Microorganisms in the Air Over Wastewater Treatment Plants

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Abstract

We present the results of investigations of microorganisms present in aerosol emitted by selected devices of two mechanical-biological wastewater treatment plants (“Debogorze” and “Gdańsk-Wschód”). In 2002, 5 series of measurements were conducted at each of the plants. The samples of air were collected by means of sedimentation method (PN-89/Z-04008/08) and by means of filtration method, using an air sampler produced by Merck.

Apart from measurements of microbiological analyses, consisting of determinations of the total number of psychrophillic and mezophillic bacteria, manitol-positive and manitol-negative staphylococcus, hemolyzing staphylococcus, *Pseudomonas fluorescens*, coliform bacteria and the number of spores of phycomycetes, weather conditions were also monitored.

The most numerous microorganisms in the monitored air were psychrophillic bacteria and spores of phycomycetes. The number of psychrophillic bacteria varied in the wide range from 14 to 5255 CFU/m$^3$, the number of mezophillic bacteria changed from 1 to 1324 CFU/m$^3$, the number of staphylococcus – from 1 to 150 CFU/m$^3$, the numbers of *Pseudomonas fluorescens* and the coliforms varied from a few to 50 CFU/m$^3$ and the spores of phycomycetes varied from a few to 5,250 CFU/m$^3$ of air (collected by means of Merck air sampler).

It was found out that in both plants the main sources of microorganisms were sludge recirculation chambers and sand traps. The air around aeration chambers of activated sludge contained similar numbers of microorganisms as the background. It was proved that the sampling method is of critical effect on measurement results. The number of microorganisms detected in the samples collected by means of sedimentation method was by one order of magnitude higher than in the samples collected by means of filtration method (using air sampler), allowing for strict control of the filtered air volume.

Keywords: aerosol, microorganisms, wastewater treatment plant.

Introduction

Despite the fact that atmospheric air does not favour growth of microorganisms due to lack of nutrients, the microorganisms are suspended in the air in an aerosol form. 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 viruses, pathogenic bacteria and fungi, capable of causing humans allergies and skin disease are present [1]. The process of aerosol spread is influenced by a number of atmospheric factors such as temperature, wind velocity and specific humidity [2]. Survival time of microorganisms in the aerosols depends on various environmental factors such as UV radiation or lack of moisture.

Wastewater treatment plants are considered potential sources of airborne pathogenic microorganisms. Bioaero-
sols are generated from bursting bubbles produced by the aeration system, which provides oxygen for biodegradation processes [3]. The number of psychrophilic bacteria varied in a wide range from $1.4 \times 10^2$ CFU/m$^3$ - sedimentation method [4] to $1.2 \times 10^5$ CFU/m$^3$ - impaction method [5] for the samples of air collected near aeration chamber. As raw wastewater is also a potential source of pathogenic microorganisms, inhalation, contact and ingestion can endanger human health through water or air pathways. Different authors have reported the incidence of special form of illness among workers in sewers and wastewater treatment plants [6]. The transfer of the microorganisms from water to air occurs mainly during aeration, when the bubbles reach the surface they burst and little film drops are ejected above the surface [7]. Blanchard and Syzdek [8] in their experiments proved that bacterial concentrations in the drops ejected from bubbles could exceed 600 times the concentration of bacteria in water source, depending on the drop size.

Bioaerosols emitted by wastewater treatment plants can strongly influence air quality. There are two large wastewater treatment plants (WWTPs) in the Three-city area, “Debogorze” and “Wschod”, receiving 60,000 m$^3$/d and 93,000 m$^3$/d, respectively. Multiphase activated sludge systems are working in both WWTPs. Treated sewage is discharged to marine coastal waters. The outflows from both wastewater treatment plants to the Gulf of Gdansk are still bacteriologically polluted (with concentration of $E. coli$/100ml from $10^5$ to $10^6$) and are potential sources of pathogenic bacteria [9,10]. The objectives of our study were to investigate the emission of bacteria and fungi from the different wastewater treatment plants into the atmosphere and find out the relationship between sampling points and the number of bioaerosols.

### Materials and Methods

In the article the results of investigations of microorganisms present in aerosol emitted by selected devices of two mechanical-biological wastewater treatment plants (“Debogorze” and “Wschod”) are presented. The samples of air were collected at four sampling points: background – sampling point located outside the WWTP area (windward), sand trap, sludge recirculation chamber and aeration chamber. Bioaerosols were sampled in spring 2002, 5 series of measurements were conducted at each of the plants. Locations of sampling points are presented in Fig.1. In addition, a set of meteorological data on wind speed, direction and air temperature were collected.

The samples of air were collected by means of sedimentation method (SM) and impaction method (IM). In the sedimentation method, according to Polish Standards (PN-89/ Z-04008/08) [11], the microorganisms settled

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**Table 1. Evaluation of microbiological air pollution according to Polish Regulation [11] (PN-89/ Z 0411/02 and 03).**

<table>
<thead>
<tr>
<th>Type of microorganisms</th>
<th>Number of microorganisms in m$^3$ of atmospheric air</th>
</tr>
</thead>
<tbody>
<tr>
<td>psychrophic bacteria</td>
<td>$&lt; 1\ 000$</td>
</tr>
<tr>
<td><em>Pseudomonas fluorescens</em></td>
<td>$1\ 000 - 3\ 000$</td>
</tr>
<tr>
<td></td>
<td>$&gt; 3\ 000$</td>
</tr>
<tr>
<td>hemolytic staphylococci</td>
<td>No</td>
</tr>
<tr>
<td></td>
<td>$&lt; 50$</td>
</tr>
<tr>
<td></td>
<td>$&gt; 50$</td>
</tr>
<tr>
<td>spores of fungi</td>
<td>$&lt; 5\ 000$</td>
</tr>
<tr>
<td></td>
<td>$5\ 000 - 10\ 000$</td>
</tr>
<tr>
<td></td>
<td>$&gt; 10\ 000$</td>
</tr>
<tr>
<td>Level of air pollution</td>
<td>not polluted</td>
</tr>
<tr>
<td></td>
<td>medium pollution</td>
</tr>
<tr>
<td></td>
<td>strong pollution</td>
</tr>
</tbody>
</table>

The investigations consisted of the following determinations:

**Microorganisms**

- mesophilic bacteria
- coliform bacteria
- staphylococci:
  - mannitol-positive and mannitol-negative hemolytic bacteria
- psychrotrophic bacteria
- *Pseudomonas fluorescens*
- spores of fungi

**Culture media / incubation temperature and time**

- Nutrient Agar / 37°C 24-48h
- Chromocult Coliform Agar / 37°C 24-48h
- Chapman Agar / 37°C 24-48h
- Blood Agar / 22°C 72h
- Nutrient Agar / 22°C 72h
- King B / 26°C 120h
- Sabouraud Agar / 22°C 24-48h
from the air directly on the surface of nutrient medium were cultured – sedimentation time was 10 min. In the impaction method air sampler type MAS 100 Eco produced by Merck was used. The desired volume of air was impacted onto the head of the device, where a Petri dish with culture medium adequate for each group of microorganisms was previously placed. The air flow was 6 m³/h and the sampling volume was between 0.2 and 0.5 m³.

According to Polish Standards, sanitary air pollution is evaluated on the basis of the number of indicator bacteria and spores of fungi (Table 1).

**Results and Discussion**

Graphical presentation of measurement results of the number of psychrotrophic bacteria, mesophilic bacteria and spores of fungi are presented in Figs. 2, 3 and 4.

Results were compared to the background numbers of bacteria measured at the sampling point located outside the WWTP, winward. Total number of mesophilic bacteria fluctuated from 1 to 1324 CFU/m³ (impaction method – IM) and from several tens to 9430 CFU/m³ (sedimentation method – SM) (Fig.2). The number of psychrotrophic bacteria varied in a wide range from 14 to 5255 CFU/m³ (IM) and from 157 to 18250 CFU/m³ (SM) (Fig.3), the numbers of that bacteria were similar to the results of the other authors [4,5]. The numbers of psychrotrophic and mesophilic bacteria in samples collected close to the same devices at each WWTP were similar. Considerable increase of both groups of bacteria quantity in air (in comparison to the background) was detected in the samples collected near sand traps – the devices where raw sewage is flowing. Enhanced emission of bioaerosol was also observed in the air near activated sludge recirculation chambers. However, it was approximately 65% lower than near the sand traps.

The number of fungi spores in bioaerosols collected by means of impaction method at both WWTPs varied from a few to 5250 CFU/m³ (IM). In samples collected by means of sedimentation method (SM), it varied from several tens to 12250 CFU/m³ (Fig.4). Any relationship between the number of fungi and location of sampling point at the WWTP was not found out. The highest values
were measured outside the WWTP areas. It was found that method of sampling affected the results – usually the numbers of fungi spores in air samples collected by means of sedimentation method were 6 times higher than in samples collected by means of impaction method.

On the basis of the numbers of psychrotrophic bacteria and fungi spores (in samples collected by means of sedimentation method) the air around selected wastewater treatment devices can be classified as medium polluted and strongly polluted.

Results of measurements of potentially pathogenic bacteria numbers (staphylococci, coliforms, \textit{Pseudomonas fl.}) in air samples collected by means of impaction method are presented in Figs. 5 and 6. Maximal numbers of bac-

![Fig. 4. Comparison of the number of fungi spores in air samples collected in sampling points located at the WWTPs Debogorze (D) and Wschod (W): a) impaction method – IM; b) sedimentation method – SM.](image1)

![Fig. 5. Comparison of the numbers of bacteria in samples collected by means of impaction method in sampling points located at the WWTPs Debogorze (D) and Wschod (W); a) hemolytic staphylococci; b) mannitol-positive and mannitol-negative staphylococci.](image2)

![Fig. 6. Comparison of the numbers of bacteria in samples collected by means of impaction method in sampling points located at the WWTPs Debogorze (D) and Wschod (W); a) \textit{Pseudomonas fluorescens}; b) coliform bacteria.](image3)
teria were present near the sand traps, which turned out to be the largest bacteria emitters in both WWTPs. The number of hemolytic staphylococci determined by means of impaction method fluctuated from 0 to 70 CFU/m³, while the number of mannitol-positive and mannitol-negative staphylococci varied from 0 to 154 CFU/m³. Another substantial source of staphylococci was activated in sludge recirculation chambers.

In the air around sand traps and activated sludge recirculation junctions the highest average number of P. fluorescens (20 and 7 CFU/m³, respectively) and coliforms (132 and 17 CFU/m³) was detected (Fig.6). On the basis of these values the air around these sampling points can be classified as medium polluted. The number of bacteria from both groups around aeration chambers was from 0 to 4 CFU/m and was close to the background values. The air outside WWTP areas contained maximally 8 CFU/m³ of P. fluorescens and faecal coli.

Results of microbiological analyses of the air over the WWTP areas presented in Figs. 2-6 indicate that number of emitted microorganisms depending on type of wastewater treatment device. This is confirmed by observations of Ossowska-Cypryk [12], who found out that the quantity of emitted bioaerosol is related to type of sewage and type of microorganisms present in sewage, the type of aeration system (turbine aerators, Kessner aerator, perforated pipes) and aeration method (large, medium or small air bubbles). The number of microorganisms in the air around aeration chambers was similar to background values (outside the WWTP area). The typical indicator of microbiological air pollution over wastewater treatment plants were Pseudomonas fluorescens and coliform bacteria.

In our study it was confirmed that sampling method affects the results of measurements of the number of microorganisms in the air. In the years 2000-01 several measurement series were carried out in order to compare different methods of air sampling. The ratio of the number of some groups of heterotrophic microorganisms determined by means of sedimentation and filtration method was calculated. The numbers of microorganisms measured by means of impactation method were on average 14 times lower than in the case of sedimentation method [13]. The ratios of microorganism numbers obtained by means of different sampling methods are presented in Table 2 (the ratios were calculated on the basis of 40 measurement series carried out in “Debogorze” and “Wschod” WWTPs).

Table 2. Ratios of microorganism numbers determined by means of sedimentation method (SM) and impaction method (IM).

<table>
<thead>
<tr>
<th>Microorganism Type</th>
<th>SM / IM</th>
<th>Psychrotrophic bacteria</th>
<th>Spores of fungi</th>
<th>Hemolytic Staphylococci</th>
<th>Mannitol + - Staphylococci</th>
<th>Coliforms</th>
<th>Ps. fluorescens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mesophilic bacteria</td>
<td>35</td>
<td>25</td>
<td>6</td>
<td>29</td>
<td>18</td>
<td>32</td>
<td>21</td>
</tr>
<tr>
<td>Psychrotrophic bacteria</td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

The effect of sampling method on measurement result was smallest in the case of fungi spores (SM/IM 6) and the largest in case of mesophilic bacteria (total number, coliforms, staphylococci – SM/IM from 35 to 29). Such large differences result from different volumes of analyzed air samples. In samples collected by means of sedimentation method particles of bioaerosol settle gravitationally. This process is interrupted by wind or moisture. In the impaction method the volume of filtered air is strictly controlled, resulting in more accurate measurement of the number of bacteria in the open air.

Conclusions

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

- The air over wastewater treatment plants Debogorze and Wschod is medium microbiologically polluted according to Polish Standards.

- The largest emitters of bioaerosols at WWTPs are sand traps and activated sludge recirculation junctions.

- The number of microorganisms in the air around aeration chambers is similar to the background values (outside both WWTPs areas).

Acknowledgements

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References


11. POLISH REGULATION
