable fraction was found, and it was 71 % for Bochnia, and 85.6 % for Szczawnica, respectively.

Information about the concentration and distribution of the microflora particle sizes in the air in the chosen examination points allows to define their abilities to potentially influence the human organism [5, 16].

## Conclusions

1. The fungal aerosol concentrations in winter and in spring in sanatorium chambers in Bochnia were definitely lower than in the outside environment, and it was with the presence of the bathers as well as without them. In Szczawnica health resort, similar results were gained only during the absence of the patients.

2. Specific conditions in the sanatorium chambers in Salt Mine in Bochnia cause the decrease of the amount of fungal bioaerosol.

3. The proportional share of the respirable fraction of the fungal bioaerosol in the underground sanatorium was independent of the tourists' presence, but the opposite relation was found in the overground health resort.

4. The ascertained amounts of the fungal bioaerosol in each case fulfill the criteria of the Polish Standard, however they transgress the values suggested by the other authors several times.

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#### BADANIA AEROZOLU GRZYBOWEGO W SANATORIUM NADZIEMNYM I PODZIEMNYM

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**Abstrakt:** Lecznictwo sanatoryjne zarówno w Polsce, jak i za granicą jest bardzo rozpowszechnione. Jak wynika z wielu badań szczególnie w schorzeniach dróg oddechowych takie leczenie przynosi bardzo dobre efekty. Na te efekty w znaczący sposób wpływa specyficzny mikroklimat panujący zarówno w podziemnych komorach subterraneoterapii, jak i w pomieszczeniach sanatoriów naziemnych. Zasadniczym celem wykonywanych badań było oznaczenie i rozdział cząstek aerozolu biologicznego na frakcje (w zależności od ich średnic aerodynamicznych: powyżej 7,0 µm; 7,0–4,7 µm; 4,7–3,3 µm, 3,3–2,1 µm; 2,1–1,1 µm i 1,1–0,65 µm) występującego w komorach leczniczych w Kopalni Soli w Bochni oraz w pomieszczeniach sanatoryjnych Przedsiębiorstwa „Uzdrowisko Szczawnica”. Badania mikrobiologiczne powietrza zostały przeprowadzone w okresie zimy i wiosny 2008 r. w dwóch podziemnych komorach sanatoryjnych w Bochni oraz w pomieszczeniach sanatoryjnych w Szczawnicy. Pomiary zostały wykonane za pomocą sześciostopniowego aeroskopu Graseby-Andersena ze strefy oddechowej (tj. na wysokości jamy ustnej i nosowej) człowieka w stanie tzw. „pierwotnej jałowości”, tj. przed wprowadzeniem chorych i personelu do pomieszczeń sanatoryjnych oraz w czasie przebywania tam kuracjuszy. W wyniku przeprowadzonych analiz ilościowych stwierdzono znaczne zróżnicowanie aerozolu grzybowego na różnych stanowiskach pomiarowych. W czasie turnusów sanatoryjnych liczebności grzybów były znacząco większe w stosunku do tych, które występowały podczas nieobecności kuracjuszy – zarówno w sanatorium naziemnym, jak i podziemnym. Największe stężenia grzybów w Kopalni Soli w Bochni zanotowano w Komorze Kołdrasa oraz na boisku sportowym w Komorze Ważyn; z kolei w sanatorium w Szczawnicy największe stężenia bioaerozolu grzybowego stwierdzono w pijalni wód mineralnych.

**Słowa kluczowe:** uzdrowiska, grzyby, powietrze, kopalnia soli