Evaluation of Airborne Microbial Contamination in Chemistry Faculty Laboratories at Universidad Autónoma de Nuevo León

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Resumen

La calidad del aire interior en los laboratorios es un factor crítico para garantizar la seguridad ocupacional y prevenir la contaminación microbiológica de experimentos químicos y biológicos. Sin embargo, la información disponible sobre la composición microbiana del aire en laboratorios académicos de México sigue siendo limitada. En este estudio, se recolectaron muestras de aire en dos laboratorios de la Facultad de Química de la Universidad Autónoma de Nuevo León, México, mediante un método de muestreo pasivo realizado dos veces al día (mañana y tarde). Los microorganismos fueron identificados a través de caracterización morfológica y bioquímica. La carga microbiana total varió de 286 a 2076 UFC/m³ para bacterias y de 174 a 1103 UFC/m³ para hongos. Los microorganismos aislados incluyeron *Rhizopus* spp., *Penicillium* spp., *Alternaria* spp., *Aureobasidium* spp., *Klebsiella* spp., *Salmonella* spp. y *Yersinia* spp., algunas de ellas clasificadas como potencialmente patógenas. La carga microbiana mostró una correlación significativa con el número de ocupantes, la humedad relativa y el periodo de muestreo, mientras que la temperatura no presentó un efecto medible. Estos hallazgos ponen de manifiesto la presencia de una amplia diversidad de microorganismos aerotransportados, incluidos posibles patógenos, en ambiente s de laboratorio, lo que resalta la necesidad de un monitoreo continuo de la calidad microbiológica del aire para apoyar la evaluación de riesgos y el desarrollo de estrategias que fortalezcan la bioseguridad en laboratorios académicos.

Palabras clave: Calidad de aire interior, Carga microbiana, Bioseguridad

Abstract

Indoor air quality in laboratories is a critical factor for ensuring occupational safety and preventing microbiological contamination of chemical and biological experiments. However, data on the microbial composition of laboratory air in academic settings in Mexico remain limited. In this study, air samples were collected from two laboratories at the Faculty of Chemistry, Universidad Autónoma de Nuevo León, Mexico, using a passive sampling method performed twice daily (morning and afternoon). Microorganisms were identified through morphological and biochemical characterization. The total microbial load ranged from 286 to 2076 CFU/m³ for bacteria and from 174 to 1103 CFU/m³ for fungi. Isolated microorganisms included *Rhizopus* spp., *Penicillium* spp., *Alternaria* spp., *Aureobasidium* spp., *Klebsiella* spp., *Salmonella* spp., and *Yersinia* spp., with some species classified as potentially pathogenic. Microbial load was significantly correlated with the number of occupants, relative humidity, and sampling period,



whereas temperature had no measurable effect. These findings highlight the presence of diverse airborne microorganisms, including potential pathogens, in laboratory environments, underscoring the need for continuous monitoring of microbial air quality to support risk assessment and the development of strategies that strengthen biosafety in academic laboratories.

Keywords: Indoor air quality, Microbial load, Biosafety

Introduction

People spend nearly 90% of their time indoors, typically inhaling 10-14 m3 of air per day in environments such as classrooms, homes, and offices [1-3]. Indoor air often contains pollutants originating from construction materials, organic waste, or domestic animals, which are generally classified as physical, chemical, or biological [4,5]. Among these, pollutants or bioaerosols-airborne particles of biological origin derived from plants, animals, or humans-represent 5-34% of total indoor contaminants [1,2,4]. Bioaerosols may include pathogenic microorganisms capable of causing respiratory and allergic diseases such as pneumonia, asthma, influenza, and hypersensitivity reactions. For this reason, the microbiological evaluation of indoor air has gained increasing relevance, not only to quantify contamination levels and identify microbial species, but also to understand the environmental and anthropogenic factors that influence their presence [1-8]

In recent years, numerous studies have focused on assessing the microbiological load in indoor environments such as laboratories, where studentsparticularly those engaged in chemical and biological sciences-spend a substantial amount of time. A wide diversity of microbial species has been detected in the air of both teaching and research laboratories. Among the bacteria most frequently identified are Staphylococcus spp. [5,7,8], Klebsiella spp. [6,8], and Escherichia coli [6], along with other genera such

as Bacillus, Pseudomonas, Micrococcus, Gemella, and Listeria [5-8]. Fungal species have also been commonly reported, particularly Penicillium spp. [5,8] and Aspergillus niger [5]. Given the potential pathogenicity of several of these microorganisms and their implications for both occupational health and experimental integrity, the systematic monitoring of laboratory air quality is essential to ensure biosafety and minimize risks in academic and research settings. Several studies have reported correlations between microbial load and environmental characteristics in laboratories. Among these, the number of occupants has consistently been identified as the most influential factor, whereas temperature and relative humidity do not always show a significant effect. Additional parameters, such as ventilation type and air exchange rates, can also strongly influence microbial concentrations [9,10]. To date, no official Mexican regulations have been established regarding microbiological quality of international Nevertheless. agencies, national standards, and independent researchers have proposed guideline values. For instance, the World Health Organization (WHO) recommends that bacterial concentrations in indoor air should not exceed 1000 CFU/m3 to be considered safe [11]. Other countries, however, enforce stricter limits, such as Sweden (700 CFU/m3) [3], Brazil (750 CFU/m3) [12], and Hong Kong (1000 CFU/m3) [13].

The present study aims to evaluate microbiological quality of indoor air in three laboratories of the Faculty of Chemistry at the Universidad Autonoma de Nuevo Leon, where students spend prolonged periods potentially exposed to bioaerosols. Specifically, the study seeks to identify the presence and frequency of enterobacteria and fungi, to assess correlations between microbial load and selected environmental and physical parameters, and to propose strategies to improve indoor air quality in laboratory settings.

Materials and Methods

Air samples were collected from two laboratories: the Physicochemical Processes Laboratory (PCP) and the Biotechnology Laboratory (BT). A passive air sampling technique, following standardized procedures [14], was employed. Petri dishes containing nutrient agar (for bacteria), and potato dextrose agar (for fungi) were used. Plates were exposed to laboratory air for 4 h. Sampling was carried out in the morning and afternoon on two different days, separated by one week. Bacterial samples were incubated at 37 °C for 24 h, while fungal samples were incubated at 37 °C for 3–5 days. Microbial load was expressed as CFU/m³ and calculated using Omeliansky's equation (Equation 1) [15,16].

$$N = 5a * 10^4 (Bt)^{-1}$$

Where:

- N = microbial load (CFU/m³)
- a = number of colonies counted on the Petri dish
- b = surface area of the Petri dish (cm²)
- t = exposure time (min)

Bacteria and fungi from indoor air samples were identified based on macroscopic morphological characteristics (colony description), microscopic features (Gram staining for bacteria and methylene blue staining for fungi), and, in the case of bacteria, biochemical assays including catalase, indole production, citrate utilization, SIM, TSI, and MRVP tests. For each microorganism identified, the frequency percentage was calculated [17–19]. Only bacteria belonging to the Enterobacteriaceae family were analyzed. To explore potential correlations between microbial load and specific environmental or laboratory variables, a principal component analysis (PCA) was performed using Infostat statistical software [20].

Results and Discussions

The microbial load, estimated by passive sampling, ranged between 286–2076 CFU/m³ for bacteria and 174–1103 CFU/m³ for fungi. The Biotechnology Laboratory showed both the minimum and maximum bacterial counts, as well as the maximum fungal load, whereas the minimum fungal load was recorded in the Physicochemical Processes Laboratory (Table 1).

Table 1. Microbial loads present in the laboratories.

	PCP Lab		BT Lab	
Sampling period	Bacteria	Fungi	Bacteria	Fungi
Morning	1417.8 (a)	306.7	571.0	192.9
Afternoon	700.2	237.3	308.6	347.2
Morning	1209.5 (a)	497.7	2075.6 (a)	1103.4 (a)
Afternoon	399.3	173.6	285.5	200.6

(a)

xceeding reference limit values

In the PCP Laboratory, bacterial load values exceeded the WHO recommended limit of 1000 CFU/m³ on two occasions. However, according to international standards—and considering the absence of specific regulations in Mexico—these values correspond to moderate contamination, representing minimal risk. The observed increase in bacterial load may be related to the higher number of occupants in the morning, since anthropogenic activity is one of the main sources of bioaerosols. In contrast, fungal load did not exceed the recommended limit at any time, indicating no risk associated with airborne fungi in this laboratory [5].

In the BT Laboratory, both bacterial and fungal loads exceeded the limit value on one occasion. This may be attributed to the greater number of occupants and the use of microorganisms during laboratory activities. Similar findings have been reported by other authors, who observed that increased occupancy combined with microbial handling contributes to higher microbial loads in indoor air [5–7].

Identification of microorganisms

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Based on morphological characteristics and biochemical tests, microorganisms were identified at the genus level. For fungi, the genera presumptively identified included *Rhizopus*, *Penicillium*, *Alternaria*, *Aureobasidium*, and yeast, with *Rhizopus* being the most frequently isolated genus in both laboratories (Table 2). Three fungal isolates could not be identified or assigned to any clinically relevant genus

Table 2. Frequency distribution of isolated fungal genera in each laboratory.

Genus	Isolations (a)	PCP Lab (a)	BT Lab (a)
Rhizopus	8 (47.05%)	3 (42.86%)	5 (50%)
Penicillium	3 (17.65%)	1 (14.28%)	2 (20%)
Alternaria	1 (5.88%)	1 (14.28%)	0 (0%)
Aureobasidium	1 (5.88%)	0 (0%)	1 (10%)
Yeast	1 (5.88%)	0 (0%)	1 (10%)
Unidentified	3 (17.65%)	2 (28.57%)	1 (10%)

(a)

umber of isolations (frequency percentage)

Penicillium is a well-known genus and one of the most commonly occurring fungi, widely distributed in habitats such as soil, vegetation, air, indoor environments, food products, and water-damaged building materials. Rhizopus, a representative genus of the phylum Zygomycota, comprises saprophytic fungi typically found in decaying organic matter, including plants and soil. Species of Alternaria are frequently isolated from soil and plants, as well as from cereal grains, wood, grass, and construction materials. In contrast, Aureobasidium is more commonly associated with indoor environments, as it requires high moisture levels for growth, thriving in damp or continuously wet areas such as bathrooms and kitchen surfaces [21-23]. These genera, together with yeasts, are among the fungi most frequently detected in indoor air, largely due to their constant presence outdoors, which constitutes a major source of bioaerosols [24]. Although these fungal genera are considered part of the natural indoor microbiota, they have been associated with recurrent human diseases such as allergies, opportunistic infections, and, in severe cases, mucormycosis. Therefore, caution is required when working in laboratories where they are present, particularly in the case of immunocompromised individuals [25].

Regarding bacteria, isolates were identified as belonging to the genera *Klebsiella*, *Salmonella*, and *Yersinia*, each detected only once and therefore presenting the same frequency (Table 3). The remaining samples could not be assigned to the family *Enterobacteriaceae*.

Table 3. Frequency distribution of isolated bacterial genera in each laboratory.

Genus	Isolations ^(a)	PCP Lab ^(a)	BT Lab (a)
Klebsiella	1 (3.57%)	1 (12.5%)	0 (0%)
Salmonella	1 (3.57%)	1 (12.5%)	0 (0%)
Yersinia	1 (3.57%)	0 (0%)	1 (5%)
Non-enteric	25 (89.29%)	6 (75%)	19 (95%)

umber of isolations (frequency percentage)

Bacteria of the genus Klebsiella are commonly found in nature as well as in humans, primarily in the digestive tract, oral cavity, and feces. Their presence in sites such as the mouth facilitates their release into the environment, contributing to indoor air contamination. Infections caused by Klebsiella generally do not occur in healthy individuals but are more frequent in patients undergoing treatment for other illnesses. Similarly, bacteria of the genus Salmonella inhabit the human intestine and can cause severe disease in immunocompromised individuals, most commonly through the consumption of contaminated food

In contrast, Yersinia is not typically found in healthy individuals. In infected hosts, it colonizes the intestines and can be shed through feces. Transmission usually occurs via direct contact with infected people or animals. The disease it causes (yersiniosis) is generally mild and can be effectively treated with standard antibiotic therapy [26]. As with fungi, the bacterial genera identified are considered common (except for Yersinia); however, all can cause disease. Therefore, appropriate precautions should be observed when working in the laboratories.

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Principal component analysis

A principal component analysis (PCA) was conducted to evaluate whether any of the three environmental variables studied (temperature, relative humidity, and number of occupants) significantly influenced the microbial load in the laboratories. The PCA results are presented in Figure 1.

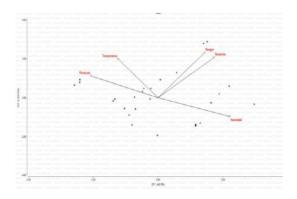


Figure 1. Principal component analysis graph.

From the graph, three distinct phenomena were identified: Firstly, a strong correlation was observed between bacterial and fungal components, as evidenced by the proximity and alignment of their vectors in the biplot. This suggests that as one increases, so does the other. This finding is consistent with the fact that microorganisms present as bioaerosols often originate from common sources—whether natural (e.g., transported by wind through diffusion mechanisms) or anthropogenic.

Secondly, a moderate correlation was found between microbial presence and relative humidity. This was inferred from the orientation of the humidity vector, which, although not perfectly aligned with the microbial vectors, was sufficiently close to indicate a degree of association. This relationship can also be explained by diffusion processes, as bioaerosols released into the air can attach to suspended water droplets, using them as a medium for transport into indoor environments.

Finally, neither temperature nor the number of occupants showed a significant influence on microbial load. The lack of temperature influence may be due to the narrow temperature range during data collection, as all laboratory environments remained close ambient temperature (approximately 25 °C), with minimal fluctuation. However, the absence of a detectable effect from human presence contradicts findings from prior studies, which identify humans as major contributors to indoor bioaerosol levels. This inconsistency may be attributed to the limited dataset used in the principal component analysis (PCA), considering that such analyses typically require larger sample sizes for robust conclusions. Similar findings regarding the limited influence of temperature and the moderate correlation with humidity have been reported in previous studies [10].

Conclusions

The average bacterial and fungal loads recorded in the laboratories during the sampling periods ranged from 286 to 2076 CFU/m³ and 174 to 1103 CFU/m³, respectively. The highest microbial load was observed in the Biotechnology Laboratory; however, values did not exceed thresholds typically associated with highly contaminated environments. Nevertheless, both laboratories surpassed the recommended microbial load limits (≤500 CFU/m³ for indoor environments, according to WHO and other international guidelines) during certain sampling periods, indicating episodes of potential air quality compromise.

Four fungal genera were identified among the isolates.

Rhizopus was the most prevalent (47%), followed by Penicillium (17%), and Alternaria, Aureobasidium, and a yeast species, each with 5%. The bacterial isolates belonged to the genera Klebsiella, Salmonella, and Yersinia, each with a frequency of 33%. Except for Yersinia, the identified taxa are frequently reported as part of the typical indoor air microbiota. Nonetheless, all identified organisms are opportunistic pathogens with potential health risks,

particularly in sensitive environments such as research laboratories.

Among the environmental factors studied, a strong correlation was observed between bacterial and fungal loads, with both also moderately associated with relative humidity. In contrast, temperature and occupant density did not exhibit statistically significant effects on microbial concentrations. These findings highlight the role of humidity in facilitating microbial dispersion and survival in indoor environments.

From a practical standpoint, the results underscore the need for continuous microbiological monitoring and humidity control in laboratory settings to maintain biosafety standards and minimize potential exposure risks for personnel. Further studies with larger datasets are recommended to validate these findings and to explore additional influencing factors under varying operational conditions.

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