Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering. Vol. XV, 2026
Print ISSN 2285-6064, CD-ROM ISSN 2285-6072, Online ISSN 2393-5138, ISSN-L 2285-6064

DYNAMICS OF AIRBORNE MICROORGANISMS ACCORDING TO ENVIRONMENTAL CONDITIONS

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Abstract

The present study aims to track the dynamics of airborne microorganisms in the territory of Sofia, Bulgaria in relation to the dynamics of fine particulate matter (FPM). Sampling points were 20, which cover territories with different dynamics - parks, intersections, central and peripheral parts, etc. Spin Air sampler was used. Sampling was carried out at a height of 1.5 m, for optimal consideration of the potential impact on human health. Specific microbiological characteristics were recorded - total microbial number in 1m^3 air, the main microbiological groups and their ratio and the percentage of pigment forms in relation to the change in the UV index. Specific environmental parameters related to airborne microorganisms were also recorded - amount of FPM, air temperature, humidity, atmospheric pressure, wind direction and speed, UV index. The data show a specific distribution of microbial groups, as well as a certain microbial load in points with intensive car traffic. There is a direct correlation between the amount of pigment forms and UV index ($r=0.92$; $p<0.05$). In the present pilot study, no direct correlation was found between the increase in fine particulate matter and microbial abundance (Pearson correlation matrix, significance thresholds 95%, $p<0.05$). However, there is a clear relationship between the reduced dust level and the higher percentage of woody vegetation.

Key words*: airborne microorganisms, air quality, fine particulate matter, pigment forms, UV index.*

INTRODUCTION

Understanding the complex interaction between physical pollutants and biological components of air is crucial for modern urban ecology and public health. The air we breathe in urban areas is not just a mixture of gases, but a complex multiphase system containing solid particles and biological agents known as bioaerosols. Bioaerosols contain particles of biological origin, including all kinds of biological components and fragments, microorganisms, pollen, parts of plant fragments, etc. (Despres et al., 2012). For this reason, it is very important to distinguish between bioaerosols (which include any type of biological fragments) and airborne microorganisms (which include microorganisms from different groups such as bacteria, micromycetes, actinomycetes, etc.), as Kim et al. (2018), point out. In recent years, Sofia has been among the European capitals with the most serious challenges in terms of ambient air quality, mainly due to its geographical location in a valley and intense car traffic, which is why the study of ground-level air quality is of utmost importance. Modern research shows that airborne microorganisms are not just passive passengers. They use fine particulate matter (FPM) as a "vehicle" and substrate for growth and survival (Tignat-Perrier et al., 2020).

As Meola et al. (2015) and Haas et al. (2025) point out in their studies, fine particulate matter (FPM) act as transport nuclei for the transfer and spread of airborne microorganisms. It is important to note that airborne microorganisms are an important component of ambient air. Although they sometimes have a harmful effect on people and worsen the quality of the environment in urban areas, they are extremely important in the upper layers of the atmosphere, where they play the role of condensation nuclei, as a result of which it is believed that their increase as suspended units may have an impact on the ongoing climate changes (Zhai et al., 2018). It is precisely this complexity and intricacy that necessitates the in-depth study of airborne microorganisms.

Recent data suggest that the chemical composition of PM may selectively favor certain microbial taxa, making urban air a potential reservoir for pathogens or allergens (Cao et al., 2014; Wang et al., 2015). The link between fine

dust particles and the number of microorganisms in the air is undeniable. Last but not least, fine dust particles pose a risk to human health, as their increase not only leads to physical changes in air quality, but can also increase the risk of allergies and respiratory problems, given that large quantities of airborne microorganisms are adhered to PM (Arca-Lafuente et al., 2025). What is specific about fine dust particles is that, due to their size and extremely low relative weight, they can remain suspended in the air for a very long time, with their spread strongly depending on the speed and direction of the wind (He et al., 2021). The increase in PM concentration is not only due to natural sources such as volcanic eruptions, fires, earthquakes or landslides of large land masses (Zanoletti et al., 2024; Whitty et al., 2026), but also exclusively to human activity, such as construction, industrial combustion, car traffic, etc. (Edo et al., 2024; Morales et al., 2025). If wind is the main factor influencing the concentration and distribution of FPM, then changes in temperature, solar radiation index and humidity are of key importance for the vitality, quantity and distribution of airborne microorganisms (Tang, 2009).

The main goal of this study is to track the dynamics of airborne microorganisms in the territory of Sofia, Bulgaria in relation to the dynamics of fine particulate matter (FPM) and specific environmental conditions.

MATERIALS AND METHODS

Airborne microorganisms are a major component of both atmospheric aerosols and the composition of air, which is highly dynamic. The quantitative and qualitative distribution of microbial groups is of great importance for both urban air quality and public health. However, airborne microorganisms remain poorly understood. In order to identify differences in environmental influences (type of urban environment, intensity of car traffic, vegetation cover) twenty samples were collected in three replicates from different locations with different levels of anthropogenic pressure in the capital of Bulgaria, Sofia, in the autumn of 2025. Points with increased anthropogenic pressure are marked as hot points (HP), and points with low anthropogenic pressure, located in park areas, as green zones (GZ), Figure 1:

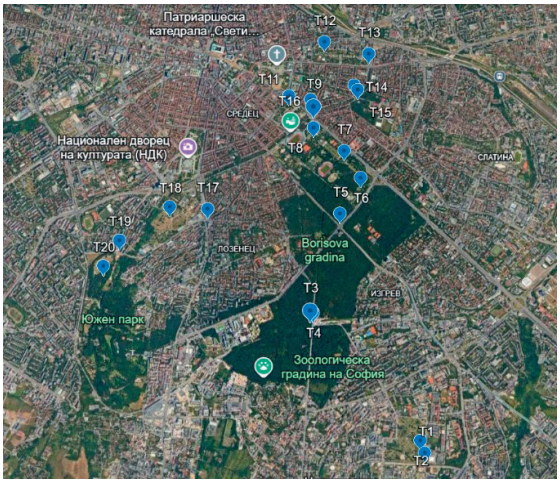


Figure 1. Location of selected sampling points (T1 Crossroad of Osmi Dekemvri Street and Rosario Street (HP); T2 Student Park (GZ); T3 Hunting Park (GZ); T4 Crossroad of Simeonovsko Shosse and Nikola Gabrovski Blvd. – "Loven Park Intersection" (HP); T5 Television Tower - Roundabout (HP); T6 Observatory, in Upper part of Borisova Gpadina Park (GZ); T7 Lily Pond - Middle part of Borisova Gradina Park (GZ); T8 Lower part of Borisova Gradina Park (GZ); T9 Orlov Bridge (HP); T10 V. Levski Blvd. intersection with Tsar Osvoboditel Blvd. (HP); T11 Prince's Garden (GZ); T12 Zaimov Park (GZ); T13 Madrid blvd. Crossroad (HP); T14 Crossroad Evlogii Geogriev Blvd And Shipka str. (HP); T15 Military Academy Park (GZ); T16 Iranian Embassy on Evlogii Georgiev Blvd (HP); T17 Hotel Hemus (HP); T18 Park behind the Museum of Land and People (GZ); T19 Crossroad Vitoshka str. и South Park (HP); T20 South Park (GZ))

For the purposes of the study, the following nutrient media were used: Mesopeptone agar for the cultivation of bacteria, Czapek-Dox agar for the cultivation of micromycetes, and Starch Ammonia Agar for the cultivation of actinomycetes. Air sampling was carried out using an IUL Spin Air impactor, which has high accuracy and resolution. The device works by means of a rotary mechanism, which allows for complete utilization of the nutrient medium layer in the Petri dish, thus ensuring an even distribution of the columns in the Petri dish, which facilitates reading. The measurement was carried out at a height of 1.5 m above the ground surface, corresponding to the human respiratory zone. The sampling parameters are as follows: 120 seconds for each studied group of microorganisms at a flow rate of 100 L/min. These parameters were selected after a series of preliminary test analyses to establish the optimal air flow rate. After sampling, the samples were transferred in specialized bags to the laboratory, where subsequent incubation was performed. Bacteria were cultured for 48 hours at 28°C, fungi - 7 days at 30°C, and actinomycetes - 10 days at 35°C. The counting of the developed colonies was performed using a digital colony counter, with the data converted into CFU*$10^2/m^3$.

Along with the taking of microbiological samples, the levels of fine particulate matter were measured, and meteorological parameters were also recorded. The concentration of PM was recorded using the Envea PM Cairsense device. The device simultaneously measures the PM10, PM2.5 and PM1 fractions, using laser diffraction. During sampling, meteorological data were also recorded: air temperature, humidity, UV index, wind speed and direction. The data for the microbiological analyses were statistically processed using the StatSoft Statistica 12 program, Pearson correlation matrix, at significance thresholds of 95%, $p<0.05$.

RESULTS AND DISCUSSIONS

The results for the reported meteorological parameters are presented in Figure 2.
Meteorological data indicate slightly lower temperatures than usual for the season. Humidity is also low, ranging between 48% and 65% throughout the sampling period, with the lowest values shortly after noon. This is the period when the temperature and UV index increase. The wind speed of 6.7 m/s from the west-northwest is a constant factor for all measurements.

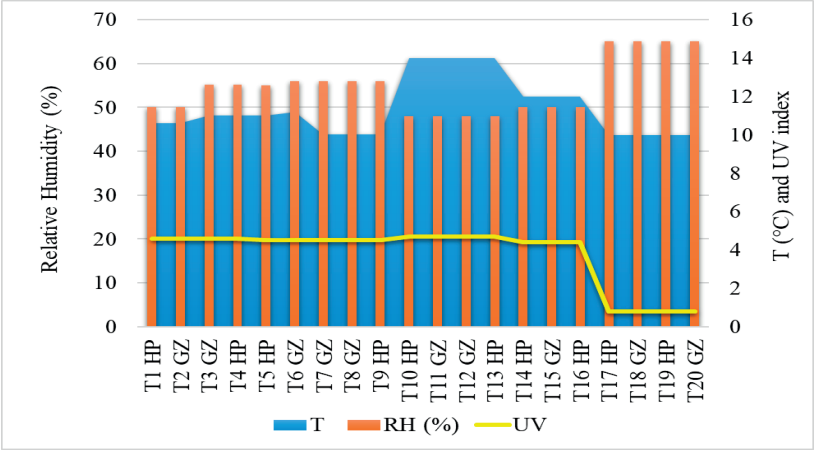


Figure 2. Recorded climatic characteristics during sampling

Such a speed is theoretically sufficient to facilitate the transport of fine dust, bioaerosols, and airborne microorganisms from the outskirts to the city center, suggesting a potential directional transport pathway that warrants further meteorological analysis, while also helping to disperse larger PM10 particles.

The temperature range of 10°C to 14°C is relatively favorable for many of the mesophilic microorganisms commonly found in the air environment (Haas et al., 2020).

The results for the reported amounts of fine particulate matter, by individual fractions, are presented in Table 1.

Table 1. Fine particulate matter (μg/m^3)

	PM 10	PM 2.5	PM 1
T1 HP	48.83 ± 12.85	31.83 ± 7.90	23.17 ± 5.34
T2 GZ	44.33 ± 5.35	32.08 ± 4.34	23.67 ± 3.49
T3 GZ	44.25 ± 5.35	35.50 ± 4.34	27.25 ± 3.49
T4 HP	67.25 ± 12.85	42.87 ± 7.90	29.87 ± 5.34
T5 HP	35.56 ± 12.85	28.56 ± 7.90	20.78 ± 5.34
T6 GZ	31.55 ± 5.35	26.20 ± 4.34	19.60 ± 3.49
T7 GZ	38.89 ± 5.35	30.88 ± 4.34	22.67 ± 3.49
T8 GZ	38.78 ± 5.35	30.70 ± 4.34	22.78 ± 3.49
T9 HP	47.00 ± 12.85	38.00 ± 7.90	27.75 ± 5.34
T10 HP	52.22 ± 12.85	39.44 ± 7.90	28.78 ± 5.34
T11 GZ	39.62 ± 5.35	31.62 ± 4.34	23.25 ± 3.49
T12 GZ	30.70 ± 5.35	23.80 ± 4.34	17.30 ± 3.49
T13 HP	33.50 ± 12.85	26.60 ± 7.90	19.70 ± 5.34
T14 HP	23.40 ± 12.85	18.50 ± 7.90	13.87 ± 5.34
T15 GZ	35.72 ± 5.35	24.81 ± 4.34	17.90 ± 3.49
T16 HP	31.50 ± 12.85	23.07 ± 7.90	16.93 ± 5.34
T17 HP	38.76 ± 12.85	29.38 ± 7.90	21.61 ± 5.34
T18 GZ	47.11 ± 5.35	37.00 ± 4.34	27.44 ± 3.49
T19 HP	31.86 ± 12.85	23.28 ± 7.90	18.00 ± 5.34
T20 GZ	39.90 ± 5.35	28.50 ± 4.34	20.30 ± 3.49

The data from the measurements conducted in Sofia reveal clear spatial heterogeneity in the levels of particulate matter pollution of the fractions PM10, PM2.5 and PM1. These differences reflect the influence of local emission sources, such as road transport, and the protective role of urban vegetation.

The highest levels of PM10 were recorded at point T4 (Loven Park Intersection) - 67.25 μg/m^3, which exceeds the average values with 68% (average value = 40.01 μg/m$^{3)}$ for the inner parts of the parks. This fact illustrates the so-called "edge effect", in which areas in the immediate vicinity of busy transport arteries accumulate both primary emissions from fuel combustion and re-suspended dust from the road surface, which is rich in microorganisms, including those of soil origin, as has been found in other studies (Rathnayake et al., 2016). For comparison, point T14 (Military Academy Crossroad) shows the lowest PM10 values (23.4 μg/m^3), which is probably due to specific air circulation in the area of this crossroad, which is also confirmed by our other studies (Grigorova-Pesheva & Peshev, 2025).

Analysis of the ratio between the different fractions is essential for microbiology. Fine particles PM2.5 and PM1 have a larger relative surface area, making them more effective carriers of small bacterial cells and fragments of fungal mycelium, which increase the risk of allergies (Thangavel et al., 2022). The average PM2.5/PM10 ratio at the surveyed points varies, but in heavily congested areas such as Orlov Most (T9) and Blvd. V. Levski (T10), PM2.5 concentrations reach 38.00 μg/m^3 and 39.44 μg/m^3, respectively.

The results of the microbiological analyses performed are presented in Table 2.

Figure 3 presents a comparison of the reported biogenicity of the individual points, for which purpose the green areas are depicted in green and the hot points in red. The graph also shows the reported values of fine particulate matter in μm/m^3.

Table 2. Microbiological parameters (CFU*10^2/m^3)

	TMN	Bacteria	Actinomycetes	Micromycetes
T1 HP	4.90 ± 2.11	3.00 ± 1.53	1.80 ± 2.08	0.10 ± 0.41
T2 GZ	6.05 ± 2.75	2.30 ± 0.48	3.55 ± 2.62	0.20 ± 0.33
T3 GZ	2.55 ± 2.75	1.30 ± 0.48	1.10 ± 2.62	0.15 ± 0.33
T4 HP	6.20 ± 2.11	4.85 ± 1.53	1.30 ± 2.08	0.05 ± 0.41
T5 HP	9.70 ± 2.11	7.35 ± 1.53	1.75 ± 2.08	0.60 ± 0.41
T6 GZ	7.70 ± 2.75	2.35 ± 0.48	5.10 ± 2.62	0.25 ± 0.33
T7 GZ	3.79 ± 2.75	2.40 ± 0.48	1.20 ± 2.62	0.19 0.33
T8 GZ	3.00 ± 2.75	1.50 ± 0.48	1.30 ± 2.62	0.20 ± 0.33
T9 HP	5.65 ± 2.11	3.45 ± 1.53	1.95 ± 2.08	0.25 ± 0.41
T10 HP	5.50 ± 2.11	3.10 ± 1.53	2.30 ± 2.08	0.10 ± 0.41
T11 GZ	3.95 ± 2.75	2.20 ± 0.48	1.55 ± 2.62	0.20 ± 0.33
T12 GZ	3.95 ± 2.75	2.10 ± 0.48	1.70 ± 2.62	0.15 ± 0.33
T13 HP	7.25 ± 2.11	5.90 ± 1.53	1.30 ± 2.08	0.05 ± 0.41
T14 HP	7.15 ± 2.11	4.45 ± 1.53	2.65 ± 2.08	0.05 ± 0.41
T15 GZ	4.45 ± 2.75	2.45 ± 0.48	1.85 ± 2.62	0.15 ± 0.33
T16 HP	5.85 ± 2.11	3.60 ± 1.53	2.10 ± 2.08	0.15 ± 0.41
T17 HP	7.22 ± 2.11	4.45 ± 0.48	1.95 ± 2.62	0.82 ± 0.33
T18 GZ	11.73 ± 2.75	1.30 ± 0.48	9.45 ± 2.62	0.98 ± 0.33
T19 HP	11.75 ± 2.11	2.15 ± 1.53	8.35 ± 2.08	1.25 ± 0.41
T20 GZ	6.25 ± 2.75	1.45 ± 0.48	3.85 ± 2.62	0.95 ± 0.33

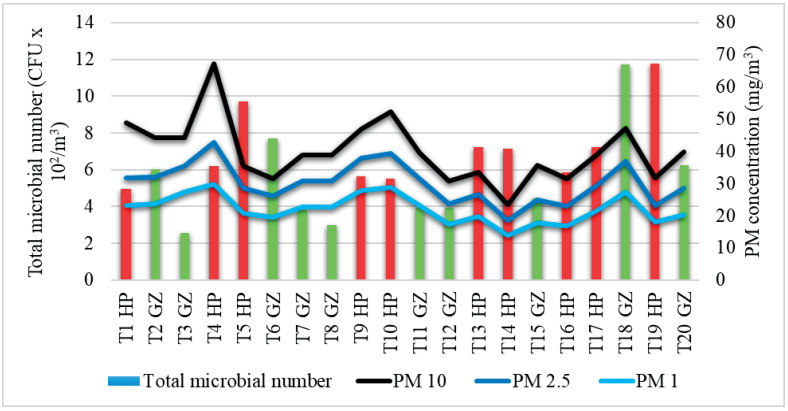


Figure 3. Total microbial number of the studied points and FPM load

Analysis of the results shows that green areas generally have a lower microbial load than hot points, although there are exceptions. Green areas with a low percentage of woody vegetation (T2 and T18) stand out with a higher microbial load, close to that of hot points. These are park areas dominated by grasslands and low shrub vegetation. Similar data have been found in our previous studies, as well as in the work of other colleagues (Mhuireach et al., 2016; Grigorova-Pesheva & Peshev, 2025). Specifically, green zone T2 once again shows high levels of microbial abundance. This is a park area that is built at a lower level than the street. During sampling, dust swirling and its transfer to the park area were observed. Similar to previous studies, higher levels of both fine dust particles and microorganisms were observed here again. Such results confirm the need to comply with the requirements and guidelines of environmental hygiene when carrying out urban planning. As indicated by (Revich, 2022), green areas

increase not only the physical, but also the mental health of the population, therefore their proper planning is key to building an environmentally friendly urban environment.

In the present study, no direct correlation was found between the increase in fine particulate matter and microbial abundance. (*Pearson correlation matrix, significance thresholds 95%, $p<0.05$*). However, there is a clear relationship between the reduced dust level and the higher percentage of woody vegetation. As Cohen et al. (2014), point out in their study, the presence of trees strongly reduces the amount of dust in green areas, which leads to an increase in air quality.

The specific dynamics of the results prove that when analyzing it is necessary to consider the data in pairs, green zone, hot spot, because depending on the specific conditions and dynamics of the characteristics of the environment, specific differences are observed both in the fine dust load and in the microbial abundance. However, the influence of the way the green zones are managed on the amount of PM reported and the total number of airborne microorganisms is undeniable. So again, at T3 (Hunting Park), where we have a green area imitating a natural dense forest, the levels of microorganisms are the lowest (2.55 CFU*10^2/m^3), including much lower levels of dust compared to the nearby heavily trafficked intersection (T4). Although the sample in the green zone was taken only 70 meters inland, in the park, a strong positive impact on air quality is observed. Similar results are also obtained for T7 (3.79 CFU*10^2/m^3) and T8 (3.00 CFU*10^2/m^3), which are located in Borisova Gradina - one of the largest green zones in Sofia, which has a large percentage occupied by woody vegetation - not accidentally called the "Tsar's Garden". Interesting data are for T11 (3.95 CFU*10^2/m^3), this is a green zone that has a smaller percentage of woody vegetation, but is closed on two sides with pedestrian zones, and in the center, there is a large water area. This again shows how specific urban planning can improve air quality, not only by increasing the percentage of woody vegetation, but also by reducing the intensity of car traffic.

The results for the percentage distribution of the individual microbiological groups are presented in Figure 4.

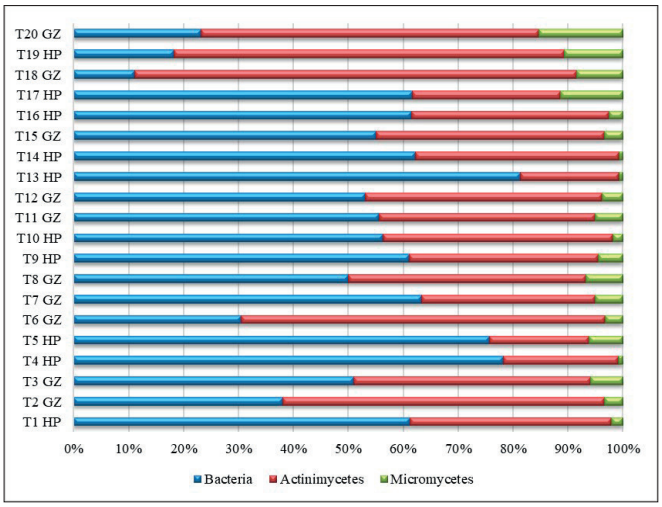


Figure 4. Percentage distribution of individual microbiological groups

The results show a very low percentage participation of the micromycetes group, which is probably due to the reduced humidity for the season.

One of the main microbiological parameters related to the vitality of microorganisms is the relative humidity (RH). Research shows that humidity levels in the range of 40-60% can be suppressive for many airborne microorganisms due to specific physicochemical processes in aerosol particles. However, most studies focus on indoor air quality, indicating that the optimal

humidity at which the possibility of transmitting pathogens and other microorganisms is seriously reduced is 40-60% (Skanata et al., 2022). There is insufficient data on the effect of humidity on airborne microorganisms in outdoor environments. Our data show that as the % humidity increases, the percentage of the micromycetes group (T17, T18, T19 and T20) increases. There are other studies available that prove that as humidity increases, the number of airborne fungi increases, explaining that high humidity levels facilitate the growth of this microbial group (Sajjad et al., 2023).

The results for the percentage of pigment forms, as well as the reported UV index levels, are presented in Figure 5.

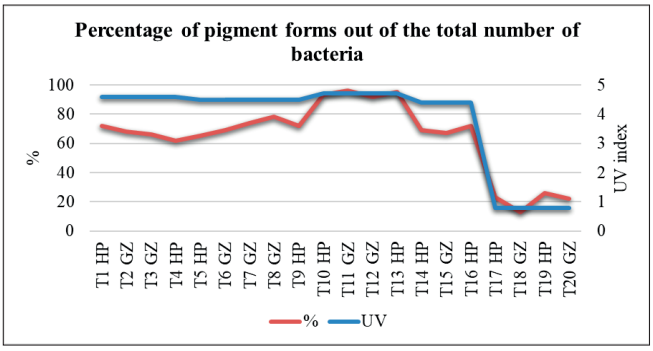


Figure 5. Percentage of pigment forms relative to the reported UV index

The results of the percentage participation show an increase in the number of pigment forms relative to the total number of bacterial colonies with increasing UV index. Similar results were also found by Tong & Lighthart, 1997. As the authors point out, pigmentation is a protective mechanism that ensures the survival of bacteria at high levels of UV radiation. At high UV index, non-pigmented bacteria, which are sensitive to light, die rapidly in the atmosphere. This leads to a relative increase in the proportion of surviving microorganisms that form pigment forms, which then form colonies on the nutrient medium in microbiological analysis (Figure 5). The increase in the percentage of pigmented colonies at higher UV index is a direct result of the defense mechanisms of microorganisms against intense solar radiation and the process of selective pressure in the atmosphere. Namely, such high results for the percentage of pigment forms indicate the strong adaptability of the air microflora (Park & Fowler, 2026).

It is important to note, however, that most studies indicate that the maximum proportion of pigment forms during peak hours of UV radiation is around 50-60% (Tong & Lighthart, 1997), while in the present study the results indicate higher values, which suggests that the air microflora probably responds not only to an increase in the UV index, but also to additional stress factors such as increased oxidative stress, increased concentration of pollutants in the air, etc.

CONCLUSIONS

The main objective of this study was to analyze the dynamics of airborne microorganisms according to environmental conditions. A total of 20 sample points were studied - 10 green areas and 10 hot points. No direct correlation was established between the fine dust load, regardless of the fraction, and the microbial abundance. The results confirm the need to analyze the data in pairs - green area - hot spot. The results indicate that in green areas with a high percentage of woody vegetation, both the microbial load and the fine dust load are lower. The present study confirmed the importance of urban planning. Thus, for park areas located below the roadway level, increased levels of PM and microorganisms were found. The present study found that the microbial community is influenced by environmental factors – humidity and UV index. Thus, in samples with increased % humidity, the participation of the micromycetes group increases. It was also found that an increase in the UV index leads to an

increase in the percentage of pigment forms regardless of the study point. This pilot study can serve as the basis for an expanded study covering major cities in Bulgaria to account for the influence of regional climate on shaping the microbial air community.

ACKNOWLEDGEMENTS

This research work was carried out with the support of Research Fund of Ministry of Education and Science in Bulgaria, project №: КП-06-Н74/5 „Relationships between the amount of fine particulate matter and airborne microorganisms in "hot spots", park areas, urban background areas and control forest areas''.

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