

Article



# Concentration and Size Distribution of Culturable Bacteria in Ambient Air during Spring and Winter in Gliwice: A Typical Urban Area

# Ewa Brągoszewska, Anna Mainka \* and Jozef S. Pastuszka

Department of Air Protection, Silesian University of Technology, 22B Konarskiego St., 44-100 Gliwice, Poland; Ewa.Bragoszewska@polsl.pl (E.B.); Jozef.Pastuszka@polsl.p (J.S.P.)

\* Correspondence: Anna.Mainka@polsl.pl; Tel.: +48-322-371-060

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Abstract: The concentrations and size distributions of culturable bacterial aerosols were measured during spring and winter in outdoor air in Gliwice, Upper Silesia, Poland. This research on culturable bacteria was carried over a period of two years. The samples were collected using a six-stage Andersen cascade impactor (with aerodynamic cut-off diameters of 7.0, 4.7, 3.3, 2.1, 1.1, and 0.65  $\mu$ m). The results showed that the average concentration of culturable bacterial aerosol was 355 CFU m<sup>-3</sup> in spring, which was four times higher than during winter (65 CFU m<sup>-3</sup>). Bacterial aerosol concentrations showed the unimodal size distribution with the highest range of 3.3–4.7  $\mu$ m particles. The seasonal distributions of bacterial aerosol grain clearly indicate that, in winter, the size distribution of particles <7  $\mu$ m is more "flattened" and is characterized by an increased share of fine fractions and a decreased share of coarse ones. Environmental parameters, such as temperature, UV radiation, relative humidity, wind velocity, as well as PM<sub>10</sub> and PM<sub>2.5</sub> concentrations, were measured in order to analyse whether environmental factors had any effect on bacterial aerosols. Statistically, the most important meteorological factors in the viability of airborne bacteria were temperature and UV radiation.

Keywords: outdoor air; bacterial aerosols; size distribution; meteorological parameters; correlation

## 1. Introduction

Airborne microbes are an abundant component of atmospheric aerosols, with thousands, or even millions, of cells in each cubic metre of air [1,2]. The comprehensive monitoring of atmospheric bioaerosol concentrations is very important, not only for environmental management, but also to assess the health impact of air pollution. This is because bioaerosols can cause a variety of infectious diseases, as well as allergic and toxic reactions [3–6]. Bacterial aerosols are dispersed into the air from various sources, such as soil, plants, and water, as well as through the activities of animals, humans, and industrial operations, all of which can occur in the urban environment [7]. In ambient air, bacterial aerosols are often less abundant than fungal spores or pollen grains and rarely cause serious human illness [8]. Particularly well documented are negative effects that bioaerosols can play on the human respiratory system spanning from infectious diseases to allergies, and asthma [9,10]. However, they can contain cell membrane antigens such as endotoxin and peptidoglycans, which may damage lung tissue and induce serious respiratory symptoms [7,11]. It is important to note, that infectious and non-infectious diseases depend not only on the biological properties of bioaerosols, but also on the site of their deposition in the respiratory tract. The deposition of particles in the respiratory tract is directly related to the aerodynamic diameter of the particles and their effect on human health depends highly on their physical properties, and particularly on their size distribution [12].

Bacteria and microscopic fungi cells are 0.3 to 100  $\mu$ m in diameter (aerodynamic diameter). The size distribution of bioaerosols depends on the type of microorganism species, the age of the spore and nutrient medium, humidity, differences in aggregation rates of the spores, and the type of particles with which the spores are associated, such as mist or dust. It should be noted that bioaerosol particles can occur in the air as single cells or aggregates of cells, as well as in fragments. They are often transported attached to other particles, such as skin flakes, soil, dust, saliva, or water droplets [3,13].

Numerous studies have demonstrated the adverse health effects of exposure to biological agents, but most of them have focused on occupational and indoor environments because humans spend the majority of their time in these environments [14–21]. There is still very little information about the level of bacterial aerosol concentrations in ambient air. Meanwhile, information about the concentration of bacteria in the ground layer of the atmosphere is crucial in predicting the effects of breathing outdoor air on human health.

To date, bioaerosols have been measured in many indoor environments [18,20–22], including residential and public buildings [12,15], schools and nurseries [14,16,17,23]. Some studies have indicated that the concentration and size distribution of bioaerosols vary greatly by region and by season, depending on biotic and abiotic factors, such as the species of microorganism, environmental conditions, or human activities [24–30]. The role of geography and climate on concentrations of outdoor bacteria is important because the transport of bioaerosols is primarily governed by hydrodynamic and kinetic factors, while their fate is dependent on their specific chemical composition and the meteorological factors to which they are exposed [24,31–33]. The meteorological factors with major roles to play in bioaerosol concentrations and their transport are air temperature (T), relative humidity (RH), wind velocity (WV) and UV radiation (UV) [24,27,28]. Moreover, particulate matter (PM) in the atmosphere may influence bioaerosol concentrations. Gao et al. [34] showed that  $PM_{10}$  and  $PM_{2.5}$  have a statistically significant relationship with some bioaerosol characteristics. PM<sub>2.5</sub> appeared to have a unanimously negative correlation with culturable bioaerosol concentrations. A possible reason for this is that PM<sub>2.5</sub> usually contains soot, metals, and secondary inorganic components, which have a negative effect on the cultivation and survival of bioaerosols. However, the effect of  $PM_{10}$  on bioaerosols was different. Probably because the concentration of bioaerosols attached to coarse solid particles increases as PM accumulates. Meanwhile, limited chemicals in coarse particles, including mineral species from soil and particles of biological origin, had no obvious poisonous effect [34].

Although there have been studies presenting the relationship between different weather conditions and bioaerosol concentrations, the size distribution of bacterial aerosols and its relationship with different meteorological factors has been only preliminary recognized. Moreover, our previous studies conducted in Upper Silesia, Poland, showed that the highest concentrations of airborne bacteria in outdoor air occur in spring, while the lowest occur in winter [17,23,35]. Therefore, it is essential to explain the influence of meteorological parameters on the concentration and size distribution of bacterial bioaerosols in moderate climate zones.

The present study was carried out in a typical Eastern European urban city, with the aim of filling these knowledge gaps. Therefore, in this study, we investigated the concentration levels of culturable bacterial aerosols, their size distribution, as well as seasonal (spring-winter) variations in the outdoor air in Gliwice, a town in Southern Poland. In this work, we focused on culturable bacteria only, because these microorganisms are very sensitive to interaction with other air pollutants, e.g., PM<sub>2.5</sub> and PM<sub>10</sub>, and seem to be highly influenced by a variety of meteorological factors. Thus, to better understand these atmospheric dynamics under different conditions, a particular investigation of the complicated interrelationships between environmental factors and bioaerosols is needed. Moreover, this study can increase awareness of the influence of bioaerosols on human health, as well as provide references for a better understanding of OAQ (outdoor air quality) in the urban areas of developing countries. The data presented herein also indicate the need to control environmental pollution (through the future determination of an official European OAQ standard), thereby preventing human respiratory disease as a result of outdoor atmospheric exposure.

#### 1.1. Sampling Sites

The study was carried out in Gliwice (50°17′37.1″ N 18°40′54.9″ E), a typical city in the industrial region of Upper Silesia, Poland (a region containing 4.5 million people). The measurement point was located in the city centre, about 500 m from a busy road (Figure 1). The nearest surroundings to the sampling point were characterized by compact building development, i.e., buildings, roads, pavements, etc., covering most of the surface of this part of the city. The field sampling of ambient bioaerosols was carried out at human head height (approx. 1.5 m) above ground level.

Air sampling was conducted during two seasons (spring and winter) in 2015 and 2016. The spring season was conventionally assumed to last from mid-March to mid-June, while the winter months were assumed to last from mid-December to mid-March. A total of 150 measurements were taken for this study. Every measurement was conducted between 10:00 a.m. and 11:00 a.m. during the campaign period. The most important meteorological parameters, such as air temperature (T), relative humidity (RH) and wind velocity (WV), were recorded at the sampling sites using an Oregon Scientific WMR200 weather station (Oregon Scientific, Portland, OR, USA). On the other hand, PM<sub>10</sub>, PM<sub>2.5</sub>, and UV radiation (UV) measurements were obtained from automatic measurement stations made accessible by the Provincial Inspectorate for Environmental Protection (WIOŚ). They were collected during the sampling periods at the urban background station in Zabrze and the regional background station in Gliwice, located beyond the range of influence of Gliwice city.

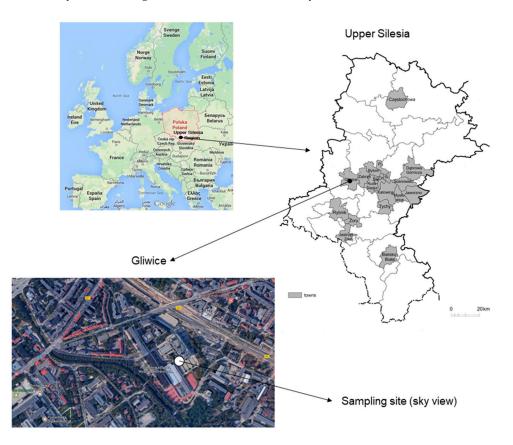


Figure 1. Location of bioaerosol measurement campaign (map data: 2017© Google, ORION-ME).

#### 1.2. Sampling and Analysis Methods

The air samples were collected using a six-stage Andersen cascade impactor (Tisch Environmental Inc., Cleves, OH, USA). The aerodynamic diameter ranges for each stage are as follows: stage 1 (>7  $\mu$ m), stage 2 (4.7–7  $\mu$ m), stage 3 (3.3–4.7  $\mu$ m), stage 4 (2.1–3.3  $\mu$ m), stage 5 (1.1–2.1  $\mu$ m), and stage 6 (0.65–1.1  $\mu$ m). The impactor also comprises a pump, which provides a constant flow

rate (28.3 L min<sup>-1</sup>) during measurement. The sampling time, following Nevalainen et al. [3,36], was 10 min. The microorganisms were collected on nutrient media in Petri dishes located on all impactor stages. The impactors were disinfected with 70% ethanol-immersed cotton balls between each sampling. Tryptic soy agar (TSA) was used for the bacteria, with cycloheximide added to inhibit fungal growth. The bacteria were counted according to the Polish Standard [23]. It is important to note that, although it is extremely difficult to directly measure the concentrations of living airborne bacteria, the commonly-used substitute for the concentrations of living microorganisms present in the air is the number of colony forming units in the volume of air (CFU m<sup>-3</sup>). It should be noted that the culture-based method employed in this study includes probably only about 10% of the total microorganisms [29]; however, it still provides valuable information for the assessment of human exposure and the establishment of a Polish official standard governing the levels of bioaerosols in ambient air.

The quality control was performed by using PN-EN12322 [37] and ISO 11133 [38] standards, with the same operation details as in our previous studies [17,23].

#### 1.3. Statistical Analysis

All statistical analyses were performed using the Statistica 12 statistical package (StatSoft). The concentration values reported below were presented as the mean values and standard deviation. Given the fact that the data were not normally distributed, a nonparametric method was employed. The Mann-Whitney test was applied to assess seasonal differences at the sampling site. Spearman's rank correlation coefficient was used to evaluate the statistical dependence between airborne bacterial concentrations and meteorological parameters, including temperature (T), relative humidity (RH), wind velocity (WV), UV radiation (UV), and  $PM_{10}$  and  $PM_{2.5}$  concentrations, separately for spring and winter. Significant differences were determined when a probability lower than 0.05 (p < 0.05) was considered.

#### 2. Results and Discussion

#### 2.1. Bacterial Aerosol Concentrations during the Spring and Winter Seasons

Figure 2A,B shows the concentrations of aerosol particles in the air in spring and winter. The concentrations of bacterial aerosols in the outdoor air ranged from 80 to 1124 CFU m<sup>-3</sup> in spring and from four to 265 CFU m<sup>-3</sup> in winter. The average concentration of bacterial aerosol was found in spring (355 CFU m<sup>-3</sup>), and was more than five times higher than the average level recorded in winter (65 CFU m<sup>-3</sup>).

The values recorded in the outdoor air in Gliwice in winter are higher than to those obtained in studies conducted in Warsaw, Poland, where the average concentration of airborne bacteria in winter was 39 CFU m<sup>-3</sup> (range 35–126 CFU m<sup>-3</sup>) [22], and in Seoul, South Korea, where the average concentration of airborne bacteria in winter was 11 CFU m<sup>-3</sup> (range 0–118 CFU m<sup>-3</sup>) [6]. In Ankara, Turkey, Mentese et al. [18], reported an average concentration of bacterial aerosol of 57 CFU m<sup>-3</sup> from December to March 2007. The lowest bacterial concentrations are observed in winter because extreme conditions, such as a decreased temperature and the heaviest rainfall and snowfall of the year, might contribute to decreased bacterial levels [28,39]. The values obtained in Gliwice are also comparable to data obtained in Colorado, USA and Montreal, Canada, where the lowest average bacterial aerosol concentrations were found in the winter and the highest were found in spring [1,40]. On the other hand, the definition of the season cannot be used as the only criteria for seasonal patterns. For example, Genitsaris et al. [41] underlined the higher concentration of bacterial aerosol in winter comparing to spring, while our results present the opposite trend. However, winter in Greece is characterized by the similar positive temperature levels, which promotes the growth of bacteria, as in Poland during spring.

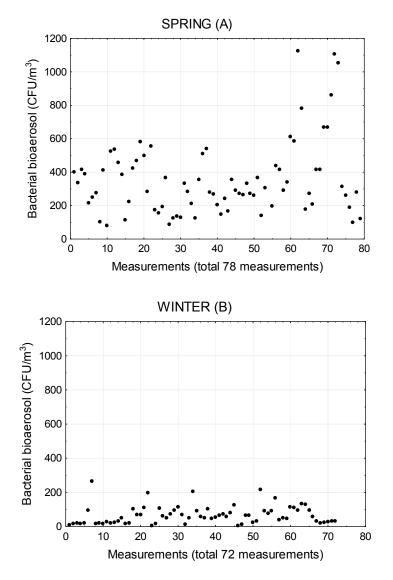


Figure 2. Concentration of culturable bacterial aerosols in outdoor air in spring (A) and winter (B).

Although in Poland, and many other countries, the legislation governing microbiological air pollution standards has not been developed and implemented according to the available literature, it can be seen that the concentration levels of bioaerosols obtained in our study are below the proposed standards. Górny [42], in a review of propositions for outdoor air, proposed a total bacterial concentration of 5000 CFU m<sup>-3</sup> as the upper limit for regulatory bacterial aerosol levels. However, research by the Robert Sauvé Occupational Health and Safety Research Institute (IRSST) indicates that if the total concentration of airborne bacteria concentrations is 1000 CFU m<sup>-3</sup>, then microbial contamination is possible; this justifies further investigation of the situation and the requirement to take action [43].

#### 2.2. The Size Distribution of Bacterial Aerosols

The aerodynamic diameters of bioaerosols determine their potential to be deposited either in the tracheal, bronchial, or alveolar regions of the lungs; thus, the effects of bioaerosols on human health are not only related to their concentrations, but also to their size distribution. The size distribution of airborne bacteria in spring and winter is shown in Figures 3 and 4. It can be seen that the airborne culturable bacteria collected in spring were distributed unimodally, however, in winter,

the size distribution of bacterial particles was more "flattened" and was characterized by the increased contribution of fine bioparticles and a decreased contribution of coarse ones.

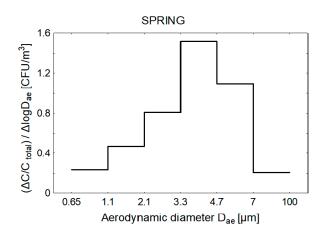


Figure 3. Size distribution of bacterial aerosols in outdoor air in spring.

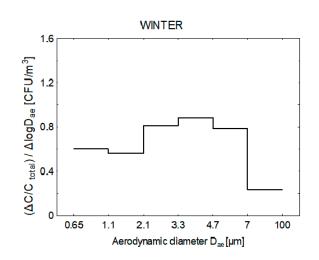


Figure 4. Size distribution of t bacterial aerosols in outdoor air in winter.

During both seasons, bacteria were primarily found as the coarse particles larger than 2.1  $\mu$ m, with contributions of 81.8% and 78.2% during spring and winter, respectively. This seasonal difference was not statistically significant (p > 0.05).

It is also interesting that 53.9% and 59.2% of airborne bacteria were found within the size range of less than 4.7  $\mu$ m in spring and winter, respectively. This indicates that more than 50% of bacterial aerosols were in the respirable size range, regardless of the season. Respirable particles have the potential to be deposited either in the upper or lower respiratory tract. Therefore, exposure to ambient bacteria has potential adverse effects on human health during both spring and winter. Furthermore, the present results suggest that the respirable fraction of the bacteria in ambient air is comparable to those obtained in indoor environments [12,15,17,29].

The size distribution of bioaerosols depends upon the species of microorganism, the age of the spore and the nutrient medium, differences in spore aggregation rates, as well as the type of particles they are associated with, such as mist or particulate matter [23]. Differences in size distribution may be also affected by meteorological factors, which explains the strong interaction between the two opposing mechanisms stimulating the growth and reduction of bacterial aerosols.

In order to check the assumed influence of meteorological parameters on bacterial size distribution, we checked the size distributions of bacterial aerosols at the minimum and maximum values of the

selected meteorological parameters: ambient temperature, UV radiation, relative humidity, and wind velocity. A particularly different size distribution was observed between spring and winter for the highest and the lowest UV radiation values (Figures 5 and 6). Generally, spring conditions are better for the growth of bacterial particles, mainly for the formation of a larger cells. On the other hand, a substantial percentage of airborne bacteria died in stronger ultraviolet light. It seems that the particles most sensitive to UV radiation are single bacterial cells and, therefore, fine bacterial aerosol particles are present in lower numbers in spring and higher numbers in winter. In winter, UV radiation gradually loses importance as a bactericidal factor; on the other hand, the growth of microorganisms preferring to form in agglomerates is much slower. As a result, an increasing proportion of fine particles is observed, while the proportion of coarse grains decreases. It should be noted that the size distribution of bacterial grains mainly affects the extreme fractions.

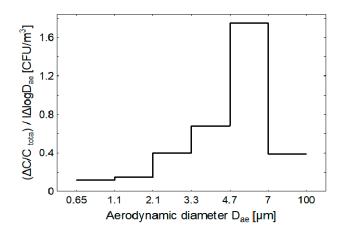


Figure 5. Size distribution of bacterial aerosols recorded at the maximum UV radiation value (886 W/m<sup>2</sup>).

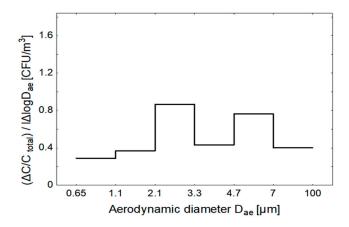


Figure 6. Size distribution of bacterial aerosols recorded at the minimum UV radiation value (39 W/m<sup>2</sup>).

Meanwhile, bacterial particles with aerodynamic diameters measuring between 1.1 and 3.3  $\mu$ m, do not significantly affect particle size distribution, while they can significantly alter the ratio of concentrations of respirable fractions to coarse fractions. Their levels can fluctuate strongly depending on the instantaneous values of the meteorological factors and the local bioaerosol structure. Therefore, proposals for particle size distribution are not directly transferable to the analysis of concentrations of respirable particles to coarse particles.

#### 2.3. The Influence of Meteorological Factors

The factors influencing the concentration and size distribution of culturable bacteria are very complex and include meteorological parameters, climatic conditions, source strength, and the geographical environment. Due to the great variation in weather conditions and meteorological factors, the most important factors influencing bioaerosols vary according to different seasons and locations [28]. In this study, environmental parameters such as temperature, UV radiation, relative humidity, wind velocity, as well as  $PM_{10}$  and  $PM_{2.5}$  concentrations, were measured in order to investigate whether environmental factors had any effect on bacterial aerosols. Table 1 provides a general overview of these environmental parameters during spring and winter. They were measured at the same time as the bioaerosols.

Outdoor Parameters, Mean $\pm$ SD							
Parameters	Spring	Winter					
Temperature, °C	$18.0\pm4.7$	$-3.6\pm4.4$					
Relative humidity (RH), %	$72.2\pm15.7$	$77.4 \pm 11.2$					
Wind velocity, km/h	$9.30\pm5.2$	$14.4\pm6.3$					
UV radiation, $W/m^2$	$461.1\pm246.5$	$145.4\pm118.5$					
PM <sub>10</sub> , μg/m <sup>3</sup>	$27.4\pm21.5$	$90.5\pm50.4$					
$PM_{2.5}, \mu g/m^3$	$12.9\pm8.4$	$78.8 \pm 47.8$					

Table 1. Environmental parameters during sample collection.

#### 2.3.1. The Influence of Temperature

Our previous research clearly indicates that, in Poland, spring is more suitable for the survival and growth of culturable bacteria than summer [35]. Moreover, plant growth is particularly observed in spring; this may also contribute to the increase of bioaerosol sources, as well as to an increased variety of media in which microorganisms can produce and grow [28,44]. During the study, the temperature ranged from 8–27 °C and -11-6.5 °C in spring and winter, respectively. The temperature ranges might suggest that bioaerosol concentrations would be significantly and positively correlated with atmospheric temperature, i.e., the concentration of bioaerosols will increase as the temperature increases.

As shown in Table 2, there is a significant positive correlation between temperature and bacterial bioaerosol concentrations in winter, but there is a negative correlation in spring. The Spearman correlation coefficient was 0.675 and -0.316 (p < 0.05) in winter and spring, respectively.

During winter, the low temperatures decrease cell membrane fluidity and slow down microbial activity; the increased atmospheric temperature promotes the growth and release of bacteria—thus, a positive correlation is observed. On the other hand, in spring, the temperature is too high and is related to strong ultraviolet radiation. This is confirmed by a significant positive Spearman correlation coefficient of 0.687 (p < 0.05) meaning that these conditions are not suitable for the propagation and growth of plants and lead to the denaturation and inactivation of proteins [28].

	Bacteria	Т, °С	RH, %	W, km/h	UV, W/m <sup>2</sup>	PM <sub>10</sub>	PM <sub>2.5</sub>		
Spring									
Bacteria	1								
T, °C	-0.316	1							
RH, %	0.156	-0.430	1						
W, km/h	-0.040	0.049	0.063	1					
UV, $W/m^2$	-0.329	0.687	-0.497	0.148	1				
$PM_{10}$	-0.067	0.121	-0.308	-0.361	0.337	1			
PM <sub>2.5</sub>	0.020	0.080	0.037	-0.509	0.090	0.616	1		
Winter									
Bacteria	1								
T, °C	0.675	1							
RH, %	0.351	0.397	1						
W, km/h	-0.102	-0.201	-0.043	1					
UV, $W/m^2$	-0.603	-0.640	-0.614	0.070	1				
$PM_{10}$	-0.301	-0.209	-0.218	-0.053	0.176	1			
PM <sub>2.5</sub>	-0.261	-0.138	-0.161	-0.119	0.123	0.984	1		

**Table 2.** Spearman correlation coefficients between bacterial aerosol concentrations and environmental parameters (n = 150).

Italic entries indicate that the correlation is significant at the 0.05 level; Temperature, T (°C); Relative humidity, RH (%); Wind velocity, W (km/h); UV radiation, UV (W/m<sup>2</sup>); concentration of particulate matter PM<sub>10</sub> and PM<sub>2.5</sub> ( $\mu$ g/m<sup>3</sup>).

## 2.3.2. The Influence of UV Radiation

Solar radiation is known to constitute an effective sterilization method for bioaerosols [29]. During our study, UV radiation was significantly different for each season. The average UV radiation was 461.1 W/m<sup>2</sup> and 145.4 W/m<sup>2</sup> in spring and winter, respectively. Table 2 shows the correlation between bacterial aerosol concentrations and UV radiation. During both seasons, we observed a statistically significant negative relationship between bacterial aerosol concentrations and UV radiation. The Spearman correlation coefficient was -0.329 and -0.603 (p < 0.05) in spring and winter, respectively. Therefore, airborne bacteria in Gliwice would be more likely to propagate and grow at relatively low UV radiation levels, whereas a comparatively high UV radiation value would inhibit their growth.

## 2.3.3. The Influence of Relative Humidity (RH)

The viability of bioaerosols becomes inhibited if RH is too low, because a dry environment will decrease the metabolism and physiological activities of microorganisms. The RH in Gliwice ranged from 44–97% and 56–94% in spring and winter, respectively. The average RH was relatively stable and changed little between seasons; the average was 72.2% and 77.4% in spring and winter, respectively (Table 1). Positive correlations were found between concentrations of viable bacteria and RH. This was consistent with the results reported by Li et al. [45]. As indicated in Table 2, the correlation was higher and only statistically significant in winter (p < 0.05). According to Jones and Harrison [46], moisture in the air could alter the integrity of cell walls and viral coats and, thus, increased air humidity could promote the growth and release of bacteria.

## 2.3.4. The Influence of Wind Velocity

Generally, wind has been confirmed to have an effect on bioaerosols, varying according to different speeds and directions. For example Montero et al. [47] reported that during summer season wind speed and total coarse bioaerosols were significantly correlated. Wind velocity is clearly associated with the release and dispersal of spores, although its effect varies according to its intensity and the height at which it is recorded. Wind speeds that exceed a threshold value will accelerate the process of

releasing microbes from soil or plant surfaces by blowing them off or moving them. However, at high wind speeds, microbial concentrations will become diluted [46]. In our study, the Spearman correlation coefficient was -0.040 and -0.102 (p > 0.05) in spring and winter, respectively. Therefore, there was no relationship between wind velocity and bacterial aerosol concentrations in either season. This result is in a good agreement with the observations of Li et al. [26,48]. To find, however, a more precise relationship between the level of airborne bacteria and wind speed very large datasets should be analyzed. Especially, it is important to measure the concentration of bacterial aerosols for the velocity of wind ranging from less than 1 km h<sup>-1</sup> to more than 20 km h<sup>-1</sup>.

## 2.3.5. The Influence of PM

Bioaerosols, including moulds and yeasts, bacteria, and viruses, as well as particles of plant origin pollens and algae, are bound to PM and, therefore, its biological and morphological characteristics may undergo considerable changes. In Poland, this phenomenon might be particularly important in winter due to high PM emissions from coal-based domestic sources [49]. Changes in microbial dispersal patterns and aerodynamic diameters could lead to variations in deposition locations in human respiratory tracts. At high PM concentrations, the potential synergistic effects between biological and chemical pollutants may further intensify their hazardous effects on human health [34]. Polish legislation [50] specifies a 24 h mean concentration of  $PM_{10}$  in ambient air, which is 50 µg/m<sup>3</sup>.  $PM_{2.5}$  has no corresponding short term (24 h) limit, but there is an annual limit of 25  $\mu$ g/m<sup>3</sup>. The World Health Organization (WHO) is more strict in this regard: for PM<sub>2.5</sub>, it recommends a 24 h average standard of 25  $\mu$ g/m<sup>3</sup> [51] and recommends applying the same guidelines to ambient air as for indoor spaces [52]. During the spring part of the study, the average concentrations of  $PM_{10}$  and  $PM_{2.5}$ (27.4 and 12.9  $\mu$ g/m<sup>3</sup>) did not exceed Polish and WHO guidelines. However, in winter, the PM<sub>10</sub> standard was exceeded by roughly 60% and the  $PM_{2.5}$  standard was exceeded by more than 300%. In spring, no consistent relationship between PM and bioaerosols was observed; however, in winter, a decrease in culturable airborne bacteria was observed as PM<sub>10</sub> and PM<sub>2.5</sub> concentrations increased. The Spearman correlation coefficient was 0.301 and -0.261 (p > 0.05) for PM<sub>10</sub> and PM<sub>2.5</sub>, respectively. Therefore, a weak negative relationship was found between PM and bacterial aerosol in winter. A possible reason for this is that winter PM contains components that have a negative effect on the cultivation and survival of bioaerosols [34].

## 3. Conclusions

The term "bioaerosol" is not very clearly defined and is frequently used with different meanings, we focused only on bacterial aerosol. This study provided information on the concentrations and size distribution of ambient culturable bacteria in Gliwice City during spring and winter. The average concentration of culturable bacterial aerosols in outdoor air differed significantly over the study period. The average values were  $355 \pm 222$  CFU m<sup>-3</sup> and  $65 \pm 53$  CFU m<sup>-3</sup> in spring and winter, respectively. The highest concentrations of bacterial aerosol were recorded during spring (1124 CFU m<sup>-3</sup>), while the lowest were in winter (4 CFU m<sup>-3</sup>). Comparing the concentration levels of bacterial aerosols with those recorded in other studies, it can be concluded that the atmospheric air in Gliwice is relatively clean, microbiologically. The examination of seasonal size distributions of bacterial grains clearly indicated that during the cooler season, the size distribution is more "flattened" and is also characterized by an increased contribution of fine bioparticles and a decreased contribution of coarse bacteria. Another important finding was that more than 50% of bacterial aerosols fell within the respiratory range.

We analysed the correlation between bacterial aerosol concentrations and meteorological parameters. Statistically, the most important meteorological factors in the viability of airborne bacteria were temperature (T) and UV radiation (UV). In spring, the Spearman correlation coefficients for T or UV with bacterial aerosol concentrations were -0.316 and -0.329 (p < 0.05) and 0.675 and -0.603 (p < 0.05) in spring and winter, respectively. The influence of UV radiation was negative during both

seasons. Low UV radiation led to higher airborne bacteria concentrations, while high RH contributed to lower bacterial aerosol concentrations. However, the temperature had a different impact in spring and in winter. During winter, a strong positive correlation was observed between temperature and bacterial aerosol concentrations. These concentrations were high in relatively warm temperatures and were low in cooler temperatures. During spring, the opposite was observed. The higher temperature decreased the ability of ambient bacteria to survive. This dependence was caused by the significant correlation between temperature and UV radiation. The Spearman correlation coefficients between T and UV

were of 0.687 and -0.640 (p < 0.05) in spring and winter, respectively. Relative humidity was positively correlated with bacterial aerosol concentrations; however, this relationship was only statistically significant in winter (0.351; p < 0.05). Wind velocity was found to have no statistically significant correlation with bacterial aerosol concentrations. The statistically significant weak relationship between bacterial aerosols and particulate matter was only observed in winter. The Spearman correlation coefficients were -0.301 and -0.261 for PM<sub>10</sub> and PM<sub>2.5</sub>, respectively; therefore, high PM<sub>10</sub> and PM<sub>2.5</sub> levels during winter could reduce bacterial viability.

In addition to the limitations of the culture-based method employed in the present study (a culture-based method employed in the present study probably accounts for only ~10% of the total microorganisms), the results of this campaign may still be useful in developing control devices for bacterial aerosols and in predicting of the occurrence of diseases caused by bioaerosols. However, to precisely anticipate the adverse health effects caused by the exposure to airborne bacteria the taxonomic identification of collected bacterial particles should be made. If not, the typical percentage of genera/species in the total bacteria concentration around the sampling point could be assumed using the literature data. It should be noted that such approach may be applied in the preliminary assessment of the influence of bacterial aerosol on the human health only.

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**Author Contributions:** Ewa Bragoszewska performed the measurements and drafted the manuscript. Anna Mainka prepared the final manuscript. Jozef S. Pastuszka helped with the data analysis.

**Conflicts of Interest:** The authors declare no conflict of interest.

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