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Airborne Microorganisms Fluctuations Over the Gulf of Gdansk Coastal Zone (Southern Baltic)

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Abstract

A one-year record of airborne microorganisms content in the region of the seaside resort in Sopot and in a few locations along the coastline of the Gulf of Gdansk are presented. The air samples were collected by means of filtration method using an apparatus produced by Sartorius. In addition, several investigations were performed to compare the results of three sampling methods: sedimentation method according to Polish Regulation [1], filtration, and impaction.

The total number of mesophilic, psychrophilic bacteria and fungi concentrations in air were determined. Between January and December 1998 the number of mesophilic bacteria in air varied from a few to 308 CFU/m^3 (CFU-Colony Forming Units), while the number of psychrophilic bacteria varied from 1 to 190 CFU/m^3. The respective fungi content varied from 5 to 1100 CFU/m^3. The monthly and seasonal geometrical mean values were calculated showing that the maximum seasonal occurrence of studied groups of microorganisms were observed at different times.

Keywords: aerosol, microorganisms, airborne particles.

Introduction

Despite the fact that atmospheric air does not favour growth of microorganisms due to lack of nutrients, the microorganisms are present in aerosol form, suspended in the air. The basic sources of microbes are soil, water, animals and humans. The bioaerosol consists of different types of microorganisms usually stuck to the particles of dust or suspended in tiny drops of water. Among them pathogenic viruses, bacteria and fungi, capable of causing human allergies and skin disease are present [2].

The survival time of microorganisms in bioaerosol decreases at low contents of humidity and high UV radiation. Therefore, the number of microorganisms in the air fluctuates during the year. The impact of UV radiation on the airborne microbes is inversely proportional to dustiness of the air.

The basic process of self-purification of air is sedimentation of bioaerosols. However, pigmentary and sporulating bacteria as well as spores of fungi can be carried by the wind for long distances. For instance it was proved that spores of *Bacillus* bacteria were transported with a mass of air for a distance of 1800 km [3].

Due to numerous reports there are up to 100 species of microorganisms, mostly saprophites, that are present in the air. Most common are *Micrococcus* and *Sarcina* bacteria, forming yellow or orange-coloured colonies when grown on nutrient agars, aerobic sporulating bacteria of *Bacillus* species and fungi [4].

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In Poland previous studies were limited to the areas surrounding wastewater treatment plants or landfills and focused on assessment of environmental impact of WWTPs and landfills on microbiological quality of air [5, 6, 7, 8, 9]. Only a few published studies concerned evaluation of seasonal fluctuations of the number of microorganisms in cities, for instance in Szczecin [10] and Krakow [11]. In the majority of the published studies the number of microorganisms was determined by means of sedimentation method, assuming gravitational settling of microorganisms on the surface of nutrient media.

Another poorly investigated task is evaluation of occurrence of microorganisms in the air over marine coastal zones. Results of investigations carried out by Shaffer and Lighthart [12], who used filtration method (with induced air flow) for samples collection, indicated that the number of bacteria was several times lower in the coastal zone (103 CFU/m^3) than in the city (609 CFU/m^3).

Progress in elucidation of processes influencing the release of microorganisms from water and the formation of marine aerosol followed publication of the results of laboratory investigations conducted by Blanchard and Syzdek [13]. Authors proved that each drop formed after bursting of individual air bubbles become enriched in bacteria. They showed that the enrichment of *Serratia marcescens* bacteria in droplets (ejected by bubbles migrating in the water column more than 10 cm) could exceed 600 times the concentration of bacteria in water. The authors found out that enrichment of drops with bacteria is directly proportional to the distance of migration of maternal bubble in the surface water. It may be assumed that there exists a relationship between the number of microorganisms in the air and the level of microbiological contamination of marine water, especially under moderate or strong air-sea interactions. For instance, in August 1995 and 1996, in the air above the mixing zone of strongly bacteriologically contaminated waters of the Vistula River and marine waters, the higher number of mesophilic bacteria was measured than in other sampling points located near Gdynia and Sopot [14].

Conditions that especially favour marine aerosol formation are incorporated with wave breaking areas as well as with periods when the water is supersaturated with dissolved gases. Since drops of marine aerosol may contain bacteria, potential migration of bacteriological contamination may impose a sanitary risk.

Bioaerosols in outflows from wastewater treatment plants are important factors influencing the air quality. In the Three-City area there are two large wastewater treatment plants: Gdansk-Wschod and Gdynia-Debogorze, of 100 000 m^3 capacity per day and 87 000 m^3/d, capacity. The outflows from both wastewater treatment plants to the Gulf of Gdansk are bacteriologically polluted (with concentration of E.coli/l00ml from 10^5 to 10^6) and are potential reservoirs of pathogenic bacteria [15, 16]. Hence, the watering places located in the Gulf of Gdansk areas influenced by the outflows have been closed for many seasons because of low sanitary quality of water. In addition the mixing zone of fresh and saline waters in the mouth of the Vistula River maintain all year round poor sanitary quality as controlled in Kiezmark [17].

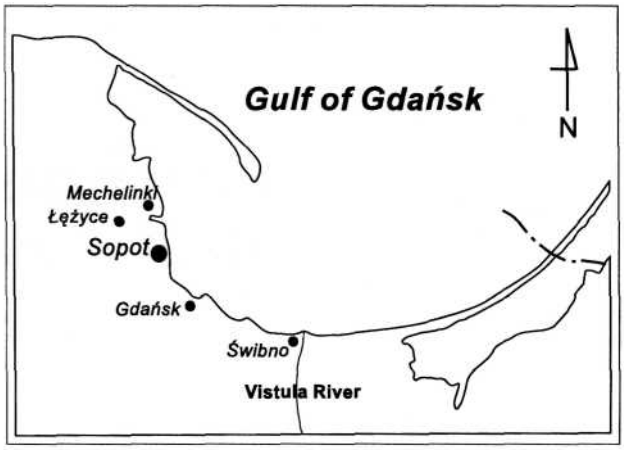

Fig. 1. Location of sampling points.

Current studies aim at bioaerosols transfer from water to the air in the coastal zone of the Gulf of Gdansk. The purpose of the study was to evaluate the possibility of carrying away microbiological pollutants with aerosols from marine waters to the air, especially in the neighbourhood of wastewater treatment plant outflows. Another task undertaken was comparison of different sampling methods on the recovery of microorganisms.

Materials and Methods

Bioaerosols were sampled in several coastal locations along the Gulf of Gdansk in Sopot, Mechelinki, Gdansk and Swibno from 1998 to 2001. Locations of sampling points are presented in Fig. 1. In addition, a set of meteorological data on wind speed and direction, air temperature, humidity, and the rate of precipitation were collected.

The sampling station in Sopot was located in the Northern Bathes Building, approximately 300 m from the shore, 10 m above sea level. The numbers of microorganisms in the air were measured out from January to December 1998. The total number of 198 samples were collected, 4 or 5 times a week, 14-24 measurements each month, apart from March (Table 1).

Table 1. The number of sampling days in subsequent months of 1998.

Months	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
The number of sampling days	19	15	–	16	16	14	16	16	23	24	21	18

Table 2. The ratio of the number of mesophilic bacteria (BM) to psychrophilic bacteria (BP) in air collected in subsequent months in 1998.

Months	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII
BM / BP	0.31	0.39	–	0.20	0.24	0.47	0.33	0.41	0.64	0.69	0.76	0.67

Sterilized gelatinous Sartorius filters, SM 12602-080 ALK (collection efficiency of 99.9%), were used for air filtration and incubated on nutrient agar. Then the number of mesophilic bacteria (incubated at 37°C), total number of psychrophilic bacteria (incubated at 20°C), and spores of fungi according to Polish Regulation [18] was determined. The results were calculated as colony forming units (CFU) in a cubic meter of air.

In the period 2000-2001 a complementary series of measurements were performed in order to find out the influence of different sampling methods on the detected number of microorganisms. In each of five sampling points, at the same time and on the same elevation, a filtration sample and sedimentation on the surface of selected nutrient medium were performed. Altogether 84 measurements comparing sedimentation and filtration method as well as 84 measurements comparing sedimentation and impaction methods were performed. As impaction method a Merck MAS-100 air sampler for standard Petri dishes with integrated anemometer for correct and reproducible results was used. MAS-100 sampler works based on the Andersen sampler principles [19]. The investigations were carried out on the communal beach in Sopot, on the beach in Gdansk, in Mechelinki, close to the outflow from the wastewater treatment plant Debogorze, and around a communal landfill in Lezyce (Fig- 1).

The number of microorganisms was determined by means of culturing them on nutrient media. The samples of air were collected by means of sedimentation, filtration and impaction method. In the sedimentation method according to PN-89/ Z-04008/08 the microorganisms settled from the air directly on the surface of nutrient medium were cultured. In that case an apparatus Sartorius MD8 was used, a known volume (0.5 or 1 m^3) of air was filtered through sterile gelatinous filters. The filters were then put on adequate culture media. In the impaction method air sampler type MAS 100 Eco™ produced by Merck was used. The desired volume of air was impacted on to the head of the device, where a Petri dish with culture medium adequate for each group of microorganisms was previously placed. Above methods were compared with air filtration method.

Discussion of the Results

The results of microbiological measurements performed near the beach in Sopot are presented as monthly mean geometrical values in Figs. 2-5. The number of mesophilic bacteria varied in range from 2 to 308 CFU/m^3 (Fig. 2), while the number of psychrophilic bacteria changed from 1 to 190 CFU/m^3 (Fig. 3). The highest mean concentration of psychrophilic bacteria, equal to about 50 CFU/m^3, was observed in the period from April to May, and then gradually decreased, reaching its minimum of 2 CFU/m^3 in December. The highest mean values of mesophilic bacteria, equal to 15-16 CFU/m^3 (about three times less than psychrophilic bacteria) were observed in September-October. In each month the mean number of mesophilic bacteria was lower than the corresponding value of psychrophilic bacteria. The highest numbers of mesophilic bacteria (308 CFU/m^3 and 98 CFU/m^3) were observed on the days with easterly winds, which was responsible for carrying away the microorganisms from the zone where the Vistula River and Gulf of Gdansk sea waters mixed.

The colony forming units ratios of mesophilic bacteria (BM) to psychrophilic bacteria (BP) in subsequent months in 1998 are presented in Table 2. In January to August the BM/BP changed from 0.2 (in April) to 0.47 (in June), which indicates that in air psychrophilic bacteria dominated. From September to December BM/BP

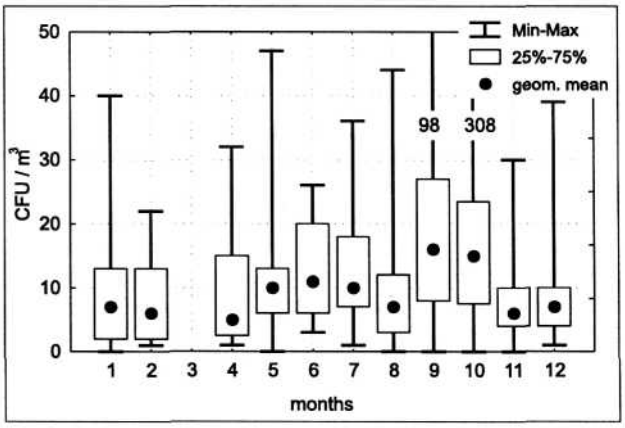

Fig.2. The number of mesophilic bacteria in 1 m^3 of air in subsequent months in 1998.

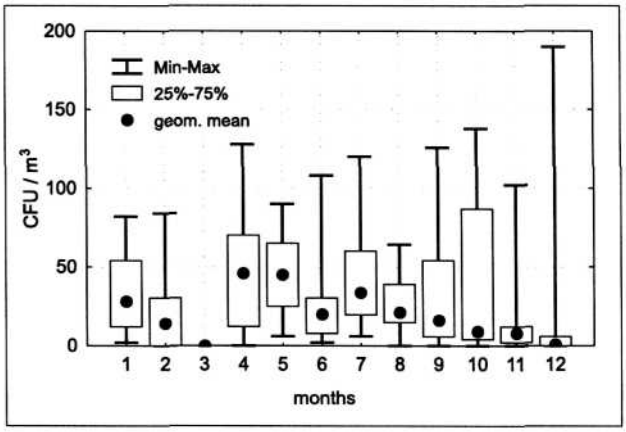

Fig. 3. The number of psychrophilic bacteria in 1 m^3 of air in subsequent months in 1998.

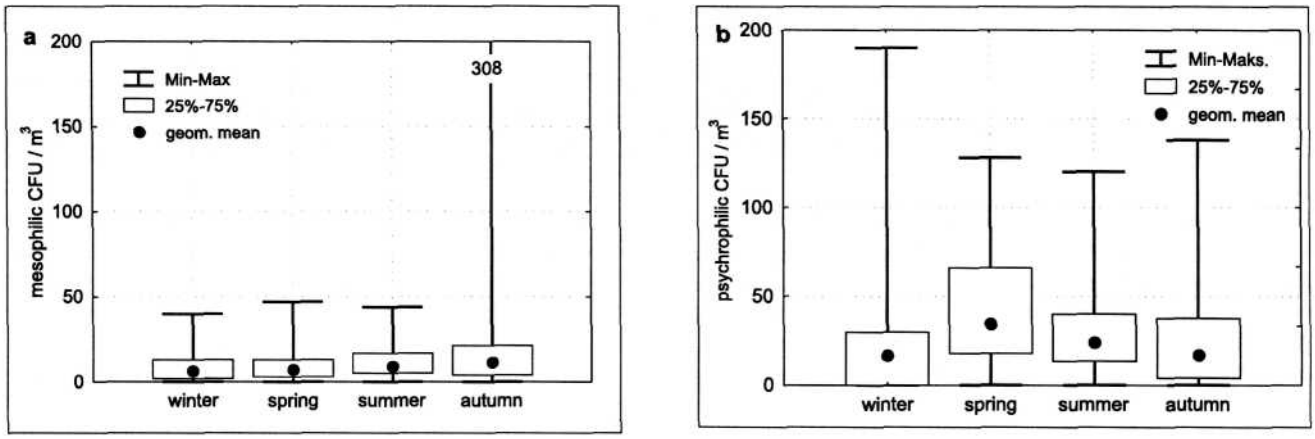

Fig. 4. Seasonal fluctuations of the number of mesophilic (a) and psychrophilic (b) bacteria in 1 m^3 of air in 1998.

were displayed by higher values range from 0.64-0.76, which was probably due to depletion of environmental bacteria (especially in November and December).

Fig.4 illustrates the results for both groups of bacteria in subsequent seasons. For each season, the widest range of 50% of results for psychrophilic bacteria was observed in spring and varied from 18 to 66 CFU/m^3. The number of mesophilic bacteria in subsequent seasons changed from 2 to 21 CFU/m^3.

The number of fungi in aerosols changed from a few to 1100 CFU/m^3 (see Fig. 5). In the period from July to August, the mean geometrical values reached maximum of 380-500 CFU/m^3 (in June approximately 200 CFU/m^3), while the lowest values, equal to 10-25 CFU/m^3 were observed from December to April (see Fig. 5a). The comparison of the mean geometrical values in different seasons showed that the number of fungi was highest in summer, reaching 300 CFU/m^3. The lowest geometrical mean value was observed in winter (see Fig. 5b).

In general the results of microbiological analyses indicate that among investigated airborne microorganisms the most numerous were fungi. The content of microorganisms in the air in the coastal zone of the Gulf of Gdansk in 1998 indicate some tendencies of fluctuations. The highest geometrical mean values of mesophilic bacteria (BM), psychrophilic bacteria (BP) and fungi were equal to 16 CFU/m^3, 50 CFU/m^3 and 500 CFU/m^3, respectively. The maximum of occurrences for each group of microorganisms differ: for BM it was September-October, for BP April-May and for fungi July-August.

In the period 2000-2001 several series of investigations of air were conducted in order to compare three methods of sample collection (see Fig. 6). The obtained concentrations of each group of heterotrophic microorganisms determined by means of sedimentation and filtration methods were calculated. The number of microorganisms observed when the filtration method was used were on average 7 times lower than in the case of sedimentation method, while the microorganisms detected by means of impaction method were 14 times lower compare to

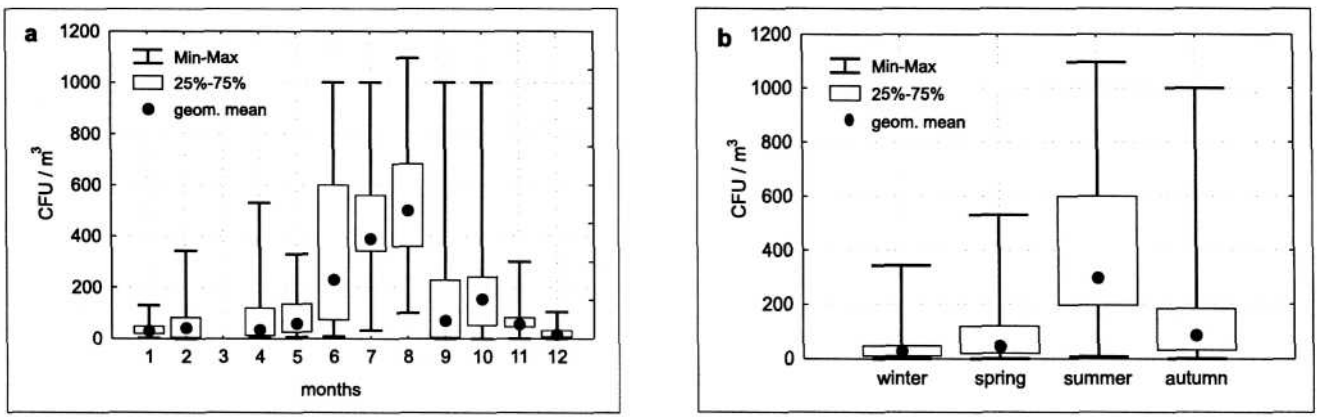

Fig. 5. a) The number of fungi in 1 m^3 of air in subsequent months of 1998, b) Seasonal changes of the number of fungi in 1 m^3 of air in 1998.

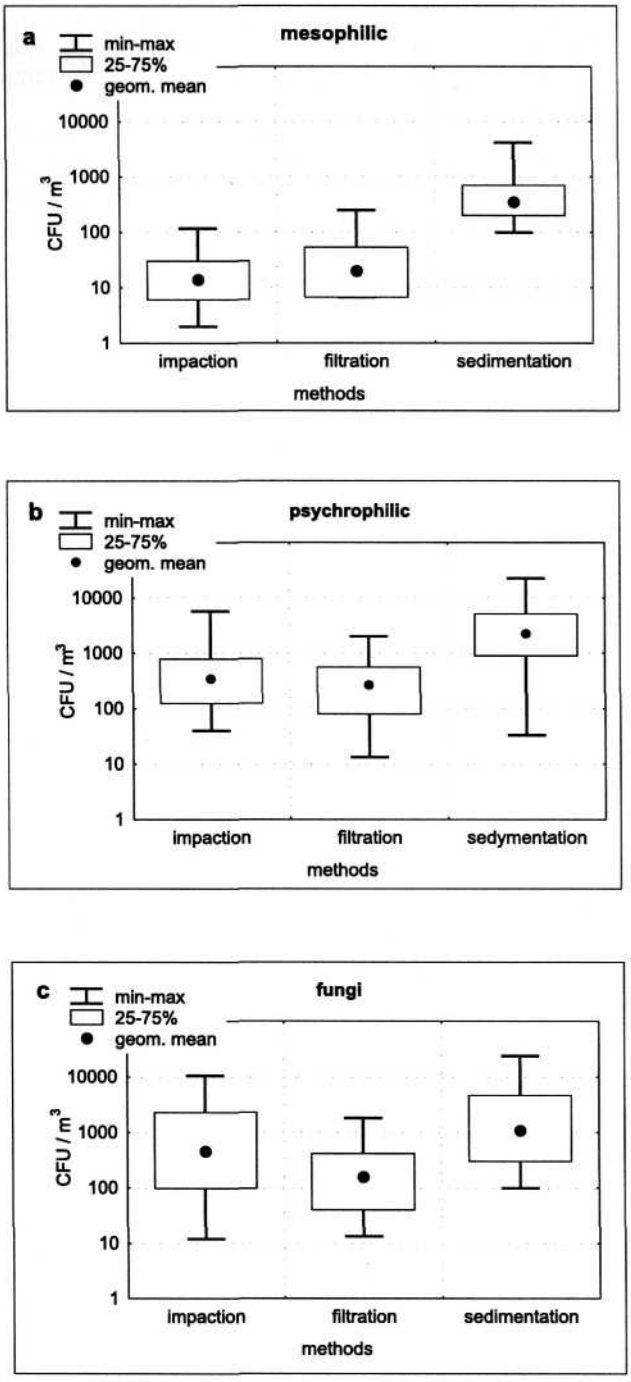

Fig. 6. Comparison of the of mesophilic (a), psychrophilic (b) bacteria and fungi (c) CFU measured by means of impaction, filtration and sedimentation methods.

sedimentation method. This accounts for the relatively low but more accurate numbers of microorganisms observed during the one-year investigations in Sopot, in comparison to published studies using sedimentation method of sample collection, which is more appropriate for indoor sampling.

The number of mesophilic and psychrophilic bacteria in one cubic meter of air, measured by means of filtration and impaction methods are similar. However if sedimentation method was used, the average numbers of CFU/m^3 were by an order of magnitude higher than in the other methods.

The number of fungi was less sensitive for the choice of sampling method. The highest results for impaction method and sedimentation methods were equal to 10 500 CFU/m^3 and 23 500 CFU/m^3, respectively, while the lowest values were obtained when using filtration.

Conclusions

The performed investigations on the airborne microorganism content over the marine coastal region allow us to verbalize the following conclusions.

1. Seasonal fluctuations of the number of microorganisms present in the air were more pronounced for psychrophilic than for mesophilic bacteria:
- the maximum for psychrophilic bacteria were found in April-May, while the minimum occurred during the winter months,
- the maximum of mesophile bacteria were found in September-October,
- the elevated concentrations of fungi occurred in spring;
- the most abundant microorganisms in the Gulf of Gdansk coastal air are fungi.

2. The method of sampling proved to be critical for the recovery of microorganisms from aerosol. When sedimentation method was used, the number of detected microorganisms was by an order of magnitude higher than in the case of impaction and filtration methods, thus the filtration method is recommended as more accurate due to the strict control of the sampled air volume.

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