

## OPINION

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Member of the Scientific Jury, confirmed by Order No. RD-38-388/13.07.2022 of the Rector of the Sofia University "St. Kliment Ohridski" Prof. Dr. Anastas Gerdjikov, DSc

**Subject:** Ph.D. thesis for acquiring the educational and scientific degree "Doctor" to Boyanka Nikolaeva Angelova, a full-time Ph.D. student at the Department of "General and Industrial Microbiology" on the topic: "**Characterization of the microbiome in the complex study of particulate matter (PM) in the atmosphere of urban areas and risk assessment**", Professional direction 4.3. Biological Sciences, Scientific specialty Microbiology

Boyanka Angelova's Ph.D. thesis is an in-depth, interdisciplinary study of air pollution with microbial bioaerosols and particulate matter (PM), in an area of the highly urbanized part of the city of Sofia, namely the roof space of the Faculty of Biology. The location of the city, its active infrastructural reconstruction, and heavy traffic in recent years create conditions for strong atmospheric pollution, which contributes to negative climate changes, as well as poses a serious threat to human health. In her research, the Ph.D. student conducts one-year microbiological monitoring of the quantitative content of microorganisms in the air, as well as tracks the interrelationship between air quality, the presence of particulate matter, and concentration and type of bioaerosols. The lack of sufficient scientific data regarding the year-round dynamics of microorganisms in the air and the fact that no similar full-scale monitoring study has been conducted in the territory of the city of Sofia determines the relevance of the presented research.

In addition to the above, the dissertation contains new results on spatio-temporal air pollution with fine dust particles and their composition, structure, and morphology. In this regard, the presented complex research is the first of its kind on a national scale and provides valuable information about the dynamics and sources of air pollution in the capital, which must be taken into account when developing strategies to protect air purity.

The presented dissertation is written in 300 pages and is structured in 13 sections, the first 9 being the standard sections for this type of scientific work, namely: Introduction (1 page), Literature review (63 pages), Aim and Objectives (1 page), Materials and Methods (22 pages), Results and Discussion (150 pages), Conclusions (2 pages), Contributions (1 page),

Recommendations (1 page), and References (30 pages). A large number of cited literary sources - a total of 641 titles deserve special attention.

The dissertation work is illustrated with 112 figures, 44 tables, and an appendix, which presents in tabular form the morphological and physiological-biochemical characteristics of the microorganisms isolated from the air. A very good impression is made by the synthesized list of a large number of figures and tables at the beginning of the thesis, in order to make them easier to perceive and reflect on.

The title of the dissertation fully corresponds to its content. The introduction is written briefly and concisely and logically outlines the problem of the complex nature of air pollution over highly urbanized areas and the need for in-depth research in this direction, tracing the relationship between air quality, the presence of pollutants, and the level of the microbial component associated with dust pollution.

The literature review presents in detail the composition of microorganisms and the main pollutants in atmospheric air, the standards that are applied in the EU for air quality and the monitoring systems in the city of Sofia, as well as the methods and approaches for collecting and studying bioaerosols and fine dust particles from the air. This section has been written after reviewing a large number of literature sources and the information is very well summarized and analyzed.

The aim of the dissertation is focused on a full-scale complex one-year study of bioaerosol contamination levels, with the identification of dominant microbial species, and full characterization of associated particulate matter pollution. It is realized through the implementation of 5 main tasks.

The microbiological work of the Ph.D. involved the use of a variety of culture-dependent and culture-independent approaches for the taxonomic determination of airborne bacterial and fungal isolates.

The lidar monitoring and physicochemical characterization of the PM were done respectively in the Institute of Electronics and the Institute of Catalysis, Bulgarian Academy of Sciences. As can be seen, the Ph.D. student has mastered a large number of modern microbiological, genetic and physicochemical analysis methods, which are precisely described in the Materials and Methods section.

As a result of the quantitative microbiological monitoring carried out, based on 54 weekly samplings within a full calendar year, new data were obtained regarding:

- the number of bacterial and fungal bioaerosols in the air of the studied location.
- the monthly and seasonal dynamics in the levels of both types of bioaerosols, with higher values recorded in spring, the first half of the summer season, and autumn, while significantly lower levels of microbial bioaerosols were detected in the winter.
- the different dominant presence of bacteria and fungi in the air during different periods of the year.
- the influence of anthropogenic activity on the daily and weekly dynamics in the concentration of microbial bioaerosols.
- the impact of certain meteorological phenomena (fog, rain, snow, and transboundary dust pollution) on the number of microbial bioaerosols.

As a result of the performed cultivation-dependent and metagenomic analyses, original results were obtained regarding the taxonomic diversity of the air microbiota in the studied air samples from the selected location. An extremely high diversity of culturable and non-culturable bacterial and fungal taxa was found, with the bacterial microbiota being dominant.

New results were obtained regarding the morphology and chemical composition of the dust content (PM<sub>10/2.5</sub>) in the air of the investigated area, as well as a directly proportional relationship between the concentration of PM and microorganisms in the air, was established.

Based on the data from the conducted complex research, 14 conclusions were formulated, four recommendations were made and 7 contributions were indicated, which emphasize the originality of the conducted research on a national scale and reveal the importance of the obtained scientific results in the development of strategies for control, risk assessment and protection of the air purity.

The results of the dissertation work were published in two articles in a scientific journal with an impact factor and were presented at 5 international and 3 national scientific forums. To realize the experimental work on the dissertation, the doctoral student participated in 3 scientific projects.

The abstract attached to the thesis has a volume of 64 pages and is prepared as a synthesized version of the dissertation, which fully reflects all its main sections and is well illustrated with figures and tables.

In conclusion, I would like to summarize that the submitted dissertation and accompanying documents fully meet the legal criteria for obtaining the educational and scientific degree "Doctor". In view of the large-scale and in-depth research conducted and the contributions derived from it, the use of various modern methods, and the importance of the obtained original results from a fundamental and applied perspective, I give my positive assessment and recommend the Scientific Jury to award the educational and scientific degree "Doctor" to Boyanka Angelova.

25 of August 2022 г.

Sofia

Member of the Scientific Jury:

/Assoc. prof. Anna Tomova/