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*Changes in the concentration of microbial aerosol
in the premises of the Jagiellonian University Museum during
the Night of Museums*

ABSTRACT

Changes in the concentration of microbial aerosol were examined in the selected premises of the Jagiellonian University Museum in Kraków, made available to visitors during the Night of Museums, i.e. Hall, Libraria and Treasury. The samples were collected four times using the collision method with MAS-100 air sampler. The concentration of mesophilic and heatolytic bacteria, molds, staphylococci and actinomycetes was examined and an attempt was made to find the correlation between the number of museum visitors and the concentration of microbial aerosol. The smallest number of visitors (600 persons) was recorded in Libraria, while the greatest (900 persons) in the Hall. The prevalence of microorganisms varied significantly between the hours of sampling, from the smallest number in the morning, after opening, to the greatest recorded while there were most visitors (9 p.m.). The largest number of molds was observed after the museum was closed (0:30). A strong positive correlation was found between the concentration of airborne mesophilic bacteria and actinomycetes with the number of visitors, the concentration of fungi was negatively correlated and there was a weak positive correlation in the case of staphylococci. On average, the greatest concentration of mesophilic bacteria (860 CFU/m³), actinomycetes (60 CFU/m³) and staphylococci (26 CFU/m³) was recorded in the Hall at 9 p.m. The largest concentration of molds (1,290 CFU/m³) was found in the Hall and Treasury at 0:30. The observed concentrations of airborne microorganisms do not exceed acceptable levels for public utility premises, therefore they do not pose a threat to the health of the Museum employees, tourists and to the condition of the Museum collections.

Keywords: bioaerosol, Jagiellonian University Museum, tourism, the Night of Museums

Introduction

Kraków is among most interesting touristic sites in Poland and as a city with rich history has many attractive places to visit by tourists, among which museums have a significant share. These institutions were established to collect and protect objects of historical value. One of those numerous museums in Kraków is the object of this study, i.e. Jagiellonian University Museum, situated in the Collegium Maius, nearby the Main Market Square in Kraków. Each year museums are visited by crowds of tourists. Several hundreds of people walking daily around the exhibition halls undoubtedly have impact on the quality of indoor air of museums, as every living organism is the source of microorganisms. It was demonstrated that in non-industrial indoor environments, the presence of human beings is one of the most important sources of airborne bacteria (Stetzenbach 1997). Concentrations of airborne microorganisms in occupied premises are higher compared to unoccupied conditions (Meadow et al. 2014) and people have been reported as a source of bacteria and fungi in settled dust samples (Niesler et al. 2010). Some of human activities, such as talking, sneezing, coughing, washing and toilet flushing can generate airborne biological particulate matter. Moreover, dust-covered monuments and museum artifacts, as well as damp or contaminated barrier constructions can be another source of airborne microorganisms, allergenic molds in particular (Buczyńska et al. 2007).

Microorganisms occur in air in the form of bioaerosols, composed of not only bacteria, fungi and viruses but also organic compounds of microbial origin, such as endotoxins, metabolites, toxins or other microbial fragments (Heikkinen et al. 2005). Indoor air is a very dynamic system in which particles of biological and non-biological origin are distributed and transported. It has been shown that bioaerosols contribute to about 5 to 34% of indoor air pollution (Srikanth et al. 2008). Among the threats related to the exposure to bioaerosols is the contact with pathogenic microorganisms. They occur in atmospheric air relatively rarely as compared to indoor air, where there are people or animals. Bioaerosols, through respiratory droplets, contribute to transmission of various bacterial diseases (e.g. pulmonary tuberculosis, tonsillitis, whooping cough), fungal diseases (surface and deep mycoses) or viral infections. Moreover, many airborne microorganisms are allergenic, in particular mold fungi, but also thermophilic actinomycetes or Gram-negative bacteria (Walczak et al. 2013).

Airborne microorganisms also secrete various types of chemical substances, such as endotoxins, enterotoxins, enzymes and mycotoxins. Endotoxins are compounds released by cell walls of Gram-negative bacteria and they may affect humans by causing fever, breathing problems, hypoglycemia or alter the number of leukocytes in blood (Gołofit-Szymczak, Skowroń 2005). Mycotoxins are secondary metabolites produced by fungi, particularly harmful to humans and animals. It is estimated that approximately 30% of health problems are associated with the exposure to mold fungi. Spores which they produce penetrate airways and deposit on the skin. Molds are the causes of various diseases such as sick building syndrome, dermatitis, allergies, respiratory diseases, mycotoxicoses and poisonings by volatile compounds. What is important, is that with the lack of indoor sources, the predominant source of airborne fungi indoors is the process of fungal aerosol migration from the outdoor air (Niesler et al. 2010). Mycotoxins are produced by

airborne species such as *Aspergillus flavus*, *Aspergillus parasiticus*, *Stachybotrys atra* or *Penicillium meleagrinum*. Inhalation of airborne mycelia fragments of mycotoxin-producing fungi leads to poisonings, dermatitis, diarrhea or immune mechanisms disorders. In addition, aflatoxins produced by *A. flavus* or *A. parasiticus* are compounds that proven to be carcinogenic (Jankowska, Pośniak 2007). The above described health effects, related to the exposure to microbiological aerosols, resulted in numerous studies on the concentrations of bioaerosol in museum premises (Niesler et al. 2010; Skóra et al. 2015; Zielińska-Jankiewicz et al. 2008) but there are no reports related to the number of their visitors or during particular events, when museums are visited by crowds of tourists.

Having regard to the above, the aim of this study was to assess the changes in the concentration of bacterial and fungal aerosol in the premises of the Jagiellonian University Museum, made available to visitors during the annual event – the Night of Museums. An attempt was also made to correlate the concentration of bioaerosols with the number of visitors.

Material and Methods

The study was conducted during the annual event, i.e. the Night of Museums, which takes place in many Polish cities every May. The samples of air were collected on May 15th 2015 in three rooms of the Jagiellonian University Museum, which were made available to tourists, i.e. The Hall, Libraria and Treasury. The examined premises do not have any ventilation or air conditioning system. Sampling was conducted four times during the day of the test – at 9 a.m. (before opening of the museum), 3 p.m. (when the museum was closed for the break after the morning round of visiting), 9 p.m. (2 hours after the museum was open for the visitors of the Night of Museums, during the expected “peak” of visitors) and at 0:30 a.m. (after the last visitors of the Night of Museums left the premises).

Air samples of 100 L were taken using MAS-100 (Merck) air sampler (Fig. 1). The measurements were conducted according to the procedure described in the PN-Z-04008-08:1989 standard, which means that the sampler was placed at c.a. 1.5 m, i.e. the human breathing zone and the volume of air collected was empirically adjusted to the expected concentration of microbiological aerosol. Types of applied media and incubation conditions are given in brackets. The number of the total mesophilic bacteria (Trypticase Soy Agar – Biocorp, 48 h at 36±1°C), haemolytic bacteria (5% sheep blood agar; Atlas, Parks 2004; 48 h at 36±1°C), mold fungi (Malt Extract Agar – Biocorp, 3–5 days at 24°C), actinomycetes (Gauze agar; Atlas, Parks 2004; 7 days at 24°C) and staphylococci (Chapman agar – Biocorp, 48 h at 36±1°C) was assessed. After incubation, the number of colonies characteristic for different microbial groups were counted and expressed as colony forming units per cubic meter of air (CFU/m³). The actual colony count per each culture plate was corrected according to the positive hole correction table (Operator’s manual MAS-100). All measurements were conducted in triplicates and the results are presented as means of those three replicates. Air temperature was measured with an electronic thermometer (Elmetron PT-105) during all samplings and the number of visitors was counted in the premises during the Night of Museums event.

Statistical analysis was performed using Statistica v.10 (StatSoft) – basic descriptive characteristics were calculated and a one-way ANOVA test (F) was employed to verify the significance of differences in the number of microorganisms between the hours of sampling and the differences between individual rooms of the museum. The Pearson coefficient of correlation (r) was calculated to assess the relationship between the concentration of microbial components of bioaerosol and the number of visitors during the Night of Museums.

Results and Discussion

The observed changes in the temperature were very small, i.e. the lowest temperature was recorded at 9 a.m. in The Hall (19.2 °C) and the highest (22.3°C) – in The Treasury. In most cases the temperature fluctuated around 20–21°C. The number of visitors differed between the sites – at 9 p.m. there were 600 persons in the Library, 730 in the Treasury and 900 in the Hall. After closure of the Museum, the number of visitors reached the total of 2,080.

As shown in Figure II, the mean concentrations of the tested microorganisms also ranged between the hours of sampling with the lowest values of all microorganisms recorded in the morning, before the museum was opened. Then the concentration of most microbial components of bioaerosol increased to their maximums reached at 9 p.m., when there were also most tourists visiting the museum premises. Only the number of mold fungi increased even more, to reach the maximum of 1,158 CFU/m³ of air at 0:30 a.m. The ANOVA test confirmed the significance of the observed differences between the hours of sampling (with values accordingly: actinomycetes $F=4.03$; molds $F=3.96$; mesophilic bacteria $F=10.49$, values significant at $p<0.05$) except from staphylococci, for which the F value was 1.79. When trying to assess the relationship between the number of the Museum visitors and the bioaerosol concentration, the highest values of the Pearson correlation coefficient were obtained for mesophilic bacteria and actinomycetes, i.e. $r=0.84$ and $r=0.90$ ($p<0.05$), respectively, indicating a strong positive correlation for these two groups of microorganisms. On the other hand, there was also very strong but negative correlation between the concentration of mold fungi and the number of visitors ($r=-0.99$, $p<0.05$). For the concentration of staphylococci the correlation was weak, with the r value of 0.23 ($p<0.05$).

Among the tested groups of microorganisms, mold fungi were the ones whose mean concentration was the highest in the Treasury and the Hall. In the Libraria hemolytic bacteria were the most numerous group (Fig. III). Analysis of variance indicated that the differences in the concentration of individual components of bioaerosol between different sampling sites were statistically significant in the case of actinomycetes (F value = 6.75) and mesophilic bacteria ($F=7.86$, values significant at $p<0.05$), whereas the differences for molds, staphylococci and hemolytic bacteria were not significant (F values of 1.65, 0.78 and 1.19, respectively, $p<0.05$).

When comparing the observed concentrations of mesophilic bacteria, actinomycetes and mold fungi with values proposed by the Expert Group on Biological Agents at the

Polish Interdepartmental Commission for Maximum Admissible Concentrations and Intensities for Agents Harmful to Health in the Working Environment (Skowroń, Górny 2014), it can be concluded that the numbers of microorganisms do not exceed acceptable levels for residential and public utility premises (i.e. 5,000; 200 and 5,000 CFU/m³ of air respectively for mesophilic bacteria, actinomycetes and molds). Nevertheless, issues related to the microbiological contamination of air in workplaces and public utility buildings have become of great interest, mainly due to the related threats. Contact with air containing fungal spores, and cells of bacteria and actinomycetes may cause allergies and infections of the upper and lower respiratory tract, as well as asthma (Grinshpun et al. 1997). However, Skóra et al. (2015) in their study conducted in four museums in Poland on the concentrations of airborne bacteria and fungi recorded fungal values between less than 100 CFU/m³ to more than 10,000 CFU/m³ of air and bacterial values ranging from c.a. 500 CFU/m³ to c.a. 800 CFU/m³. This indicates that the mean values recorded in our examinations are comparable to or lower than those recorded in the mentioned study. The concentration of fungal aerosol detected by Zielińska-Jankiewicz et al. (2008) in their study conducted in libraries and archive storage facilities ranged from the minimum of 60 CFU/m³ to the maximum of 2,900 CFU/m³ in archive storage facilities and from 60 to 1,800 CFU/m³ in libraries. Mean concentrations of fungal aerosol in the mentioned study ranged from 230 to 630 CFU/m³ in archive storage facilities and from 180 to 830 CFU/m³ in libraries. The mean values obtained in our study are smaller than the ones obtained by the mentioned authors.

Differences in values obtained by various researchers may result from the fact that the bioaerosol levels depend on a number of factors, including the room furnishings, building materials or microbiological contamination of walls and ceilings, which can be a significant cause in the case of old buildings (Mandal, Brandl 2011). Another important source of bioaerosol indoors are their occupants and people staying there temporarily (Loftness et al. 2007). Indoor bioaerosol levels may also increase due to outdoor sources, when particles are transferred through windows and doors. This might be the reason for the negative correlation between fungal aerosol concentration and the number of the Museum visitors, observed in our study, since according to Niesler et al. (2010) in the case of lack of internal sources, outdoor air migration is the main source of indoor fungal aerosol. On the other hand, the presence of airborne bacteria in non-industrial indoor environments is mostly related to the presence of people (Mandal, Brandl 2011), which also proves to be the case in our study, as the highest concentrations of these microorganisms were recorded when the Museum was visited by the greatest number of tourists. Among other factors that should be taken into consideration while analyzing or predicting the indoor bioaerosol concentrations there are building heating, ventilation and/or air conditioning which affect the temperature and relative humidity of the premises (Stetzenbach et al. 2004).

Conclusion

The concentration of microbial components of bioaerosol in the studied premises of the Jagiellonian University Museum differed depending on the group of analyzed microorganisms with the lowest values observed for staphylococci and the highest for mold fungi. The highest concentrations of all microorganisms was recorded when the Museum was visited by the highest number of tourists, except fungi, whose number increased until the last measurement. These differences may result from different sources of the analyzed bioaerosol components, i.e. people visiting the museum for bacteria and outdoor air in the case of molds. The highest mean concentrations of molds, mesophilic bacteria and actinomycetes were recorded in the Hall. None of the concentrations exceeded the threshold values set for public utility premises.

Although it is a widely known fact that contact with fungal spores, thermophilic actinomycetes or bacterial endotoxins can cause allergies or upper respiratory tract infections in people exposed to microbial aerosol, the conducted study shows that in the studied museum premises the risk is not elevated. However, more detailed study, including determination of particle size and the predominant microbial species, would be valuable in more precise evaluation of health hazards for both museum visitors and its employees.

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