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Characterization of Fungi Bioaerosols in Family Farming Systems: Municipality of Turbaco, Colombia

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Abstract– ***Bioaerosols impact the environment, agriculture, and human health by influencing air quality, cloud formation, and climate patterns. In agriculture, they act as vectors of pathogens that harm crops but also disperse beneficial spores that enhance plant nutrition. This study aimed to identify fungal species in the air and evaluate their relationship with a family farming system. Systematic samples were collected with three replicates using a cascade impactor at various points within the agricultural system. The samples were processed in the laboratory to isolate and identify fungal species through cultivation techniques, using PDA (Potato-Dextrose-Agar) as the growth medium. Detailed macroscopic and microscopic observations were conducted to examine the morphology and characteristics of the colonies. Fungal concentrations in the air were also quantified, and the relationships between the identified species and the agricultural system's characteristics were analyzed. Preliminary results showed fungal growth in all culture plates, with notable genera such as Fusarium (phytopathogen), as well as others like Aspergillus, and Mucor. This analysis highlights the importance of understanding fungal diversity in the air and the need for continuous monitoring to mitigate its impact on crops and manage phytopathological diseases.***

Keywords– ***Aerobiology, Air quality, Curvularia, Fusarium, Phytopathogenic fungi.***

I. INTRODUCTION

Bioaerosols are recognized as fast dispersing and wide-ranging vectors, due to their small size and light weight that allows them to be transported by small air currents through different atmospheric layers. This ability allows them to reach diverse environments, from soils to water bodies and urban areas [1], [2], [3]. Thus, bioaerosols facilitate biological redistribution in different ecosystems. Compared to other transport mechanisms, bioaerosols can more rapidly alter the dynamics and structuring of microbiogeography [4], as they which can introduce microorganisms into previously uncontaminated sites. In addition, bioaerosols can act as condensation nuclei in the atmosphere, influencing the formation of clouds, ice crystals and precipitation, which affects the hydrological cycle and the climate of the environment reached [5],[6],[7]. These aerosols can be effective carriers of pathogenic microorganisms (live or dead) thus constituting a risk factor in the spread of diseases that can negatively affect impacted biota [4].

A general classification of bioaerosols, based on their composition, categorizes them into three main groups: a) viruses and parasites, b) living organisms, including bacteria and fungi, and c) microbial components or by-products, such as plant spores, pollen, endotoxins, and allergens of animal origin [1],[2],[5]. Fungi comprises 30% of the total bioaerosols present in the atmosphere [8]. While, fungal spores are usually not only fungal dissemination units, they can also transport mycotoxins, allergens, bacteria and pollutants.

Depending on the fungal species, spore contributions can be favorable and/or detrimental. Of the nearly 100,000 fungal species recorded, 8,000 correspond to phytopathogenic species responsible for about 80,000 plant diseases. In addition, approximately 50 species are pathogenic to humans and animals, while the rest correspond to saprophytic and symbiont species [9],[10],[11]. Phytopathogenic fungi, although they represent a small fraction of fungal diversity, their impact on the productivity of cropping systems is significant, given that, they can reduce plant yields and can even compromise food quality, and, consequently, increase the risk of mycotoxin contamination. Therefore, accurate identification of fungal species is essential to ensure crop health and sustainability of agricultural systems [12],[13],[14],[15],[16].

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Meanwhile, the increasing world population requires a substantial increase in food production to meet future demands, which highlights the critical need for effective crop disease control. Understanding bioaerosol dispersion, seasonal variations, and composition is crucial for developing improved strategies to manage crop diseases and increase yields [17]. Since it is estimated that approximately 30% of global agricultural production is lost due to diseases caused by pathogens, it is imperative to thoroughly investigate how bioaerosols contribute to the spread of these organisms and the implications this has for agriculture [17]. Also, Pathogenic or infectious bioaerosols can cause respiratory infections after penetration into the respiratory system of humans or animals [18].

The warm and humid tropical climate of the department of Bolivar, located on the northern coast of Colombia, favors fungal growth when there are sufficient nutrients [19], so the department constitutes a focus of fungal spore dispersal. Now, despite the importance of research on bioaerosols and fungal species to promote public health and local agriculture, currently in Cartagena de Indias there are only three previous studies in open environments; the first evaluated the impact of air pollutants, such as bioaerosols, on asthma and other respiratory diseases [20]; the second focused on quantifying and characterizing bioaerosols at Blas El Teso beach [6], and the third, on determining fungal bioaerosol concentrations in an IES [21].

Therefore, this study aims to: (1) conduct a spatially dispersed sampling of bioaerosols in a family farming system in the municipality of Turbaco to identify the fungal species present in the air; (2) determine the concentration of fungal spores in the air within the family farming environment using a cascade impactor; and (3) analyze how factors such as temperature, humidity and type of surrounding crop influence the presence and concentration of isolated fungal spores in the air.

II. METHODOLOGY

The main methodological stages developed during this study are shown below:

A. Sampling Site

This study was conducted in the city of Turbaco, department of Bolivar - Colombia is characterized by a tropical savanna climate, moderate to high humidity, and an average temperature of 30 °C. The rainy season generally occurs between May and November, with heavy rain. The dry season runs from December to April, with minimal rainfall (Ideam). The sampling site was located at Agricola Camelias (10° 20' 0.08“ N - 75° 22' 51.33” W). The locations of the sampling sites are shown in Fig. 1.

Figure 1. Location of the sampling site (https://online.mapcreator.io).

B. Sample collection and processing

The sampling instruments were placed at a height of approximately 1.5 m, which corresponds to the average height of human respiration [21],[22]. Bioaerosol sampling was conducted triplicate once a week in June 2023, and samples were collected in the morning (9:00 AM-10:00 AM). The sampling time was 10 minutes. Samples of culturable fungi were collected on Petri dishes containing PDA agar and incubated at 26 °C for 8 days. The methodology used was the one proposed by [6] and [21]. Using a non-viable cascade sampler TISCH Environmental® brand. Meteorological data (temperature, precipitation, wind speed and humidity) were obtained using the Davis Vantage Pro® weather station (Fig. 2).

Figure 2. (A) non-viable cascade sampler TISCH Environmental®. (B) Weather station, Davis Vantage Pro®

C. *Identification of the collected biological material*

The visible characteristics of the colonies (color, texture, shape, and size) were recorded daily during the 5 days of growth. Additionally, a colony count was performed, and the data were recorded in an Excel spreadsheet corresponding to the three collected replicates. Subsequently, fragments of the colonies were stained with methylene blue and observed under an optical microscope to evaluate fungal structures such as hyphae, spores, and conidia. The data obtained were photographically documented and compared with specialized literature for identification, following the methodologies proposed by [6] and [21].

III. RESULTS AND ANALYSIS

Bioaerosols have a significant impact on the environment, agriculture, and human health. They affect air quality, contribute to cloud formation, and alter climate patterns. In agriculture, they can transmit pathogens harmful to crops but also disperse beneficial spores that enhance plant nutrition. The results obtained at each stage of the procedure are described below:

A. *Macroscopic observation of fungi*

From bioaerosols collected using a cascade impactor and grown on PDA medium, cultures with a variety of morphologies and colors were observed. Among them, colonies with cottony textures and white, black, green and brown shades were identified, as well as circular shapes with defined and irregular edges, and slightly convex elevations (Fig. 3).

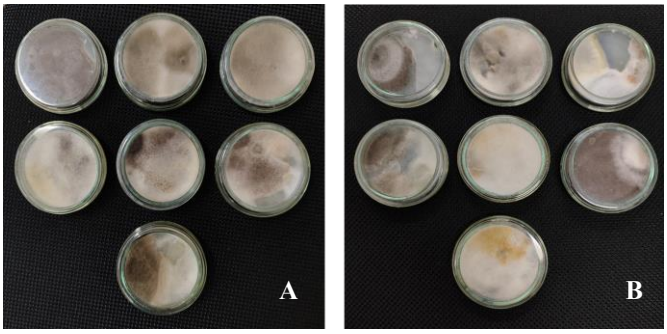

Figure 3. Macroscopic characterization of fungi. The fungal colonies obtained in replicates 1 and 2, corresponding to (A) and (B), respectively, are presented below. These images show the distinctive characteristics of the colonies, including variations in their texture, coloration and morphology.

Specific differences were observed between each replicate, which could be attributed to variations in environmental conditions, such as temperature, humidity or air flow, as well as differences in the composition of the bioaerosols collected. These factors could have influenced the diversity and abundance of microorganisms present, highlighting the importance of controlling these variables in future studies to ensure greater consistency in the results [6]. Currently, most of the fungal species detected are in the process of identification and confirmation.

B. *Microscopic observation of fungi*

The fungal structures observed by optical microscopy with a 40X objective allowed the identification of the following fungi at this stage of the investigation. In Figure 4 (A), a spore belonging to a fungus of the genus *Fusarium*, classified within the phylum Ascomycota, is presented. These fungi are usually found mainly in their mold form. Filamentous, septate, branched hyphae were observed, along with long, septate, falciform-shaped conidia, which may develop in chains or clusters [5], [21]. Figure 4 (B) illustrates a representative of the genus *Aspergillus*, also of the phylum Ascomycota. In this case, hyaline filamentous hyphae, conidiophores and conidia characteristics of this group of fungi were evident [6],[21]. In Figure 4 (C), structures characteristic of the phylum *Zygomycota* are shown, such as coenocytic hyphae and sporangiophores supporting sporangia, which can be simple or branched. These characteristics suggest the possible identification of a fungus of the genus *Mucor*. Some members of this genus are saprophytes and are commonly found in environments rich in organic matter, such as fruits, vegetables and soils [6],[21].

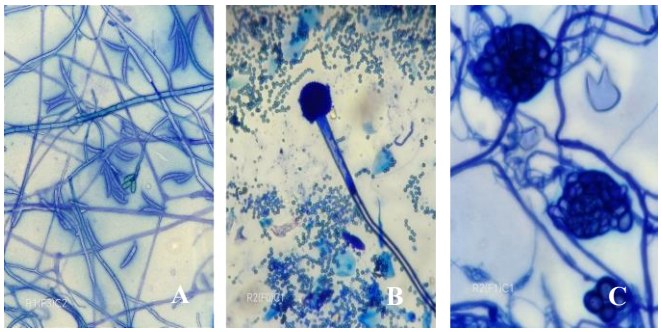

Figure 4. Microscopic characterization of fungi: Falciform-shaped conidia observed in fungi of the genus *Fusarium* (Fig.

4A). Characteristic conidiophores and conidia identified in fungi of the genus *Aspergillus* (Fig. 4B). Cenocytic hyphae and sporangiophores supporting sporangia observed in fungi of the genus *Mucor* (Fig. 4C).

C. Fungi count obtained during the analysis of two replicates

An analysis of the collected samples was carried out, in which the quantities and types of fungi present were recorded. This detailed count not only allowed us to quantify the mycological diversity in each replicate but will also allow a comparison of the composition and abundance of fungi in the different sampling environments and future research. Meanwhile, two replicates were performed for each count, and from them averages, maxima, minima, variance and standard deviation were calculated. The average values of the counts ranged from 24 to 32, depending on the data set. It was observed that the maximum and minimum values presented significant differences in some cases, suggesting some variability in the data. Variance (Var P) is a measure of the dispersion of the data. It was observed that the highest values of variance corresponded to the second column (Count 2), indicating greater dispersion in these data. In contrast, the first set of counts (Count 1) showed relatively low variances (9 and 16), suggesting that the data are more clustered around the average.

The standard deviation (DesvEst P), which is the square root of the variance, showed that Count 2 and Count 3 have greater dispersion compared to Count 1. On the other hand, more replicates may be needed to obtain a better estimate of the true average and decrease the influence of outliers (Table 1).

Table 1. Data analysis: fungal colony counting in two replicates.

	Count 1		Average 1 Replicate	Count 2		Average 2 Replicate	Count 3		Average 3 Replicate
	1	2		1	2		1	2	
Replicate 1	20	20	20	27	29	28	29	31	30
Replicate 2	26	30	28	22	35	37	33	40	29
Average Count	23	25	24	25	32	32	31	36	29
Max	26	30	28	27	35	37	33	40	30
Min	20	20	20	22	29	28	29	31	29
Var.P	9	25	16	6	9	18	4	20	1
DesvEst.P	3,0	5,0	4,0	2,5	3,0	4,3	2,0	4,5	0,8

Figure 5 shows the visual count of fungal colonies from two replicates in different conditions or categories (Count 1, Count 2 and Count 3), as well as their average values.

The main observations are detailed below:

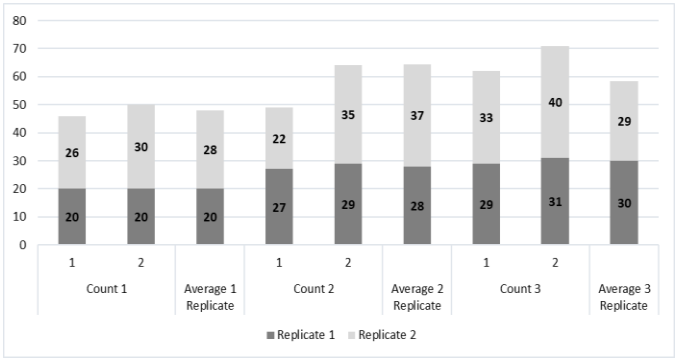

Figure 5. Visual fungal colony count.

In each count, the second replicate (Replicate 2) presented higher values than the first replicate (Replicate 1), indicating variability in the results obtained. The most notable difference between replicates was observed in Count 3 (1), where Replicate 2 reached a value of 40, while Replicate 1 registered 31, representing a significant difference. The average values for each count reflect this trend: 24, 32 and 29, respectively for each of the replicates. This indicates a greater presence of colonies in Count 2, with a slight decrease in Count 3 (Fig. 5). Simultaneously, with these observed results, we will continue with the procedures for the identification of fungal microorganisms.

IV. Discussion

Bioaerosols are airborne particles ranging in size from 0.001 to 100 μm, comprising various biological agents such as bacteria and fungi, both living and dead, pathogenic or non-pathogenic. They also include viruses, endotoxins, mycotoxins, peptidoglycans, β(1,3)-glucans, allergens, and pollen [23],[24].

Since individuals spend approximately 80% to 90% of their time indoors, with even longer durations for children, the elderly, and patients [25], indoor exposure to bioaerosols has a significant impact on health. Various studies have demonstrated that bioaerosol exposure is associated with several human health risks, including pneumonia, cancer, asthma, and respiratory syndromes [26]. Additionally, they act as vectors for the propagation and dissemination of pathogenic microorganisms, affecting both humans and crops, potentially impacting public health and agricultural productivity [17].

This study identified fungal bioaerosols in small-scale farming systems, highlighting the importance of implementing monitoring and control strategies to reduce risks in rural communities, both for human health and crop phytosanitation. Thus far, fungi belonging to the genera *Fusarium*, *Aspergillus*, and *Mucor* have been identified. Notably, metabolites from *Aspergillus*, *Penicillium*, and *Fusarium* were predominant in air and dust samples [27]. Furthermore, studies have shown that the average cytotoxicity of air samples in hospital rooms varies depending on the *Aspergillus* species present [28]. In summary, variations in microbial composition and abundance influenced cell viability and proliferation levels [17].

Previous studies on airborne fungi have reported the presence of these genera in Cartagena, Colombia. The predominant genera were *Aspergillus* (60.8%), followed by *Penicillium* (24.5%) and *Fusarium* (9.3%). *Fusarium* sp., the third most frequent genus in this study, is a group of filamentous fungi widely found in soil and plants. It is considered opportunistic due to its ability to grow at high temperatures and can cause systemic infections in immunocompromised patients. Additionally, some species produce toxins that can affect humans and animals [29]. These findings align with previous studies that have identified *Fusarium* as an important genus in outdoor environments [21], [30].

The presence of *Aspergillus* sp. is highly common in the air due to its adaptability to environmental changes and has been previously reported in bioaerosol studies [5],[31]. Moreover, this genus has been documented as highly abundant in tropical and subtropical regions [21],[31]. *Mucor* species are widely distributed in nature and can be found in various substrates, with a higher prevalence in humid environments. Their spores, released in large quantities, have a remarkable ability to absorb moisture and adhere to different surfaces, facilitating their dispersion in humid air [32].

These findings emphasize the need for continued research on the presence and distribution of fungal bioaerosols in agricultural and urban environments. Identifying genera such as *Aspergillus*, *Fusarium*, and *Mucor* reinforces the necessity of monitoring and control strategies to mitigate their potential impacts on human and plant health. Furthermore, understanding their dynamics under different environmental conditions will enable the development of more effective preventive measures. Therefore, future studies should focus on assessing their viability, toxicity, and potential long-term effects.

V. CONCLUSIONS

This research on bioaerosols in family farming systems in the municipality of Turbaco, Colombia, allowed the preliminary identification of the presence and diversity of microorganisms in the air. It highlights the importance of implementing monitoring and mitigation strategies to minimize the possible risks associated with exposure to bioaerosols, especially in rural communities where agriculture is a fundamental activity. These findings add to the knowledge of bioaerosol dynamics in agricultural settings and underscore the need for ongoing studies to assess their long-term impact.

The variability in counts could be influenced by experimental factors, such as environmental conditions, errors in visual counting or differences in fungal colony growth. To confirm the observed trend and reduce variability in the results, further replicates are recommended in future studies.

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