



Analyses of culturable microorganisms and chemical pollutants in the air of urban and rural areas in the region of São Paulo, Brazil

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Abstract Bioaerosols are particles of great importance for several fields of research, and spores produced by fungi can exist as bioaerosols when suspended in the air. Microbiological standards for environmental monitoring of outdoor air parameters can be achieved by analyzing the relationship between airborne microorganisms and the prevailing environmental conditions. The outdoor air of the Metropolitan Region of São Paulo and the rural area in a city of the state of São Paulo (Ibiúna/SP), both in Brazil, were evaluated for the presence of microorganisms using the MAS-100 ECO (Merck®, Fr.) and M Air T (Millipore®) air sample collectors. Dichloran Rose-Bengal Chloramphenicol and Tryptic Soy Agars were used for fungal and bacterial isolation, respectively. Bacterial colonies were counted, and the plates with fungal colonies were sent for phenotypic

identification up to genus and species level, respectively. Data on pollutant concentrations were obtained from the Environmental Company of the State of São Paulo. The highest number of Colony-Forming Units/m³ (CFU/m³) of microorganisms was measured in the winter and summer seasons, respectively, but the greatest Spore-Forming Units (SFU) of fungi were found in the rural area, where pollutant concentrations were lower. Nitrogen dioxide (NO₂) had a slightly positive influence on the concentration of SFU of fungi in both areas studied. Sulfur dioxide (SO₂) pollutant concentrations had both positive and negative great relations showing influence on microbial counts in the air of the rural area. In the rural area, the low bacteria count was influenced negatively by the low concentration of carbon monoxide (CO). The microbial counts were related to each other, as well as to the concentrations of pollutants, shown by all the correlations seen, indicating microorganisms as biomarkers of pollution in outdoor areas. The influence of environmental factors on the population and outdoor air biome is also explicit.

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1 Introduction

Aerosols are particles of great importance for several areas of research involving atmospheric chemistry, physics, biosphere, climate, and public health (Farmer & Riches, 2020; Pöschl, 2005). These aerosols can be divided into two fractions: the fine fraction, less than 2 μm and produced mainly by the conversion into particles of gases, and the coarse fraction, with particles greater than 2 μm (Theotônio et al., 2007; Andreeva et al., 2024). Aerosols of biological origin are generally part of this last fraction and are called bioaerosols, which consist of fragments of leaves, pollen grains, and fungi (Theotônio et al., 2007; Andreeva et al., 2024).

At certain places and times of the year, bioaerosols contribute up to 50% of the total number of aerosols or particles in the air (Morris et al., 2004; Pescott et al., 2015; Yao, 2018). Bioaerosols can change atmospheric thermodynamic properties such as temperature profile and variability of relative humidity, and they depend on these factors to circulate in the environment. The average length of stay in the atmosphere is variable, ranging from days to weeks. Many of these bioaerosols have defense mechanisms that allow them to withstand the environmental stresses of air transport, including exposure to UV radiation, dehydration, and pH in the cloud water, and also to survive long-range travel (Andreeva et al., 2024; Burrows et al., 2009; Farmer & Riches, 2020; Rosenfeld et al., 2008).

These particles are not suspended in the air as independent elements, and cell agglomerations or airborne transport in plant or animal fragments, soil particles, pollen, or spores may occur. The intact particles that are part of bioaerosols have several sizes: Most pollen types grains are 17–58 μm , fungal spores are 1–30 μm , bacteria are 0.25–8 μm , and viruses are less than 0.3 μm in diameter (Jones et al., 2004).

Kingdom Fungi can produce bioaerosols including fragments of hyphae, single-celled, and multicellular spores, allowing their dispersion. In general, diversity is inherent to the size of fungal particles, associated with the strong influence of their diameter. The taxonomic composition of these anemophilous fungi can determine how they will be distributed in the atmosphere. All fungi are active participants in the cycle of elements in nature; approximately 28 to 50 tons of fungal materials are emitted annually in the Earth's

atmosphere, and fungal particles can constitute up to 420% of primary organic aerosol emissions (Bernardi, 2007; Elbert et al., 2006; Heald & Spracklen, 2009; Yamamoto et al., 2012).

Microbiological analysis of the air can be performed by several techniques, but sample collection using impactors can determine the exact value of the air analyzed; due to its flow control, it can determine the amount of air collected (Lacey & Venette, 2020). A successful fungal analysis depends largely on the used technique, as well as the choice of the appropriate culture media in the collection, which allows a comprehensive evaluation of quantitative and qualitative characteristics (Gutarowska & Piotrowska, 2007). The characterization of bioaerosols, of fungal origin, can be done by analyzing small air samples that can provide a reasonable estimate of the typical concentration of spores. These analyses may allow the identification of potentially harmful fungal contamination, even when surface colonies are not easily visible (Egan et al., 2014).

Parameters on microbiological air conditions could be performed by analyzing the relationship between airborne fungi and bacteria present in outdoor air and the environment where they are isolated. However, other microorganisms are used in environmental controls, for their sensitivity to pollution, especially in rural areas (Martins et al., 2008; Munzi et al., 2007; Ristic et al., 2017; Stamenković et al., 2016).

On the other hand, according to the World Health Organization (World Health Organization, 2003), air quality standards are variable according to the approach adopted to balance health risks, technical feasibility, economic considerations, and various other political and social factors, which in turn depend, among other things, on the level of development and the national capacity to manage air quality, in which the number of spores in the air is one of the factors (Resolução CONAMA N° 491, 2018).

The measurement of anemophilous microorganisms is necessary to evaluate the effectiveness of microbial control strategies integrated by environmental monitoring.

Pollution influences on respiratory disorders have a great impact on public health and are related to anthropic activities producing pollutants, whether particulate matter or toxic gases such as sulfur dioxide (SO_2), nitric dioxide (NO_2), and carbon monoxide (CO), among others, which also cause enormous

inferences to the environment (Martins et al., 2002; Parsi and Görecki, 2006; Jasinski et al., 2011).

In Brazil, environmental control analyses of outdoor air are carried out characterizing the aerosols. The organic fraction has been mainly related to biomass burning and combustion, although there is a significant presence of green areas in cities that make biogenic emissions an additional source of organic carbon (Rackes & Waring, 2013; Ana Paula Mendes Emygdio, Cristiane Degobbib, Fábio Luiz Teixeira Gonçalves, 2018). This control does not focus on microorganisms in the atmosphere.

The Metropolitan Region of São Paulo (MRSP) is characterized by a large megalopolis with a high population and number of vehicles. This condition favors the emission on a scale of pollutants (particulate matter and toxic gases), creating its own biome in relation with the atmospheric and biological characteristics of the air of the region. The large circulation of people and vehicles observed in highly populated and industrialized regions can produce a high dispersion of microorganisms in these areas (Chiquetto et al., 2021).

Several types of researches conducted with environmental samples in China and India showed a strong correlation between air pollutants and the diversity of microorganisms, using culture techniques for the bioaerosols identification, showing the influence of environmental factors on the concentration of Colony-Forming Units/m³ (CFU/m³) of fungi and bacteria, correlating the concentrations of pollutants with the amount of CFU of microorganisms (Fan et al., 2019; Roy & Gupta Bhattacharya, 2020).

Identification of airborne fungi, especially those belonging to the Ascomycota phylum, can provide important information which, when related to atmospheric conditions or the pollutants concentration, would turn these microorganisms into strong predictors of environmental conditions (Roy & Gupta Bhattacharya, 2020).

In Brazil, there is a growing concern in monitoring indicators of environmental pollution and the need to expand the knowledge about these microorganisms in the atmosphere, as well as to analyze the relationship between these microorganisms and air pollutants; beyond that, we have a lack of studies on this board realized on Latin America. Based on this information, the aim of this study was to collect MRSP air samples

and analyze their relationships with air pollutant concentrations during the period.

2 Materials and methods

Outdoor air was evaluated in the MRSP and in the rural area of a city of the state of São Paulo (Ibiúna/SP) regarding the presence of fungi and bacteria, for six years, amounting to 736 collections; each collection presented a sample for fungi and another for bacteria, respectively.

Two points were analyzed: one in the city of São Paulo at the Adolfo Lutz Institute (IAL), located at Cerqueira César neighborhood. The rural area analyzed was the Votorantim neighborhood in the city of Ibiúna/SP. The two cities are 60.6 km apart (Table 1; Fig. 1). The first point is located at the downtown, and Ibiúna is considered rural.

The distribution of the collected samples is described according to the collection site and during the season of the year (Table 2).

Air was sampled using the air compactors MAS-100 ECO (Merck®, Fr.) and M Air T (Millipore®). Both have the same air flow capacity and final volume of sample collected (Moura, Caldas et al., 2015).

Three daily samples were collected at 1-h intervals during the morning (9 AM, 10 AM, and 11 AM), based on mutual schedules with highest flow of people and vehicles in the capital and the city of the interior. Each presented the final volume of 250 L (0.25 L/m³), totaling 750 L (0.75 L/m³), using the modified Dichloran Rose-Bengal Chloramphenicol (DRBCm) culture media (de Matos Castro e Silva et al., 2015) for isolation of fungi, while Tryptic Soy Agar (TSA) was supplemented with cycloheximide for count of CFU/m³ of bacteria.

Table 1 Geographic and numeric details of sites for collection of air samples in São Paulo and Ibiúna

Sites	Location	Number of samples collected	Coordinates
SITE 1	IAL	524	23°55'60" S 46°66'81" W 46°73'33" W
SITE 2	IBIÚNA	212	23°39'23" S 47°13'21" O

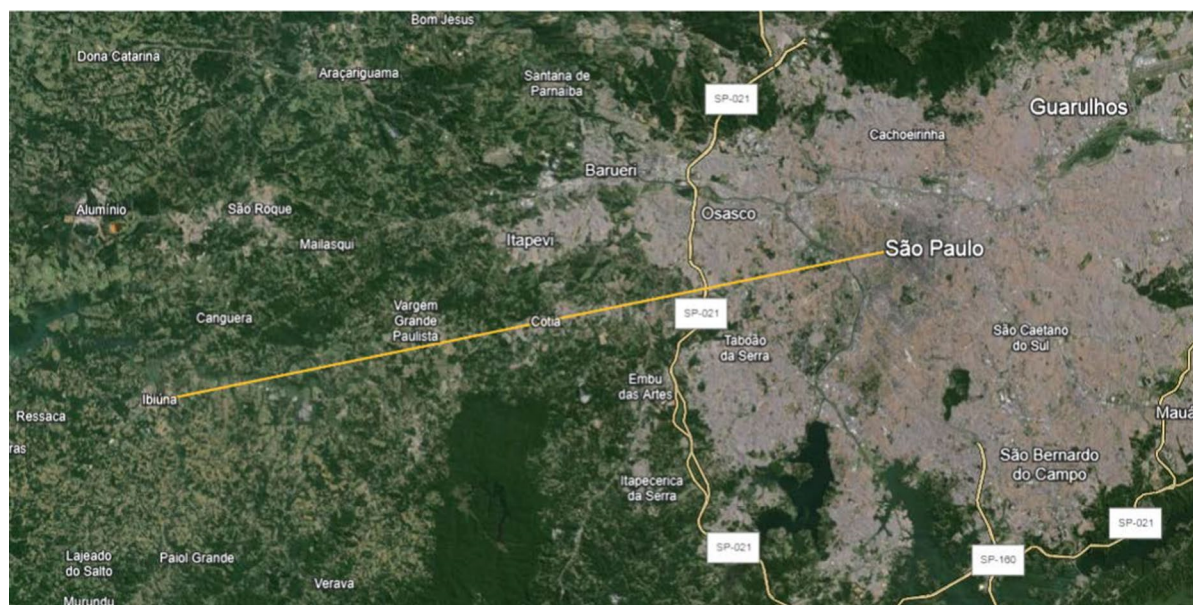


Fig. 1 Land-use sampling points

Table 2 Distribution of air sampling for a six-year period based on season, date, and collection site

Season	Collection year												Total
	2013		2014		2015		2016		2017		2018		
	R	U	R	U	R	U	R	U	R	U	R	U	
Winter	24	24	24	25	15	15	4	5	15	16	40	41	248
Autumn	41	42	–	–	–	–	4	4	–	–	62	63	216
Spring	21	21	8	8	–	–	4	5	39	39	–	–	145
Summer	–	–	–	–	–	–	4	5	9	9	50	50	127
Total	173		65		30		35		127		306		736

R: Rural area
U: Urban area

The samples collected from the TSA plates were incubated at 30 °C for three days. After this period, was performed the colony counting, and after that, the plates were discarded, since there is no need for more applied identifications for its correlation with the environment (Bragoszewska & Pastuszka, 2018).

The DRBCm media inoculated with fungi were incubated for up to seven days at 30 °C. The resulting fungal colonies after the period of incubation were counted, and only one isolate of each fungal genera was identified using phenotypic characteristics such as the macro-micromorphology and the presence of pigments (hyaline and dematiaceous), among others (Hoog, Guarro, Gené, 2014).

Data on pollutant concentrations at the time of air collection were obtained from the Environmental Company of the State of São Paulo (CETESB) in daily online reports of the stations: Cerqueira César, Pinheiros, and Sorocaba.

2.1 Statistical analysis

For statistical analysis, we applied the Kolmogorov–Smirnov test, but it was seen that none of the variables had normal distribution. Then, we performed the factorial analysis of variables and applied the Kaiser–Meyer–Olkin (KMO) and Bartlett’s tests to attest to the feasibility on factorial analysis, looking for the variance of the data, where the test could tell us

whether factor analysis was appropriate or not. Spearman's nonparametric correlation test was performed to verify the strength of the relationships between the variables, and where $p > 0.005$, we applied the Mann–Whitney U test to verify whether the comparison of two unpaired groups (≤ 100 and ≥ 101 CFU/m³) was statistically significant. All tests were performed using the Biostat software.

3 Results

No molecular analysis was performed to adjust or categorize the methodology errors due to the scarcity of resources for these analyses. All samples collected showed the presence of bacteria and fungi, respectively (Fig. 2).

Fig. 2 Collection plates with samples. **A:** DRBCm plates with fungal colonies. **B:** TSA plates with bacterial growth

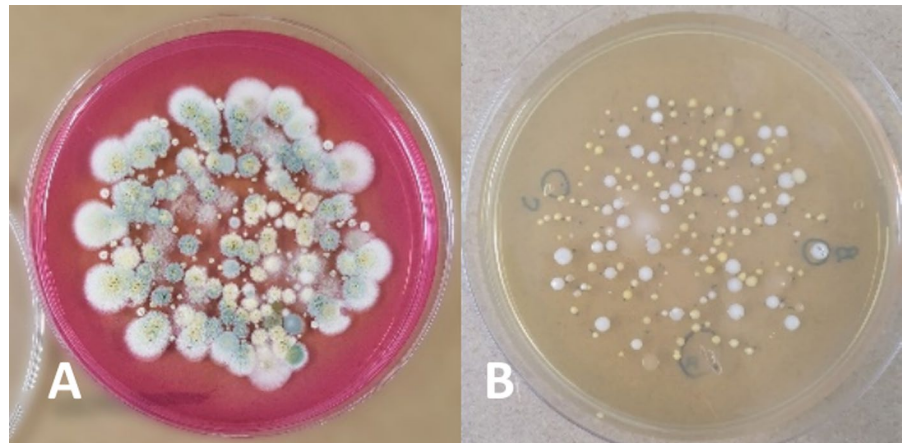
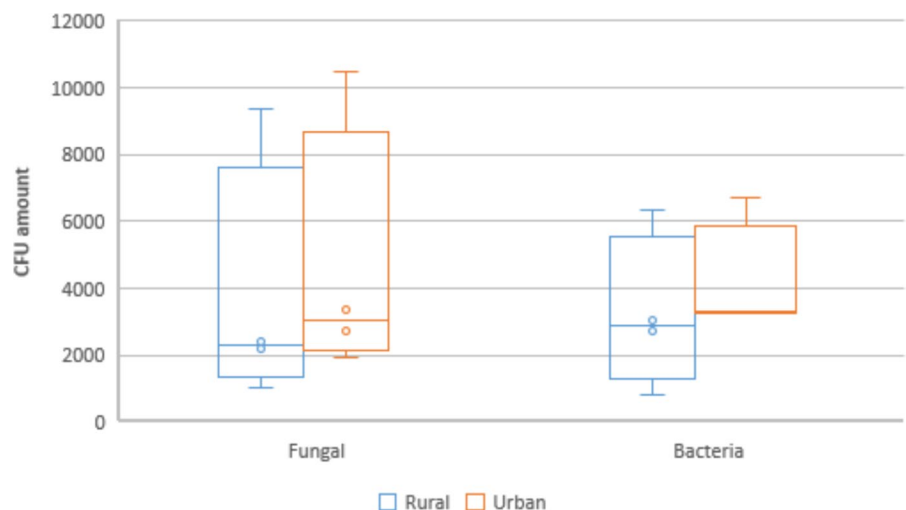


Fig. 3 Distribution of CFU/m³ of microorganisms in rural and urban areas by seasons



The concentrations of microorganisms were distributed in a similar way, but in the winter season, there was an increase in CFU/m³ of fungal spores, while in the summer season, the increase was in the CFU/m³ of bacteria (Fig. 3).

During sampling campaign, 1630 fungal isolates were obtained; 219 did not present reproduction structures and were classified as non-sporulating species. According to phenotypic analyses performed in 1411 isolates, 17 different genera were identified (Table 3).

The highest incidence of different fungal genera in the same air sample occurred in the rural area, with up to seven concomitant genera (Table 4).

Statistical analyses were based on factorial data represented in Table 5.

Table 3 Airborne fungi genera

Genera	Number of isolates	Percentile
<i>Acremonium</i>	2	0,14
<i>Aureobasidium</i>	2	0,14
<i>Alternaria</i>	62	4,39
<i>Aspergillus</i>	367	26,01
<i>Bipolaris</i>	8	0,57
<i>Cladosporium</i>	42	2,98
<i>Curvularia</i>	175	12,4
<i>Fusarium</i>	138	9,78
<i>Mucor</i>	9	0,64
<i>Neurospora</i>	141	9,99
<i>Nigrospora</i>	9	0,64
<i>Paecilomyces</i>	16	1,13
<i>Penicillium</i>	234	16,58
<i>Phoma</i>	6	0,43
<i>Rhizopus</i>	101	7,16
<i>Syncephalastrum</i>	9	0,64
<i>Trichoderma</i>	90	6,38
Total	1411	100

Table 4 Diversity of fungal genera in sample sites

Genera by sample	Urban	Rural	Percentile
1	197	41	32,87
2	197	59	35,36
3	92	51	19,75
4	29	32	8,43
5	12	10	3,04
6	–	3	0,41
7	–	1	0,14
Total	347	197	100%

Table 5 Factorial statistics descriptive about fungal and bacterial counts in relation to the air pollutants

Area		Mean		Standard deviation		Total (N)	
		Fungi	Bacteria	Fungi	Bacteria	Fungi	Bacteria
Urban	CFU/m ³	52.15	43.06	54.98	41.01	166	158
	SO ₂	20.68	21.66	33.76	34.32	166	158
	NO ₂	55.98	56.49	27.56	28.14	166	158
	CO	20.98	22.01	34.90	35.47	166	158
Rural	CFU/m ³	201.77	105.31	72.34	78.85	13	13
	SO ₂	5.95	5.95	2.78	2.78	13	13
	NO ₂	11.28	11.28	4.65	4.65	13	13
	CO	5.85	5.85	4.08	4.08	13	13

In the rural area, by the KMO–Bartlett's test, the analyzed data showed no significant relevance in the correlation of bacterial and fungal counts with the other variables, different from the relationship observed in the low positive influence of pollutant concentration on the number of bacterial counts (Table 6) dispersed on the air in the urban area ($p \leq 0.001$).

Analyzing the differences in the concentrations of SO₂ in the samples collected between urban and rural areas, the result was quite expressive. In the rural area, when it presented low rates, it promoted a positive influence with the increase of up to 45% in the presence of microorganisms in the samples ($p \leq 0.001$), mainly fungi. The low concentration of bacterial counts was negatively influenced by the low concentration of CO ($p = 0.003$); on the other hand, the fungi showed a significant positive influence in relation with the low concentrations of NO₂ ($p = 0.014$) (Table 7).

On the other hand, in the urban area, the concentrations of SO₂ in the analyzed data did not impact the concentration of microorganisms. High CO concentrations did not decrease bacterial concentrations, and even NO₂, with high levels in the air, continued to have a slight significant positive influence ($p = 0.006$) with CFU/m³ of fungi (Table 8).

For the Mann–Whitney *U* test, the variable of microorganisms was split into two groups (≤ 100 and ≥ 101 CFU/m³) for the analyses, and the results of their frequency are described in Table 9.

It was observed that NO₂ showed positive influences in relation with the fungal counts in both studied areas and had no correlation with the concentrations of bacteria in the analyzed samples.

Table 6 KMO–Bartlett’s test about the relationships between fungal and bacterial counts and the pollutants in the studied areas

Area	Kaiser–Meyer–Olkin (KMO) Test for Sampling Adequacy	Value	
		Fungal	Bacterial
Urban	Measure	0.710	0.724
	<i>Bartlett’s Test of Sphericity</i>		
	≈ Chi-squared	368.495	348.054
	df	6	6
	Sig.	≤0.001	≤0.001
Rural	Kaiser–Meyer–Olkin (KMO) Test for Sampling Adequacy	Fungal	Bacterial
	Measure	0.620	0.419
	<i>Bartlett’s Test of Sphericity</i>		
	≈ Chi-squared	9.470	24.172
	df	6	6
	Sig.	0.149	≤0.001

df: degrees of freedom

Sig.: *p* value

Table 7 Spearman’s rank correlation between the concentrations of total CFU/m³, fungal counts, and bacterial counts and NO₂, SO₂, and CO in the rural area

		NO ₂	SO ₂	CO	CFU/m ³	CFU/m ³ of fungi	CFU/m ³ of bacteria
NO ₂	Correlation	1.000	−0.014	−0.224	−0.248	0.336	0.072
	Sig.	–	0.947	0.462	0.073	0.014	0.621
SO ₂	Correlation	–	1.000	−0.375	0.675	−0.378	0.377
	Sig.	–	–	0.207	≤0.001*	0.057	0.076
CO	Correlation	–	–	1.000	−0.316	−0.116	−0.642
	Sig.	–	–	–	0.187	0.635	0.003

Sig.: *p* value

Table 8 Spearman’s rank correlation between the concentrations of total CFU/m³, fungal counts, and bacterial counts and NO₂, SO₂, and CO in the urban area

		NO ₂	SO ₂	CO	CFU/m ³	CFU/m ³ of fungi	CFU/m ³ of bacteria
NO ₂	Correlation	1.000	0.446	0.486	−0.002	0.204	0.086
	Sig.	–	≤0.001*	≤0.001*	0.978	0.006	0.266
SO ₂	Correlation	–	1.000	0.601	−0.053	0.036	−0.007
	Sig.	–	–	≤0.001*	0.493	0.636	0.929
CO	Correlation	–	–	1.000	−0.005*	−0.022	−0.069
	Sig.	–	–	–	0.942	0.773	0.367

Sig.: *p* value

Mann–Whitney *U* test (Table 10) verified statistical significance in the adequacy of data from these populations, for fungi (*p* = 0.004) and bacteria (*p* ≤ 0.001).

4 Discussion

The role of the atmosphere in the dispersion of fungi is quite nonlinear (Franić et al., 2023), in which each

Table 9 Frequency of microorganism groups (CFU/m³) for Mann–Whitney *U* test analysis

		CFU/m ³	Frequency	Percentile (%)	Valid percent (%)	Cumulative percent (%)
Fungi	Valid N	≤ 100	183	75	75	75
		≥ 101	61	25	25	100
		Total	244	100	100	–
			Frequency	Percentile (%)	Valid percent (%)	Cumulative percent (%)
Bacteria	Valid N	≤ 100	204	83.6	87.6	87.6
		≥ 101	29	11.9	12.4	100
		Total	233	95.5	100	–
	Missing values	System	11	4.5	–	–
		Total	244	100	–	–

Table 10 Mann–Whitney *U* test analysis between the microbial counts and air pollutants

	Parameters	N	Mann–Whitney <i>U</i> test					
			≤ 100 CFU/m ³		≥ 101 CFU/m ³		Total	
			Mean ± SD		Mean ± SD	Median	<i>p</i> value	
Fungi	SO ₂	199	18,47 ± 27,38		30,25 ± 55,69	19,15 ± 31,13	5,3	0,212
	NO ₂	230	46,31 ± 27,91		36,26 ± 46,79	44,76 ± 30,23	42,5	0,004
	CO	201	18,29 ± 30,21		25,60 ± 49,80	18,35 ± 32,35	2	0,788
		N	≤ 100 CFU/m ³	≥ 101 CFU/m ³	Total			
			Mean ± SD		Mean ± SD	Median	<i>p</i> value	
Bacteria	SO ₂	199	20,51 ± 32,58		13,55 ± 23,82	19,15 ± 31,13	5,3	0,460
	NO ₂	230	49,92 ± 29,30		28,34 ± 27,33	44,76 ± 30,23	42,5	≤ 0,001
	CO	201	19,91 ± 33,78		11,21 ± 23,89	18,35 ± 32,35	2	0,433

SD: Standard Deviation

N: Total

biological, pollution, and meteorological variable can have antagonistic or summation effects depending on the situation. What is known is that there are few investigations on microbial bioaerosols, among them, that they can be indicators of the level of biological pollution of the air and the attention that has been given to their relations as well. Since fungi and bacteria can be found as part of the microbial flora in the atmosphere, they deserve a more detailed analysis of their particles present and transported by air, as fungal spores provide a better understanding of these phenomena, and a more detailed survey of airborne particles is required (Grinn-Gofroń et al., 2011; Nowakowicz-Dębek et al., 2017).

Regarding the presence of pigments in fungal colonies, some species of dematiaceous fungi may take up to 21 days to develop (Sterflinger et al., 2012), creating a bias in the differentiation of hyaline and dematiaceous fungi in the seven-day period of growth.

Spore-forming units varied according to seasons, thus characterizing the seasonal number of fungi dispersed. Locality and seasons have already been described as influencing the aerial dispersion of the fungi most commonly isolated in the outdoor air of the city of São Paulo, and this influence was recorded during this study (Amend et al., 2010; Onofre., 2010; Borges, Monteiro, Monteiro, 2012; Filali Ben Sidel et al., 2015).

The relationships between fungal and bacterial communities are not fully understood yet. Since the environmental conditions of an area (temperature, humidity, physical and chemicals patterns) can allow the high concentration of fungi, it is more than likely that bacterial communities can proliferate in these environments, given their ease of assembly (Schmidt et al., 2014).

Previous studies have shown that regardless of the area analyzed, the greatest diversity and number of fungi occur during the winter season, which corroborates the finding of this research, where there was a significant increase in the number of fungal spores during the winter season (Dannemiller et al., 2016; Fan et al., 2019; Oliveira et al., 2009; Temperini et al., 2019). In other studies, conducted in the city of São Paulo, the highest concentrations of CO, NO₂, and SO₂ were recorded during the winter period, between the months of May and September, because of low rainfall rates, weak winds, and higher occurrence of temperature inversions, which corroborate the findings of this research (Aguar, 2015; Carvalho et al., 2015; Grinn-Gofroń et al., 2011). On the flip side, these same parameters are related to the increase in the number of fungal spores dispersed and may be related to the values of the relationships found between microorganisms and pollutants (Arbex et al., 2012; Dong et al., 2016; El-Batrawy, 2010; Mitchell et al., 2007).

Pollution concentrations have a strong impact on the diversity of genera and the number of microorganisms present in the air. Similarly, in this study, in the rural region where there is less pollution, the diversity of fungal genera is greater, and, as pollutant concentrations increase, there is a progressive decline of the diversity of microorganisms dispersed in the air, safeguarding the concentrations of bacteria that did not get affected by the presence of CO (Liu et al., 2019; Oliveira et al., 2009).

Aspergillus sp. showed as the most present fungi in the samples in all seasons and in both areas analyzed, corroborating other studies of the same area (Liu et al., 2019; Oliveira et al., 2009). Some species of this genus can cause opportunistic diseases, such as aspergillosis, especially in people with immunity issues (Cuervo-Maldonado et al., 2010); other species found, such as *Trichoderma* sp., *Penicillium* sp., and *Alternaria* sp., are much

less harmful to human health, even though they also can cause infections in immunosuppressed patients (Stathakis et al., 2015; Recio et al., 2019; 'BI17: Opportunistic fungal infection with *Alternaria* in the immunosuppressed', 2021), but these species are excellent indicators in the areas of pollution and biotechnology (Tiwari, Misra and Sangwan, 2013; Filali Ben Sidel et al., 2015; Morales-Oyervides et al., 2020).

The number of fungi and bacterial counts in the samples between urban and rural areas remained the same throughout the seasons; however, there was a big growth in the presence of fungal and bacterial spores during the winter and summer seasons, respectively. In addition, the genera diversity was lower in the urban area samples, where SO₂, NO₂, and CO concentrations were higher, revealing that the pollutants do not impact in the number of microorganisms dispersed, but at the variety of genera found (Liu et al., 2019; Oliveira et al., 2009).

The relation between low SO₂ concentrations and the number of microorganisms present in the air of the rural area could be explained by the action of this substance in the germination of spores (fungi), when high levels of this pollutant can lead to form H₂SO₄, which is toxic to fungi and bacteria. This action mechanism has been used in the agricultural sector, and its usefulness is now revealed and studied in the monitoring of outdoor air (Schoenleincrusius et al., 2001; Ana Paula Mendes Emygdio, Cristiane Degobbib, Fábio Luiz Teixeira Gonçalves, 2018).

In studies already published, NO₂ concentrations negatively impacted the concentration of fungi dispersed in the air (Schoenleincrusius et al., 2001), different from the slightly positive correlation found in the period studied. However, other studies reveal that the influence of this pollutant on the concentration of microorganisms depends on other parameters and changes temporally (Abdel Hameed et al., 2012; Gao et al., 2016).

The analysis of these pollutants, such as CO, has already revealed positive and negative influences according to the environmental conditions. As in this study, CO concentrations impacted the diversity and number of bacteria dispersed mainly in urban areas, where the concentration of this pollutant is higher (Dong et al., 2016; Liu et al., 2019).

5 Conclusions

During the collection of samples carried out in the urban region of São Paulo and in the rural region of Ibiúna for six years and in the four seasons, the highest concentration of fungi occurred in the winter season, and the concentration of bacteria did not vary during the study period. The greatest diversity of fungal genera was found in the rural area, where pollutant concentrations were lower.

Regarding the influence of pollutants NO₂, CO, and SO₂, dispersed in the air from the MRSP, the presence of NO₂ had a positive influence in the concentration of fungi in both studied areas. The presence of low CO concentrations had a negative influence on the concentration of bacteria, and the low concentration of the pollutant SO₂ impacted positively on the concentration of airborne microorganisms in the rural area.

The results indicated negative influences on the correlations between bacteria and CO concentration.

The monitoring of airborne fungi allows further studies to assist the analyses, determining bioindicators, based on their frequency and sensitivity to pollutants, which may be the most appropriate way to obtain parameters directly linked to environmental conditions for the benefit of human health.

This study has provided a possible baseline for further studies on analyzing the relationship between microorganisms and chemical components in outdoor air.

Authors' contribution Dulcilena de Matos Castro e Silva contributed to conceptualization, methodology, investigation, resources, data curation, writing—original draft preparation, writing—review and editing, and project administration; Fábio Luiz Teixeira Gonçalves contributed to conceptualization, supervision, and funding acquisition; Rosa Maria Nascimento Marcusso contributed to software and data curation; Maria Regina Alves Cardoso contributed to validation and writing—review and editing; and Valter Batista Duo Filho contributed to resources, data curation, and writing—review and editing. All authors have read and agreed to the published version of the manuscript.

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Conflict of interest Not applicable.

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