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RESEARCH ARTICLE

MYCOBIOTA ISOLATED FROM THE AIR, DUST AND THE DOCUMENTS SURFACE PRESERVED IN REPOSITORIES OF THE NATIONAL ARCHIVE OF CUBA

MICOBIOTA AISLADA DEL AIRE, EL POLVO Y LA SUPERFICIE DE DOCUMENTOS PRESERVADOS EN REPOSITORIOS DEL ARCHIVO NACIONAL DE CUBA

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ABSTRACT

This study aimed to determine the concentration and diversity of fungal species in the air, dust and documents surface kept in two repositories of the National Archive of the Republic of Cuba (ARNAC) characterized by having two different circulation systems of air. The outdoor and indoor air of two repositories (Repository-13 [R-13] with natural cross ventilation and Map library [ML] with air conditioning) were sampled with a biocollector at two different times corresponding to the two seasons of the year; the dust was passively sampled and the surfaces of 11 documents preserved in these repositories were sampled. The maximum concentrations of fungi were obtained in the season of little rain and the indoor/outdoor ratios in all cases were ≤ 1.0, demonstrating the good environmental quality of the repositories. The average dust loads collected in ML and R-13 were 11.7 and 31.6 mg/m^2/day, respectively. From dust, the predominant genera were *Cladosporium*, *Penicillium* and *Aspergillus*, but *Aureobasidium*, *Botrytis*, *Drechslera*, *Paecilomyces*, *Pithomyces* and *Trichoderma* were new records for the ARNAC´s dust; likewise, a high diversity of species was obtained. Although the documents analyzed were apparently clean, fungi were isolated in concentrations that ranged between 5 x 10 and 3.5 x 10^2 CFU/cm^2. The species *A. flavus*, *A. glaucus*, *A. niger*, *P. janthinellum* and *P.raistrickii* were common in all the documents analyzed. The species similarity (Sørensen similarity coefficient, QS) among the analyzed niches (indoor air, outdoor air, collected dust, document surface) showed that the QS_{TOTAL} obtained by comparing the species of the indoor air of the repositories with those of the dust collected, was high. The species diversity profile followed the order: settled dust > airborne > document surface.

Keywords: fungal diversity; archives; air mycobiota; dust mycobiota; mycobiota on documents surface; indoor environments.

RESUMEN

Este estudio tuvo como objetivo determinar la concentración y diversidad de especies fúngicas en el aire, el polvo y la superficie de los documentos que se conservan en dos repositorios del Archivo Nacional de la República de Cuba (ARNAC) caracterizados por tener dos sistemas diferentes de circulación de aire. El aire exterior e interior de dos repositorios (Repositorio-13 [R-13] con ventilación natural cruzada y Mapoteca [ML] con aire acondicionado) se muestrearon con biocolector en dos momentos diferentes correspondientes a las dos estaciones del año, se colectó polvo de forma pasiva y se muestreó la superficie de 11 documentos conservados en los mismos repositorios donde el aire fue analizados. Las concentraciones máximas de hongos se obtuvieron en la época de pocas lluvias y las relaciones interiores/exteriores en todos los casos fueron ≤ 1.0, demostrando la buena calidad ambiental de los repositorios. Las cargas medias de polvo recogidas en ML y R-13 fueron de 11.7 y 31.6 mg/m^2/día, respectivamente. Del polvo los géneros predominantes fueron *Cladosporium*, *Penicillium* y *Aspergillus*, pero *Aureobasidium*, *Botrytis*, *Drechslera*, *Paecilomyces*, *Pithomyces* y *Trichoderma* son nuevos registros para el polvo del ARNAC; asimismo, se obtuvo una alta diversidad de especies. A pesar de que los documentos analizados estaban aparentemente limpios, se aislaron hongos en concentraciones que oscilaron entre 5 x 10 y 3.5 x 10^2 UFC/cm^2. Las especies *A. flavus*, *A. glaucus*, *A. niger*, *P.janthinellum* y *P. raistrickii* fueron comunes en todos los documentos analizados. La similitud de especies (coeficiente de similitud de Sørensen, QS) entre los nichos analizados (aire interior, aire exterior, polvo recolectado, superficie del documento) evidenció que el QS_{TOTAL} obtenido al comparar las especies del aire interior de los repositorios con las del polvo colectado, fue elevado. El perfil de diversidad de especies siguió el orden: polvo depositado > en el aire > superficie del documento.

Palabras clave: diversidad fúngica; archivo; micobiota aérea; micobiota del polvo; micobiota en la superficie de documentos; ambientes interiores.

INTRODUCTION

The archives are the institutions in charge of conserving the Historical Memory of nations, which constitutes an important part of the legacy of humanity. The documentary wealth that they treasure has been generated by natural or legal persons in the exercise of their functions or intellectual activity. This documentation is found on various materials (papyrus, parchment, paper, metal, plastic, etc.), and includes types and formats different among which are special documents such as photographs, maps, microforms, audiovisual media, films, digital documents, etc. These materials age and deteriorate over time, but the deterioration process is accelerated by the effect of physical agents (light, temperature, relative humidity) and chemicals (atmospheric pollution), particulate matter (dust, soot) as well as biological agents (microorganisms, insects) present in the environment (Borrego, 2012).

Temperature (T) and relative humidity (RH) are among the environmental parameters that are most monitored and analyzed in archives, libraries, and museums, since they are involved in the chemical processes that occur during the aging of substrates, inks, and pigments. The same occurs with light intensity and particularly UV radiation (Cappitelli et al., 2009). However, the incidence of the settleable dust and biological agents, particularly filamentous fungi, on the collections kept by these institutions is not always monitored with the same rigor and systematicity.

The fungi existence in the indoor environment of the archives is due to their introduction from the outdoor through the air and together with the dust that penetrates through windows and doors, ventilation or air conditioning systems in poor hygienic conditions, through cracks and other openings in the walls, etc. (Hassan et al., 2021; Camargo et al., 2022). They can also be introduced into buildings through dirt dragged by footwear and attached to people's clothing and skin (Schneider, 2003; Karbowska-Berent et al., 2011) and even by insects and mites (Savoldelli et al., 2021). It is important to point out that the concentration of fungal propagules can increase inside the archives as a result of their dispersal from internal sources of contamination (Karbowska-Berent et al., 2011) such as documents, other objects and infected surfaces that are moved or handled. Dust also constitutes a contributing element of fungal contamination to the indoor environment of a premises (Schneider, 2003) and particularly in archives, libraries and museums (Skóra and Gutarowska, 2016).

The dust contains numerous elements that come from natural and anthropogenic sources, of biotic origin (mites, pollens and fungal propagules) and abiotic (chemical substances from industries and automobiles, as well as different sugars that can serve as a carbon source for fungi) (Oliva et al., 2001; Morawska and Salthammer, 2003). Therefore, it is composed of inorganic and organic substances and water (Morawska and Salthammer, 2003; Nastasi et al., 2020; Viegas et al., 2020) and is considered an important source of information in microbiological diagnostic studies in an environment indoor since it constitutes a reservoir of microbial contamination and especially of fungi from the air, so it can constitute a reference for the study of the behavior of the aerial mycobiota of an entire season (Pasquarella et al., 2012). Furthermore, it turns out to be a secondary source of environmental microbial contamination if it is agitated and re-suspended (Schneider, 2003; Skóra and Gutarowska, 2016; Shan et al., 2019). In this way, dust constitutes a potential danger for documents of heritage value (Grau-Bové et al., 2016) and human health (Nastasi et al., 2020; Salin et al., 2021).

Filament fungi are among the microorganisms that can be found in the indoor air of archives, libraries, and museums, as well as in the dust settled on materials and documents (Viegas et al., 2020). These agents use these materials as nutrients for their growth and development, causing serious effects on their chemical composition and altering their physical-mechanical structure, as well as causing aesthetic damage that affects their appearance and, in many cases, the correct reading of their message (Pinzari and Gutarowska, 2021). In addition to damaging the documentary heritage preserved in the archives, fungi can severely affect human health since they have different structures and pathogenic mechanisms that cause specific affections for human (Köhler et al., 2018; Segura-Medina et al., 2019), which is why some are considered primary pathogens, such as *Stachybotrys chartarum* and *Aspergillus fumigatus* and others, opportunistic or secondary pathogens that would only affect the human organism if it presents immunological problems (O'Gorman and Fuller, 2008; Yamamoto et al., 2012).

According some experimental results obtained in Cuba on the diversity and physiology of environmental fungi isolated in various archives (Rodríguez & Borrego, et al., 2023; Borrego and Molina, 2020; Borrego et al., 2020; Borrego et al., 2021a; Borrego et al., 2022a, b, c), and also from allergological studies made (Álvarez et al., 2020; Herrera et al., 2021), it is known with certainty that the mycological quality of the environment in the archives plays an important role in the staff health. These environments are characterized by having, at times, more or less high loads of dust and fungi that can trigger allergies and other health effects (Hay, 2020) since personnel are exposed to these allergens during the working day (Herrera et al., 2021). However, having palliative and preventive actions is vital to maintain the sustainability of the work in the archives at present, even more so when

more severe health affectations are envisioned, mainly due to climate change, which will cause emerging infections and storms of asthma caused by fungi (Reinmuth-Selzle et al., 2017; D'Amato et al., 2020).
For a correct evaluation of the environmental quality in the archives, it is essential to determine not only the concentration and diversity of fungi in the air, but also in dust and on surfaces, including the documents surfaces that constitute the three ecological niches to be studied (Pinzari, 2011). Taking these aspects into account, this study aimed to determine the concentration and diversity of fungal species in the air, dust and on the documents surface that are kept in two repositories of the National Archive of the Republic of Cuba (NARC) characterized for having two different air circulation systems.

MATERIALS AND METHODS

Characteristics of the studied repositories

Research was conducted at two NARC repositories located on the south side of the first floor of the building. The Map library (ML) is a large repository (length x width x height, m = 15.2 x 6.2 x 5) that is air conditioning all year round, so it has average values of T = 22°C ± 2°C and RH = 58.5% ± 2%. Repository 13 (R-13) has natural cross ventilation and has the same dimensions as ML. This repository has 8 windows in total, 4 on each side; it also has 64 ventilation ducts in total, 16 of them located on both sides of the premises at a height of 1 m and 16 at a height of 4.5 m. These ventilation conducts are holes that cross over the outer walls in an angle of 45°, and they are protected by metallic meshes. Through years of studies of the thermohygrometric values in the repository, it is known that it has average values of T = 30.3°C ± 2.8°C and HR = 75.5% ± 6.8%.

Mycological air sampling

Two samplings were carried out in 2019; the first was in March (month corresponding to the little rains season) and the second was done in July (month belonging to the rainy season).
Sampling was performed at five points in each repository in triplicate (Sanchis-Solera, 2002) using a SAS Super 100TM biocollector (VWR International Srl, Italy) with an air flow of 100 l/min for 1 min at a height of 1.5 m from the floor and at intervals of 1 hour to guarantee the recovery of the air in the analyzed point. Petri dishes of 90 cm in diameter were used with Malt Extract Agar (MEA) (Biocen, Cuba) supplemented with sodium chloride (7.5%) (MEA + NaCl) and MEA at pH = 5 (Borrego et al., 2022c). Subsequently, the dishes were incubated inverted at 30°C for 5 to 7 days and the colonies were counted to determine the fungal concentration expressed in colony-forming units per cubic meter of air (CFU/m3) according to the instructions described in the equipment manual (SAS Super 100™, 2001).
The indoor/outdoor (I/O) ratio was calculated according to De Aquino Neto and Goés Siqueira (2000) who indicated that if this relationship is equal to or less than 1.5 it is that the environments are not contaminated and there is good ventilation, if the relationship is between 1.5 and 2, then the environmental quality is regular and if it is greater than 2 it is because the environments are polluted and have poor ventilation.

Sampling and mycological characterization of the collected dust

Three years before the first determination of airborne fungi, dust collectors were located at the same five points used in the air sampling. As these collectors were placed over the shelves, they were at a height of 3 m from the floor in R-13 and approximately 2 m in ML. These collectors consisted of sterile and previously weighed 110 mm plastic Petri dishes that were placed at the sampling points and left open for three years. Thus, the dust settled on the surface of the dishes in the same way that it does on the documents (Borrego et al., 2022a). As the collectors from the different points were collected, their respective lids were placed on them and closed, and they were transported to the laboratory. Subsequently, they were placed in desiccators with silica gel and were weighed every 24 h until they reached constant weight.
The determination of the total sedimented dust load was carried out according to the formula proposed by Oliva et al. (2001):
Total dust load = (Ps – Pi) / A x t
Where: Ps- final dry weight of the Petri dish with dust, Pi- initial weight of the sterile Petri dish, A- area of the dish (m^2), t- time (days)
To determine the dust accumulated in the ventilation ducts of R-13 located at a height of 1 m, these ducts were not cleaned for three years. In the sampling, a vacuum cleaner was used to vacuum all the dust accumulated in the ducts and a dust pool was made.
To isolate the mycobiota from the dust, 0.001 g of dust were taken from each collector and 0.01 g of the dust pool collected form ducts, respectively. One mL of sterile saline was added to each sample and shaken well at random

intervals for 45 minutes. Serial dilutions were then made which were seeded deep into 110 mm dishes with MEA medium + NaCl and MEA pH = 5. The dishes were incubated inverted for 5 to 7 days at 30°C. Once the incubation was completed, the colony count was performed to determine the fungal concentration expressed in colony-forming units per g of collected dust (CFU/g).
CFU/g = (Number of total CFU obtained by dilution) x 1 g of dust / 0.001 g of dust collected or 0.01 g of dust pool

Mycological sampling of the surface of documents

The sampled documents were randomly selected. From ML, five maps were chosen that were extracted from their drawers to carry out this analysis and from R-13, six notarial protocols (NP) located very close to the air and dust sampling points were studied. The documents were apparently clean; however, they were carefully examined with the help of magnifying glass (X30) and no fungal growth was observed on them.
The sample was taken from an area of 4.84 cm^2 using the aseptic swab technique (Borrego et al., 2022c). Each swab was dipped into a tube containing 2 mL of sterile saline, the sample was shaken at random intervals for 45 minutes, and serial dilutions were made and seeded deep into 110 mm dishes using the culture media MEA + NaCl and MEA at pH = 5. The dishes were incubated inverted for 5 to 7 days at 30°C. Once the incubation was completed, the colony count was performed and the fungal concentration was reported as CFU/cm^2.

Fungal identification

Fungal identification was performed by considering the macroscopic characteristics by analyzing the front and back of the fungal colonies with a stereo microscope (X14). The conidiophores and conidia structures were observed using preparations in lactophenol blue in a clear field trinocular microscope (Olympus, Japan) at X40 and X100 coupled to a digital camera (Samsung, Korea). In the genera identification different mycological key manuals were used (Domsch et al., 1980; Barnett and Hunter, 2003). For the identification of species of *Aspergillus*, *Penicillium* and *Cladosporium* genera, criteria from other authors were used (Ellis, 1976; Klich &Pitt, 1994; Pitt, 2000; Klich, 2002; Varga et al., 2011a, b; Bensch et al., 2010, 2012; Samson et al., 2011; Visagie et al., 2014). The web site of MycoBank and Index Fungorum were also consulted.

Ecological approaches

Relative frequency (RF) of the fungal species detected in the three niches studied was determined according Esquivel et al. (2003) where:
RF = (times a species is detected / total number of sampling realized) x 100
The ecological categories are classified as: Abundant (A) with RF = 100-81%, Common (C) with RF = 80-61%, Frequent (F) with RF = 60-41%, Occasional (O) with RF = 40-21%, Rare (R) with RF = 20-0% (Borrego and Perdomo, 2016).
The Sørensen's coefficient of similarity (QS) was used to compare the similarities of obtained taxa among the three ecological niches (indoor air, outdoor air, and collected dust). The comparisons made were between the indoor air and the outdoor as well as between the indoor air and the collected dust (Sánchez et al., 2019; Borrego et al., 2022a):
QS = 2a/b + c
Where: a- is the number of common genera detected in the two environments that are comparing, b- the number of detected taxa only in indoor environment and c- the number of detected taxa only in the outdoor environment.
The values of QS are given in the range from 0 - 1. A value equal to 0 indicates that the taxa obtained in both compared niches are completely different and a value equal to 1 indicates that the taxa are identical (Moreno, 2001).

Statistical analysis

In the statistical processing of data Statgraphics Centurion XV program was used. Student's t test was used to compare the differences in the fungal concentrations obtained by both sampling times and to compare the differences between the averages of collected dust in ML and R-13. A simple variance analysis (ANOVA-1) and Duncan test were performed to compare the obtained data related with the fungal concentrations from the dust. A p-value smaller or equal to 0.05 was considered statistically significant.

RESULTS

Concentration and diversity of airborne fungi in the analyzed repositories

The fungal concentrations obtained in the analyzed repositories varied depending on the sampling performed (Fig. 1). In the 1st sampling carried out in March, the month corresponding to the dry season, the concentrations obtained in ML and R-13 were 420 and 770 CFU/m³ respectively, while the fungal concentration outdoor was 740 CFU/m³. This led to I/O ratios of 0.6 for ML and 1.0 for R-13.

In the 2nd sampling carried out in July, the month corresponding to the heavy rainy season, the concentrations obtained in ML and R-13 were 330 and 460 CFU/m³ respectively; however, the concentration in outdoor was 480 CFU/m³. As can be seen, the values were significantly higher in the 1st sampling both in the air inside both repositories and outside. The I/O ratios obtained were 0.7 for ML and 1.0 for R-13, which demonstrates the good environmental quality of these repositories regardless of the season of the year.

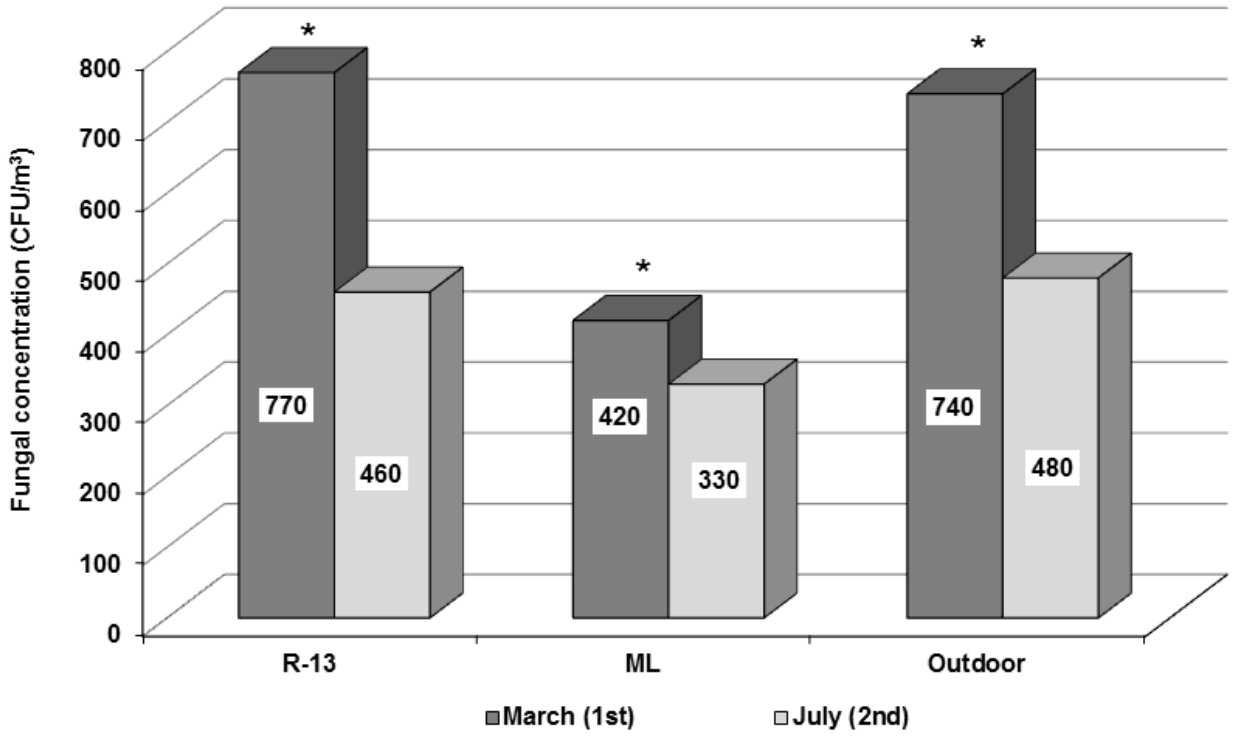

Fig. 1. *Fungal concentrations obtained in the indoor and outdoor air of the two repositories studied. *: Indicates significant differences when comparing the data obtained between month according to Student's test ($p \leq 0.05$).*

From the indoor environments, 19 genera of filamentous fungi and 3 genera of yeasts were isolated, as well as two non-sporulating mycelia, one white and the other pigmented (Fig. 2). As can be seen, the fungi of the phylum Ascomycota predominated. Of the fungi, *Aspergillus* and *Cladosporium* were the predominant genera since they were detected in the two repositories in the two samplings made, therefore they were classified as abundant. However, *Chrysosporium* and *Candida* turned out to be common genera as well as the two non-sporulating mycelia. Other genera such were classified as rare genera.

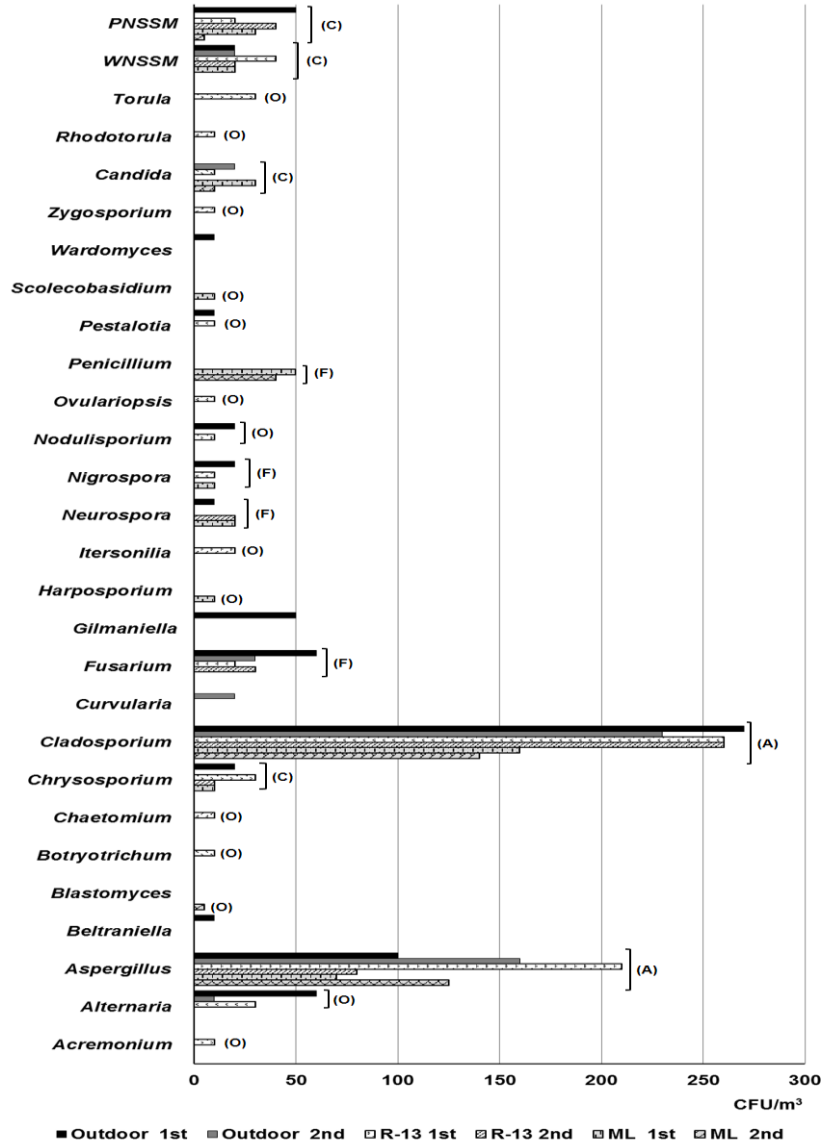

Fig. 2. *Concentrations and ecological behavior of the genera and non-sporulating mycelia detected in the indoor air of the Map library (ML), of R-13 and of the outdoor air in each sampling made. Ecological categories (EC) are Abundant taxa (A) when RF = 100-81%, Common taxa (C) when RF = 80-61%, Frequent taxa (F) when RF = 60-41%, Occasional taxa (O) when RF = 40-21% and Rare taxa (R) when RF = 20-0.1%. WNSSM: white non-sporulating septate mycelium. PNSSM: pigmented non-sporulating septate mycelium. The EC only correspond to the species detected in the indoor air of the repositories, i.e. outdoor air was not taken into account.*

Aspergillus and *Cladosporium* genera were also predominant in the outdoor air, since they were detected in both samples at concentrations between 100 and 270 CFU/m³. Other genera isolated from abroad in both samplings but at concentrations ≤ 60 CFU/m³ were *Alternaria* and *Fusarium*, as well as a non-sporulating white mycelium (WNSSM). Ten genera coincided in both the outdoor air and indoor air of the repositories and were *Alternaria*, *Aspergillus*, *Chrysosporium*, *Cladosporium*, *Fusarium*, *Neurospora*, *Nigrospora*, *Nodulisporium*, *Pestalotia*, and *Candida*. Species diversity was high in particular for *Aspergillus* and *Cladosporium*genera. Of a total of 53 species detected in the indoor air of the repositories, 15 were *Aspergillus* (28.3%), 12 *Cladosporium* (22.6%), 3 *Penicillium* (5.7%), and 2 *Alternaria* and *Fusarium* respectively (3.8%). Of the rest of the genera, only one species was isolated. From the 1st sampling carried out in ML, 21 species were isolated with a marked predominance of those belonging to the genus *Cladosporium* (11 species for 52.4%) and only 3 species corresponded to the genus *Aspergillus* (14.3%), while

one species was isolated from each of the following genera: *Chrysosporium*, *Harposporium*, *Neurospora*, *Nigrospora*, *Penicillium*, *Scolecobasidium* and *Candida* (Table 1).

On the other hand, a total of 28 species were isolated from R-13, distributed six from *Cladosporium* (21.4%), five from *Aspergillus* (17.9%), two from *Chrysosporium* and *Fusarium* respectively (7.1%), and one species from the genera *Acremonium*, *Alternaria*, *Botryotrichum*, *Chaetomium*, *Itersonilia*, *Nigrospora*, *Nodulisporium*, *Ovulariopsis*, *Pestalotia*, *Zygosporium*, *Candida*, *Rhodotorula* and *Torula*.

Table 1. Diversity and concentration (CFU/m³) of the species isolated from the air of the repositories analyzed and from outdoor

	ML				R-13				Outdoor			
	Samplings										RF [a]	
Taxa	1st	I/O	2nd	I/O	1st	I/O	2nd	I/O	1st	2nd	(%)	EC [a]
Acremonium sp. Link **	0	0	0	0	10	-	0	0	0	0	25.0	O
Alternaria cinerea (Baucom& Creamer) Woudenb. &Crous	0	0	0	0	0	0	10	-	0	0	25.0	O
Alternaria triticina Prasada&Prabhu	0	0	0	0	30	0.5	0	0	60	10	25.0	O
Aspergillus aculeatus Iizuka *	0	0	0	0	0	0	10	-	0	0	25.0	O
A. auricomus (Guég.) Saito	0	0	10	-	0	0	0	0	0	0	25.0	O
A. athecius Raper& Fennell	10	-	0	0	0	0	0	0	0	0	25.0	O
A. conicus Blochwitz *	0	0	0	0	10	-	0	0	0	0	25.0	O
A. flavus Link * [c]	45	0.9	15	0.4	60	1.2	20	0.5	50	40	100	A
A. glaucus (L.) Link	0	0	10	1.0	40	2.0	0	0	20	10	50.0	F
A. japonicus Saito *	0	0	10	0.5	0	0	10	0.5	0	20	50.0	F
A. niger Tiegh.*	10	0.3	15	0.5	50	1.7	10	0.3	30	30	100	A
A. oryzae (Ahlb.) Cohn *	0	0	20	-	0	0	0	0	0	0	25.0	O
A. restrictus G. Sm. *	0	0	15	-	50	-	0	0	0	0	50.0	F
A. sydowii (Bainier&Sartory) Thom & Church*	0	0	20	-	0	0	0	0	0	0	25.0	O
A. tamarii Kita *	0	0	0	0	0	0	10	0.5	0	20	25.0	O
A. terreus Thom *	0	0	0	0	0	0	10	0.5	0	20	25.0	O
A. ustus (Bainier) Thom & Church *	0	0	10	-	0	0	0	0	0	0	25.0	O
A. wentii Wehmer *	0	0	0	0	0	0	10	0.5	0	20	25.0	O
Beltraniella pini M.B. Ellis	0	0	0	0	0	0	0	0	10	0	0	-
Blastomyces sp. Gilchrist & W.R. Stokes **	0	0	5	-	0	0	0	0	10	0	25.0	O
Botryotrichum sp. Sacc. &Marchal	0	0	0	0	10	-	0	0	0	20	25.0	O
Chaetomium globosum Kunze *	0	0	0	0	10	-	0	0	0	0	25.0	O
Chrysosporium sp. Corda **	10	1.0	0	0	0	0	10	-	10	0	50.0	F
Chrysosporium merdarium (Ehrenb.) J.W. Carmich. *	0	0	0	0	20	2.0	0	0	10	10	25.0	O

Chrysosporium tropicum J.W. Carmich. *	0	0	0	0	10	1.0	0	0	10	0	25.0	O
Cladosporium caryigenum (Ellis &Langl.) Gottwald	20	1.0	0	0	20	1.0	0	0	20	0	50.0	F
C. cladosporioides (Fresen.) G.A. de Vries *	40	0.4	50	0.6	30	0.3	200	2.2	110	90	100	A
C. coralloides W. Yamam.	10	1.0	0	0	0	0	0	0	10	0	25.0	O
C. gossypiicola Pidopl. &Deniak	10	-	0	0	0	0	0	0	0	0	25.0	O
C. herbarum (Pers.) Link *	20	0.5	0	0	0	0	50	1.0	40	50	50.0	F
C. lignicola Corda	10	1.0	0	0	0	0	0	0	10	0	25.0	O
C. minourae Iwatsu	10	1.0	0	0	50	-	0	0	10	0	50.0	F
C. oxysporum Berk. & M.A. Curtis *	10	1.0	25	0.8	40	1.3	10	0.3	10	30	100	A
C. sphaerospermum Penz. *	20	0.5	35	1.8	0	0	0	0	40	20	50.0	F
C. staurophorum (W.B. Kendr.) M.B. Ellis	20	-	30	-	0	0	0	0	0	0	50.0	F
C. subuliforme Bensch, Crous& U. Braun *	0	0	0	0	40	2.0	0	0	20	0	25.0	O
C. tenuissimum Cooke, Grevillea *	20	-	0	0	80	-	0	0	0	0	50.0	F
Curvularia spicifera (Bainier) Boedijn	0	0	0	0	0	0	0	0	0	20	0	-
Fusarium graminearum Schwabe[b]	0	0	0	0	10	0.3	0	0	30	0	25.0	O
Fusarium xylarioides Steyaert	0	0	0	0	10	0.5	30	1.0	20	30	50.0	F
Gilmaniella sp. G.L. Barron	0	0	0	0	0	0	0	0	20	0	0	-
Harposporium sp. Lohde	10	-	0	0	0	0	0	0	0	0	25.0	O
Itersonilia sp. Derx	0	0	0	0	20	1.0	0	0	20	0	25.0	O
Neurospora crassa Shear & B.O. Dodge	20	2.0	0	0	0	0	20	-	10	0	50.0	F
Nigrospora sphaerica (Sacc.) E.W. Mason *	10	0.5	0	0	10	0.5	0	0	20	0	50.0	F
Nodulisporium sp. Preuss	0	0	0	0	10	0.5	0	0	20	0	25.0	O
Ovulariopsis sp. Pat. andHar.	0	0	0	0	10	-	0	0	0	0	25.0	O
Penicillium aurantiogriseum Dierckx * [b]	0	0	30	-	0	0	0	0	0	0	25.0	O
P. chrysogenum Thom * [b]	0	0	10	-	0	0	0	0	0	0	25.0	O
P. citrinum Thom * [b]	35	-	0	0	0	0	0	0	0	0	25.0	O
Pestalotia sp. De Not.	0	0	0	0	10	1.0	0	0	10	0	25.0	O
Scolecobasidium sp. E.V. Abbott	10	-	0	0	0	0	0	0	0	0	25.0	O
Wardomyces inflatus (Marchal) Hennebert	0	0	0	0	0	0	0	0	10	0	0	-
Zygosporium sp. Mont.	0	0	0	0	10	0.3	0	0	30	0	25.0	O

Candida sp. Berkhout **	20	2.0	10	0.5	10	1.0	0	0	10	20	75.0	C
Rhodotorula sp. F.C. Harrison **	0	0	5	-	10	-	0	0	0	0	50.0	F
Torula sp. Pers.	0	0	0	0	30	-	0	0	0	10	25.0	O

*I/O: Indoor/Outdoor ratio. *: Pathogenic species (de Hoog et al., 2000; Lysková, 2007; Sandoval-Denis et al., 2015). **: Indicative that several species of the genus are pathogenic (de Hoog et al., 2000; Guarro, 2012). [a]: Specifies that these data only correspond to the species detected in the indoor air of the repositories, i.e. outdoor air was not taken into account. [b]: It refers to species producer of mycotoxins harmful to humans (Frisvad and Samson, 2004; Zinedine et al., 2007).[c]: Indicative of species with biohazard II according to Karbowska-Berent et al. (2018). RF: Relative frequency. EC: Ecological categories are Abundant taxa (A) when RF =100-81%, Common taxa (C) when RF = 80-61%, Frequent taxa (F) when RF = 60-41%, Occasional taxa (O) when RF = 40-21% and Rare taxa (R) when RF = 20-0.1%.*

From the 2nd sampling made in ML, a total of 18 species were isolated with a marked predominance of those belonging to the *Aspergillus* genus (nine species, 50%), while were isolated four species of *Cladosporium* (22.2%), two of *Penicillium* (11.1%) and only one species was isolated from the genera *Blastomyces*, *Candida* and *Rhodotorula*. Of them, only *C. cladosporioides* was isolated at a concentration of 50 CFU/m^3, but with a low I/O ratio (0.6), revealing that its presence is related to the existing concentration outdoor (90 CFU/m^3) or perhaps to dust. From R-13, 13 species were isolated, a lower quantity than those found in the 1st sampling; of them, those of *Cladosporium* (six species, 46.2%) predominated, followed by *Aspergillus* (four species, 30.8%) and by *Chrysosporium* and *Fusarium* with two species each (15.4%). However, in this case two species were isolated at concentrations ≥ 50 CFU/m^3 (*C. cladosporioides*, *C. herbarum*), but only *C. cladosporioides* showed an I/O ratio = 2.2, while *C. herbarum* revealed an I/O ratio = 1.0. This indicates that only the species *C. cladosporioides* constitutes a contaminant of this repository or entered the indoor air together with the dust.

Concentration and diversity of dustborne fungi in the environment of the analyzed repositories

The average loads of dust collected in ML and R-13 were 11.7 and 31.6 mg/m^2/days respectively (Table 2). The load values obtained in ML was significantly lower than that obtained in R-13. This is an expected result since ML is a repository that, because it is air-conditioned, is kept closed and this means that the entry of dust is less.
The average fungal concentration obtained from the collected dust was 1.9 x 10^3 CFU/g in ML and 2.0 x 10^2 CFU/g in R-13; this last value turns out to be significantly lower than the one obtained in ML. However, the dust accumulated in the R-13 ventilation ducts showed a higher fungal concentration (8.0 x 10^5 CFU/g). An estimate of the daily fungal accumulation in the settled dust for three years in ML gives an average of 20 CFU/day and in R-13 it is 2 CFU/day, while the average fungal accumulation in the ventilation ducts of R-13 is 7 x 10^2 CFU/day. The differences between both repositories can be given by the environmental characteristics of each one.

Table 2. Total load of settleable dust collected during one year in Map library (ML) and R-13 as well as from the R-13 ventilation ducts

Repository	Points sampled	Total load of collected dust (mg/m²/days)	Fungal concentrations (CFU/g)
ML	1	19.2	3.1×10^3
	2	16.1	3.3×10^3
	3	8.6	1.7×10^3
	4	10.3	1.3×10^3
	5	6.5	1.1×10^3
	6	9.7	1.0×10^3
Average		**11.7 ***	**1.9×10^3 ***
R-13	1	36.2	3.0×10^2
	2	41.1	4.0×10^2
	3	29.6	1.0×10^2
	4	30.3	2.0×10^2
	5	24.5	1.0×10^2
	6	27.7	2.0×10^2
Average		**31.6**	**2.0×10^2**
Ventilation ducts of R-13		-	**8.0×10^5 ****

**: Indicates significant difference according to Student's t test ($P \leq 0.05$) when the averages of collected dust and fungal concentrations obtained in ML and R-13 were compared. **: Refers significant difference according to Duncan test ($P \leq 0.05$) when this value was compared with the values of fungal concentrations obtained in ML and R-13.*

From the dust collected in ML, seven genera and two non-sporulating mycelia were isolated, with a predominance of *Cladosporium* (218 CFU/g), followed by *Penicillium* (98 CFU/g) and *Aspergillus* (67 CFU/g), while in R-13, 18 genera and two non-sporulating mycelia were found, but in this case, although *Cladosporium* continued to predominate (54 CFU/g), the concentration was lower and was followed by *Aspergillus* (49 CFU/g) and *Penicillium* (26 CFU/g) (Fig. 3). On the other hand, from the dust accumulated in the ventilation ducts of R-13, a total of 26 genera and two non-sporulating mycelia were detected. From the dust aspirated from the ventilation ducts, a marked predominance of the genus *Aspergillus* (193 CFU/g) was obtained, followed by *Cladosporium* (164 CFU/g) and *Penicillium* (118 CFU/g). *Drechslera*, *Fusarium* and *Trichoderma*genera were obtained at concentrations between 20 and 25 CFU/g, while others were obtained at lower concentrations.

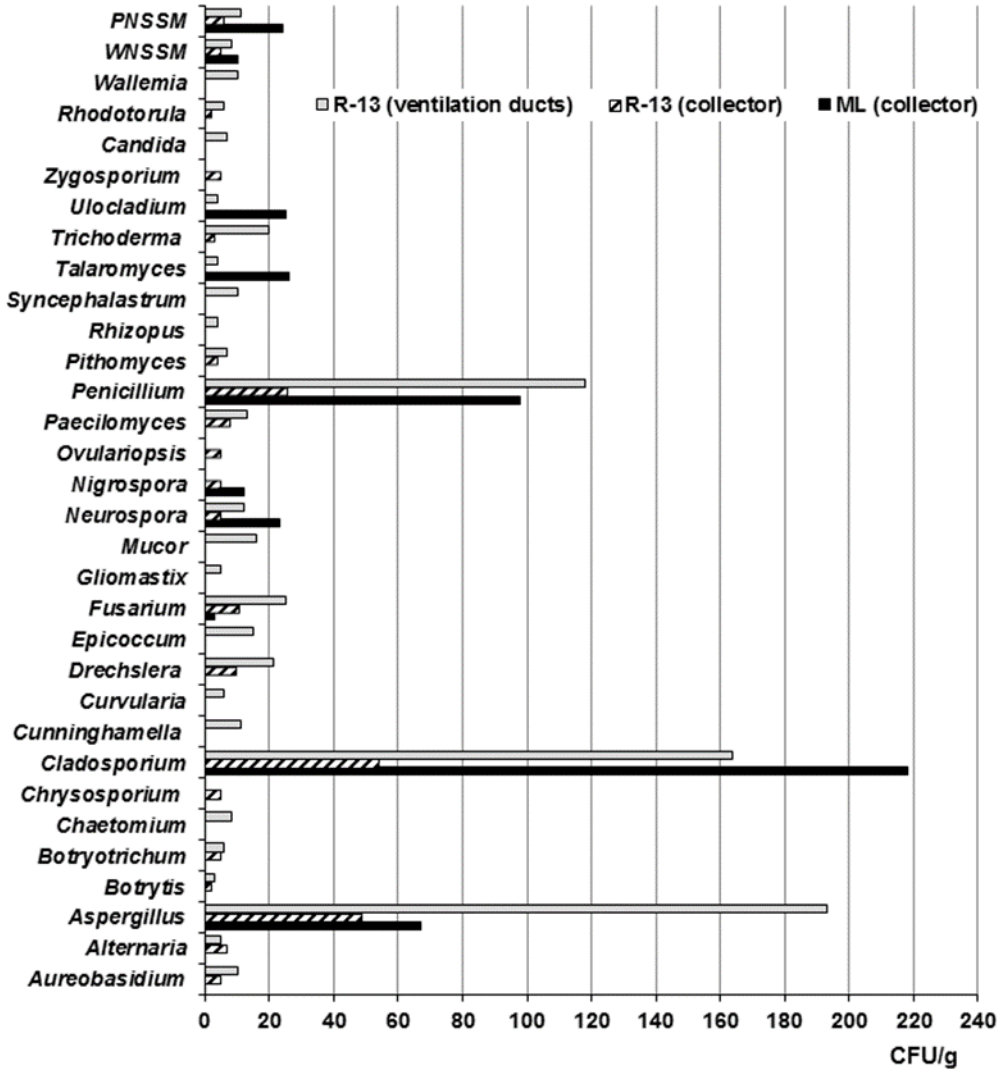

Fig. 3. *Concentration of the genera detected in the collected dust. WNSSM: white non-sporulating septate mycelium. PNSSM: pigmented non-sporulating septate mycelium*

A total of 69 species and two non-sporulating mycelia were isolated from the dust (Table 3). Of them, 21 were detected only in the dust accumulated in the R-13 ventilation ducts, which is why they are ecologically classified as occasional, 15 species were isolated from the dust collected in both repositories and from the dust accumulated in the ventilation ducts of the R-13, classified as abundant (RF = 100%), eight species were only detected in the dust collected in R-13 and seven in the dust collected in ML.

The predominant species in their entirety corresponded to the genera *Aspergillus* (12 species), *Cladosporium* and *Penicillium* (11 species of each one); but nine species of *Aspergillus*, ten of *Cladosporium* and nine of *Penicillium* were detected in the dust collected indoor the repositories. They were followed by *Fusarium* (2), *Mucor* (2), *Paecilomyces* (2), *Talaromyces* (2) and *Trichoderma* (2) species. Among the species detected in the dust, it can be seen that nine were also found in the air of the repositories in some of the samplings carried out, and they were *A. flavus*, *A. glaucus, A. niger*, *A. restrictus*, *C. caryigenum*, *C. cladosporioides*, *C. herbarum*, *N. crassa* and *N. sphaerica*. Other species were found in air and dust from R-13 (*A. aculeatus*, *Botryotrichum* sp., *Chrysosporium merdarium*, *C. subuliforme*, *Ovulariopsis* sp., *Zygosporium* sp., and *Rhodotorula* sp.), while some species were detected in air and dust from ML (*A. athecius*, *C. coralloides*, *C. gossypiicola*, *C. lignicola*, *C. sphaerospermum*, *C. staurophorum*, *P.aurantiogriseum*, *P. chrysogenum* and *P. citrinum*). These results show the interrelation of taxa between the air and the dust that circulates in the indoor environments analyzed.

Table 3. Concentration of the taxa detected in the collected dust (CFU/g) in the analyzed repositories and in the R-13 ventilation ducts, as well as their ecological behavior

Taxa	ML (collector)	R-13 (collector)	Ventilation ducts of R-13	RF (%)	EC
Alternaria alternata (Fr.) Keissl.	0	7	5	66.7	C
Aspergillus aculeatus Iizuka	0	7	0	33.3	O
A. athecius Raper& Fennell	5	3	8	100	A
A. clavatus Desm.	0	0	12	33.3	O
A. flavus Link	30	9	30	100	A
A. glaucus (L.) Link	5	5	15	100	A
A. nidulans (Eidam) G. Winter	0	0	15	33.3	O
A. niger Tiegh.	10	10	45	100	A
A. ochraceus G. Wilh.	0	0	12	33.3	O
A. penicillioides Speg.	5	0	10	66.7	C
A. restrictus G. Sm.	5	8	35	100	A
A. sydowii (Bainier&Sartory) Thom & Church	0	5	0	33.3	O
A. versicolor (Vuill.) Tirab.	7	2	11	100	A
Botrytis sp. P. Micheli ex Haller	0	0	3	33.3	O
Botrytis cinerea Pers.	0	2	0	33.3	O
Botryotrichum sp. Sacc. &Marchal	0	5	6	66.7	C
Chaetomium globosum Kunze	0	0	8	33.3	O
Chrysosporium merdarium (Ehrenb.) J.W. Carmich.	0	5	0	33.3	O
Cladosporium basiinflatum Bensch, Crous& U. Braun	25	0	20	66.7	C
C. caryigenum (Ellis &Langl.) Gottwald	20	5	0	66.7	C
C. cladosporioides Link	80	20	80	100	A
C. coralloides W. Yamam.	12	2	0	66.7	C
C. gossypiicola Pidopl. &Deniak	9	1	0	66.7	C
C. herbarum (Pers.) Link	15	10	25	100	A

C. lignicola Corda	10	2	5	100	A
C. oxysporum Berk. & M.A. Curtis	0	0	12	33.3	O
C. sphaerospermum Penz.	22	10	22	100	A
C. staurophorum (W.B. Kendr.) M.B. Ellis	25	2	0	66.7	C
C. subuliforme Bensch, Crous& U. Braun	0	2	0	33.3	O
Cunninghamella sp. Matr.	0	0	11	33.3	O
Curvularia lunata (Wakker) Boedijn	0	0	6	33.3	O
Drechslera sp. S. Ito	0	10	21	66.7	C
Epicoccum sp. Link	0	0	15	33.3	O
Fusarium graminearum Schwabe	3	8	12	100	A
Fusarium solani (Mart.) Sacc.	0	3	13	66.7	C
Gliomastix sp. Guég.	0	0	5	33.3	O
Mucor mucedo L.	0	0	9	33.3	O
Mucor racemosus Fresen.	0	0	7	33.3	O
Neurospora crassa Shear & B.O. Dodge	23	5	12	100	A
Nigrospora sphaerica (Sacc.) E.W. Mason	12	5	0	66.7	C
Ovulariopsis sp. Pat. andHar.	0	5	0	33.3	O
Paecilomyces lilacinus (Thom) Samson	0	0	10	33.3	O
Paecilomyces variotii Bainier	0	8	3	66.7	C
Penicillium aurantiogriseum Dierckx	30	3	12	100	A
P. chrysogenum Thom	5	10	32	100	A
P. citreonigrum Dierckx	10	3	0	66.7	C
P. citrinum Thom	35	5	20	100	A
P. corylophilum Dierckx	0	0	9	33.3	O
P. decumbens Thom	0	2	10	66.7	C
P. glabrum (Wehmer) Westling	0	0	4	33.3	O
P. janczewskii K.W. Zaleski	8	0	11	33.3	O
P. purpurogenum Stoll	0	2	7	66.7	C
P. spinulosum Thom	0	1	0	33.3	O
P. waksmanii K.W. Zaleski	10	0	13	66.7	C

Pithomyces chartarum (Berk. & M.A. Curtis) M.B. Ellis	0	4	7	66.7	C
Rhizopus stolonifer (Ehrenb.) Vuill.	0	0	4	33.3	O
Syncephalastrum racemosum Cohn ex J. Schröt.	0	0	10	33.3	O
Talaromyces flavus (Klöcker) Stolk& Samson	12	0	4	66.7	C
Talaromyces helicus C.R. Benj.	14	0	0	33.3	O
Trichoderma sp. Pers.	0	3	9	66.7	C
Trichoderma viride Pers.	0	0	8	33.3	O
Trichoderma lignorum (Tode) Harz	0	0	3	33.3	O
Ulocladium sp. Preuss	25	0	4	66.7	C
Zygosporium sp. Mont.	0	5	0	33.3	O
Aureobasidium sp. Viala& G. Boyer	0	5	10	66.7	C
Candida sp. Berkhout	0	0	7	33.3	O
Rhodotorula sp.	0	2	6	66.7	C
Wallemia sp. Johan-Olsen	0	0	10	33.3	O
WNSSM	10	5	8	100	A
PNSSM	24	6	11	100	A

RF: Relative frequency. EC: Ecological categories are Abundant taxa (A) when RF = 100-81%, Common taxa (C) when RF = 80-61%, Frequent taxa (F) when RF = 60-41%, Occasional taxa (O) when RF = 40-21% and Rare taxa (R) when RF = 20-0.1%. WNSSM: white non-sporulating septate mycelium. PNSSM: pigmented non-sporulating septate mycelium.

Fungal concentration and diversity on analyzed document surfaces

Of the 11 documents analyzed, fungal concentrations ranging from 5 x 10 to 3.5 x 10^2 CFU/cm^2 were isolated, evidencing the presence of viable fungi on the documents surfaces, although no fungal growth was observed with the naked eye (Table 4). From the different maps, a total of 14 species and one non-sporulating mycelium were isolated with a marked predominance of *Aspergillus* species (7) followed by *Penicillium* (4), while from the NP a total of 19 species and one non-sporulating mycelium were isolated with a prevalence of *Aspergillus* species (11) followed by those of *Penicillium* (3). Six similarspecies were detected in both types of documents (*A. flavus*, *A. glaucus*, *A. niger*, *A. penicillioides*, *P. janthinellum*, *P. raistrickii*).

Table 4. Fungal concentrations detected on documents surface and isolated taxa

Document type	Support and technique	CFU/cm^2	Taxa
Maps	Blueprint in paper	8.0 x 10	*A. flavus*, *A. glaucus*, *A. niger*, *A. penicillioides*, *A. sydowii*, *A. ustus*, *A. versicolor*, *Candida* sp., *P. citrinum*, *P. janczewskii*, *P. janthinellum*, *P. raistrickii*, *Talaromyces helicus*, *Trichophyton* sp., WNSSM
	Transparent paper	5.0 x 10	
	Paper on canvas	3.5 x 10^2	
	Linen	1.2 x 10^2	
	Linen with surface sizing	3.0 x 10^2	
Notarial Protocols (NP)	Paper 1	2.0 x 10^2	*A. candidus*, *A. chevalieri*, *A. flavus*, *A. glaucus*, *A. nidulans*, *A. niger*, *A. ornatus*, *A. parasiticus*, *A. penicillioides*, *A. restrictus*, *C. cladosporioides*, *C. sphaerospermum*, *Gliomastix* sp., *Paecilomyces variotii*, *P. aurantiogriseum*, *P. janthinellum*, *P. raistrickii*, *Torula* sp., *Zygosporium* sp., WNSSM
	Paper 2	1.5 x 10^2	
	Paper 3	2.3 x 10^2	
	Paper 4	1.0 x 10^2	
	Paper 5	1.2 x 10^2	
	Paper 6	2.5 x 10^2	

WNSSM: white non-sporulating septate mycelium

Common taxa detected in the indoor air (IA) of the repositories, the dust collected (CD) and on the surfaces of analyzed documents (DS)

Table 5 shows the results obtained by comparing the isolated species in three of the niches studied (IA, CD, DS). As can be seen, some species were detected in the three niches analyzed; others were isolated from the air and dust, while others were found in the dust and on the documents analyzed. Only three species turned out to be ecologically abundant (*A. flavus*, *A. glaucus*, *A. niger*), since they were detected in the indoor air of the two repositories analyzed, in the dust collected in both repositories, and on the surface of the documents preserved in these repositories, which shows that they are common among these niches and that they may be contributing to cross-contamination between air, dust, and documents. In the dust, the concentrations of most of these three species were low, but since there are a large number of their propagules sedimented on the documents, their dispersion into the air and their inhalation during their handling can be facilitated.

The species *C. caryigenum*, *C. herbarum*, *Neurospora crassa* and *Nigrospora sphaerica* were isolated from the air and the dust of the two repositories studied, so they were ecologically classified as common species, while *C. coralloides*, *C. gossypiicola*, *C. lignicola*, *C.staurophorum* were found in the air of ML as well as in the air and in the dust of R-13, which means which are common species. From the other species classified as common, *C.sphaerospermum*, *P. aurantiogriseum* and *P. citrinum* stand out because regardless of having been detected in the air of ML, they were also isolated from the air of R-13, from the dust collected in this repository and from the surface of maps and NP. Of the other species classified as frequent, it was obtained that *A. penicillioides* and *A. versicolor* were detected both in the dust collected in ML and on the maps surface. On the other hand, *C.coralloides*, *C. gossypiicola*, *C. lignicola*, *C. staurophorum* and *P. chrysogenum* were isolated from the air of ML and the dust collected in ML and R-13, while *Fusarium graminearum* was detected in the air of R-13 and in the dust of ML and R-13. *Zygosporium* sp. was found in the air of R-13, in the dust collected in R-13 and on notarial protocols, instead *A. sydowii* and *Candida* sp. were isolated both from the air of ML and from maps.

Table 5. Common taxa detected in the air indoor the repositories, the dust settled in the collectors and/or on the analyzed documents surface

	Indoor air		Dust collected					
	ML	R-13	ML	R-13	Documents			
Specie	(CFU/m^3)		(CFU/g)		Maps	NP	RF (%)	EC
Aspergillus aculeatus * [c]	-	10	-	7	-	-	33.3	O
A. flavus * [c]	45 - 15 [b]	60 – 20 [b]	30	9	+	+	100	A
A. glaucus *	10	40	5	5	+	+	100	A
A. nidulans * [c]	-	-	-	- [a]	-	+	16.7	R
A. niger * [c]	10 – 15 [b]	50 – 10 [b]	10	10	+	+	100	A
A. penicillioides *	-	-	5	- [a]	+	+	50.0	F
A. restrictus * [c]	15	50	5	8	-	+	83.3	A
A. sydowii * [c]	20	-	-	5	+	-	50.0	F
A. ustus * [c]	10	-	-	-	+	-	33.3	O
A. versicolor * [c]	-	-	7	2	+	-	50.0	F
Botryotrichum sp.	-	10	-	5	-	-	33.3	O
Chaetomium globosum *	-	10	-	- [a]	-	-	16.7	R
Chrysosporium merdarium	-	20	-	5	-	-	33.3	O
Cladosporium caryigenum	20	20	20	5	-	-	66.7	C
C. cladosporioides * [c]	40 – 50 [b]	30 – 200 [b]	80	20	-	+	83.3	A
C. coralloides	10	-	12	2	-	-	50.0	F
C. gossypiicola	10	-	9	1	-	-	50.0	F
C. herbarum *	20	50	15	10	-	-	66.7	C
C. lignicola	10	-	10	2	-	-	50.0	F
C. oxysporum *	10 - 25	40 - 10	-	- [a]	-	-	33.3	O
C. sphaerospermum * [c]	20 – 35 [b]	-	25	10	-	+	66.7	C
C. staurophorum	20 – 30 [b]	-	25	2	-	-	50.0	F
C. subuliforme	-	40	-	2	-	-	33.3	O
Fusarium graminearum [c]	-	10	3	8	-	-	50.0	F
Neurospora crassa	20	20	23	5	-	-	66.7	C
Nigrospora sphaerica *	10	10	12	5	-	-	66.7	C
Ovulariopsis sp.	-	10	-	5	-	-	33.3	O

Paecilomyces variotii *	-	-	-	8	-	+	33.3	O
Penicillium aurantiogriseum * [c]	30	-	30	3	-	+	66.7	C
P. chrysogenum * [c]	10	-	5	10	-	-	50.0	F
P. citrinum * [c]	35	-	35	5	+	-	66.7	C
P. janczewskii [c]	-	-	8	- [a]	+	-	33.3	O
Talaromyces helicus	-	-	14	-	+	-	33.3	O
Candida sp.	20 – 10 [b]	10	-	- [a]	+	-	50.0	F
Rhodotorula sp.	5	10	-	2	-	-	50.0	F
Torula sp.	-	30	-	-	-	+	33.3	O
Zygosporium sp.	-	10	-	5	-	+	50.0	F
WNSSM	20	40 – 20 [b]	10	5	+	+	100	A
PNSSM	30 – 5 [b]	20 – 40 [b]	24	6	-	-	66.7	C

*NP: Indicates Notarial Protocols. [a]: Indicativeof species detected in the dust accumulated in the ventilation ducts of R-13. [b]: Refers the values detected in the two samplings. *: Refers pathogenic species (de Hoog et al., 2000). [c]: Indicates species producer mycotoxins harmful to humans (Frisvad and Samson, 2004; Zinedine et al., 2007; Navale et al., 2021; Al Hallak et al., 2023). RF: Relative frequency. EC: Ecological categories are Abundant taxa (A) when RF = 100-81%, Common taxa (C) when RF = 80-61%, Frequent taxa (F) when RF = 60-41%, Occasional taxa (O) when RF = 40-21% and Rare taxa (R) when RF = 20-0.1%. WNSSM: white non-sporulating septate mycelium. PNSSM: pigmented non-sporulating septate mycelium.*

The analysis of species similarity (QS) among the studied niches showed that the QS_{TOTAL} obtained by comparing the species of the indoor air (IA) of the repositories with those of the dust collected (CD) in them, was high (0.6 - 0.8),the rest of the QS_{TOTALS} provided medium to low values ≤ 0.6 (Table 6). Similarity of *Aspergillus* species was high ($QS_{Asp.}$= 0.7 - 0.9) when comparing indoor (IA) to outdoor air (OA) and the same occurred for *Cladosporium* species ($QS_{Clad.}$ = 0.8 - 1.0). The similarity of *Aspergillus* and *Cladosporium* species obtained in indoor air (IA) with respect to the collected dust (CD) provided medium to high values depending on the sampling analyzed ($QS_{Asp.}$ = 0.5 - 0.6, $QS_{Clad.}$ = 0.8 - 0.5), while the QS obtained for *Penicillium* species was low ($QS_{Pen.}$ = 0.1 - 0.4). The other species comparisons made between indoor air (IA) and document surface (DS) as well as between document surface (DS) and collected dust (CD) showed low QS (0.1 – 0.4), only for *Aspergillus* species the DS-CD comparison showed a medium value ($QS_{Asp.}$ = 0.6).

Table 6. Sørensen's coefficient of similarity (QS) obtained by comparing the species isolated in the three studied niches

Relationship of taxa or species between:	QS_{TOTAL}	$QS_{Asp.}$	$QS_{Clad.}$	$QS_{Pen.}$
IA-OA: Indoor air and outdoor air	0.6 §	0.7 – 0.9 §§	0.8 – 1.0 §§	0 *
IA-CD: Indoor air and collected dust	0.6 – 0.8 §§	0.5 – 0.6 §§	0.8 - 0.5 §§	0.1 – 0.4 §§
IA-DS: Indoor air and document surface	0.2	0.3	0.3	0.3
DS-CD: Document surface and collected dust	0.4	0.6	0.2	0.1

*QS_{TOTAL}: It refers to the QS of all the species isolated in all the niches studied. $QS_{Asp.}$:Refers the QS of the Aspergillus species. $QS_{Clad.}$: Indicates the QS of the Cladosporium species. $QS_{Pen.}$: Refers the QS of the Penicillium species. §: Indicates similar values in both samplings. §§: Refers to variations according the sampling. *: Indicates that only the Penicillium species were detected on indoor air in the Ist sampling but they were not isolated from outdoor air.*

DISCUSSION

In indoor environments, it is proposed that a high diversity of species coincides with a high concentration of spores and fungal propagules transported through the air and deposited on materials. A dust-rich indoor area offers greater diversity than a clean one, even more than a constantly humid environment. In an indoor ecosystem fluctuating temperature and relative humidity can promote the presence of a large number of fungal species (Pinzari, 2011) and this is mainly because the availability of water is variable, and that the occurrence of species is limited by their dispersal strategies. The biological characteristics of dust depend largely on the external aerobiological situation, the seasonality of the release of spores by some fungi of the soil and/or associated with crops, and the atmospheric conditions (Mendrela-Kuder, 2003). Additionally, substrates can support different fungal communities, depending on their specific water activity, the carbon-to-nitrogen ratio, and the presence of small nutrient molecules such as sugars and peptides, which can aid in the reactivation of a spore. Therefore, air, dust and the surface of the object can be considered microbial niches (Pinzari, 2011) that must be studied to know really the quality of an indoor environment (Viegas et al., 2020).

About the concentrations of airborne fungi obtained in this work the values were similar to those reported in previous studies (Borrego & Perdomo, 2016; Borrego et al., 2022a), evidencing that the fungal load in the indoor air of the NARC repositories does not exceed those 1000 CFU/m³ of air and that the I/O relations obtained in this study (0.6 y 1.0) are indicative of good quality environments and with adequate air circulation indoor (De Aquino & Goés Siqueira, 2000; Awad et al., 2020), preventing the existence of areas of fungal amplification (Pinzari, 2011).

The differences obtained in the fungal concentrations between the two sampling may be due to the influence exerted by the intense rains during the washing of the atmosphere (Sabariego et al., 2004) which favored the decrease of the fungal concentration outdoors. As it is known, the outdoor environment influences indoor environments (Hassan et al., 2021; Camargo et al., 2022), therefore, as a consequence of the low concentrations of fungal propagules detected in July (month included in the rainy season of the year) outdoors, low concentrations were also obtained in the indoor air of the repositories regardless of whether they have air conditioning or natural cross ventilation. A similar behavior had already been detected in previous studies carried out at the NARC and other Cuban archives (Borrego &Perdomo, 2016; Borrego et al., 2020; Borrego et al., 2022a, b).

In a previous study carried out in R-13 (Borrego &Perdomo, 2012), although the outdoor fungal concentration was not reported, values of fungal load lower than or similar to those obtained in this study were reported, which ranged between 120 CFU/m^3 in February (month included in the little rainy season) and 300 CFU/m^3 in September (month that is part of the rainy season). These results show the good environmental quality that this repository has maintained over time. Regarding ML, in previous results the concentrations obtained were 50 CFU/m^3 (Borrego et al., 2017), 40.8 CFU/m^3 with an I/O ratio of 0.2 (Borrego &Molina, 2020) and 43.6 CFU/m^3 with an I/O ratio of 0.1 (Borrego et al., 2022a), which also indicates the good quality of the air from this repository despite the passage of time.

Most of the detected fungal genera corresponded to the phylum Ascomycota, which was to be expected since it is the phylum that predominates in the outdoor air of countries with a tropical climate (Fröhlich-Nowoisky et al., 2016) and in the indoor environment (Li et al., 2022). The predominance of *Aspergillus* and *Cladosporium* genera, together with *Penicillium* on some occasions, has been widely reported for Cuban archive environments (Borrego &Molina, 2020; Borrego et al., 2020; 2021; 2022a, b, c; Borrego, 2023). The other isolated genera were previously detected in NARC repository environments too (Borrego &Molina, 2018; Borrego &Molina, 2020; Borrego et al., 2022a; Borrego, 2023) and many of them were also found in air from other Cuban archives (Borrego et al., 2022b, c). Even the genera *Alternaria*, *Candida*, *Fusarium*, *Nigrospora*, *Nodulisporium*, *Zygosporium*, and *Rhodotorula* were isolated from the indoor air of libraries and museums located in Havana (Rojas et al., 2012; Borrego &Molina, 2019). Likewise, the genera *Alternaria*, *Fusarium*, *Nigrospora* and *Zygosporium* were found in the environment of a naturally ventilated museum located in Artemisa, one of the western provinces of the country and adjacent to Havana (Cruz et al., 2021).

The fact that *Aspergillus* and *Cladosporium* genera had been predominates in archive and library environments has been previously reported for other countries too (Pinheiro et al., 2019; Pirry et al., 2020; Hassan et al., 2021; Camargo et al., 2022), since these genera are considered part of the indoor mycobiota (Li et al., 2022). However, the high concentrations of *Aspergillus* and *Penicillium* genera are attributable to the small size of their spores that allow them to remain in the air for long periods of time (Richardson & Rautemaa-Richardson, 2021) and also, they are very abundant genera in the atmosphere of Havana (Almaguer et al., 2021), while *Cladosporium* is the most abundant genera in the outdoor environment (Anees-Hill et al., 2022; Camargo et al., 2022) and coincidentally very abundant also in the Havana atmosphere (Almaguer et al., 2014; Sánchez et al., 2019); perhaps for this reason they also abounded in the indoor air of the repositories analyzed. Although *Chaetomium* was detected years ago in a sampling performed in a NARC repository with natural ventilation (Borrego, 2019), it is a taxon that has not been isolated again in any Cuban archive. Even in R-13, this genus had never been isolated before, since the aforementioned report (Borrego, 2019) refers to R-14, a repository located next to R-13. Despite this, *Chaetomium* has previously been isolated from the air in museums and libraries in the capital (Rojas et al., 2012) and it is a genus that has been detected in the atmosphere of Havana in low proportion (Almaguer et al., 2014; Sánchez et al., 2019). As it is a hydrophilic fungus (Andersen et al., 2021) and the NARC repositories, where R-13 is included, do not have such a high relative humidity (RH ≥ 90%) that it can develop easily due to the natural-cross ventilation they have, it is possible that for this reason it is not frequently detected. However, it has been suggested that when the temperature is high and the relative humidity exceeds 65%, this genus can be detected (Almaguer et al., 2020) and since the annual average RH of this repository was 75.5%, perhaps for this reason it can be detected on rare occasions. Other genera such as *Alternaria*, *Fusarium*, *Nigrospora*, *Nodulisporium*, *Torula* and *Zygosporium* have also been detected in the atmosphere of Havana (Almaguer et al., 2014; Almaguer et al., 2017; Sánchez et al., 2019). Of the total of 10 coinciding genera in both environments, 9 (90%) were previously detected outdoors, evidencing that their presence indoors was due to the incidence of outdoor air, an aspect that agrees with what has been stated by several authors (Hassan et al., 2021; Camargo et al., 2022).

Despite having obtained a high species diversity of *Aspergillus* and *Cladosporium* genera, fundamentally, the concentrations were maintained below 30 CFU/m^3, according to recommended by Guild &MacDonald (2004) and Karbowska-Berent et al. (2011), and most with I/O ratios ≤1.0, indicating that these species are not indoor air pollutants in these repositories particularly of ML; only the species *Neurospora crassa* and *Candida* sp., despite having shown low concentrations (20 CFU/m^3), revealed I/O ratios = 2, which shows that these species are pollutants of the indoor air of the repository or reached the indoor air together with the dust, since it has been reported that the transport of dust particles from outdoor in tropical countries has a high content of fungal spores (Hassan et al., 2021; Savoldelli et al., 2021; Camargo et al., 2022).

Although the diversity of detected species was high, the most species were found at low concentrations (≤ 40 CFU/m^3). *Aspergillus flavus*, *A. niger*, *A. restrictus*, *C. minourae* and *C.tenuissimum* were found at impermissible

concentrations, i.e. ≥ 50 CFU/m^3 (Guild and MacDonald, 2004; Karbowska-Berent et al., 2011). Likewise, some species, despite having been found at low concentrations, revealed I/O ratios ≥ 1.5; in this case, the species *A. glaucus*, *A.niger*, *Chrysosporium merdarium* and *C. subuliforme* were found, which shows that they may be indoor air pollutants of R-13 or that they are linked to the dust that enters the repository.

Although members of the genera *Blastomyces*, *Candida*, *Chrysosporium*, *Cladosporium*,*Nigrospora*, and *Rhodotorula* have formerly been isolated from the air from NARC repositories (Borrego and Molina, 2018; Borrego and Molina, 2020; Borrego et al., 2022a), some of their species are known to be opportunistic pathogens (de Hoog et al., 2000; Guarro, 2012; Kidd et al., 2016), and the same occurs with *Chaetomium globosum*, *Cladosporium cladosporioides*, *C. herbarum*, *C. oxysporum*, *C. sphaerospermum*, *C. subuliforme*, *C. tenuissimum*, *Nigrosporasphaerica*, *Penicillium aurantiogriseum*, *P. chrysogenum*, *P. citrinum*(de Hoog et al., 2000; Kidd et al., 2016; Al Hallak et al., 2023), hence their presence in these environments can be risky for the personnel health.

Regarding the dust collected in the repositories, it was evident that the dust load collected in ML was significantly lower than that obtained in R-13, however, these values were slightly higher than that previously reported by Borrego et al. (2022a), whose indicate an average of 8.5 mg/m^2/days. Although there is no previous report of dust determinations in R-13, there is a precedent that refers to the dust load collected in R-14 (repository located next to R-13) and whose value was 22.8 mg/m^2/days, that is, lower than that found in this study (Borrego et al., 2022a). However, the dust concentrations obtained in this research are much lower than the values reported by Rodríguez (2016) in a study made in the Documentation Center of the National Museum of Music in Cuba (64 mg/m^2/day) a heritage institution located in Old Havana, very near the Port Avenue and the NARC, and are higher than those obtained in two of the repositories analyzed in the Provincial Historical Archive of Villa Clara (PHAVC) (Borrego et al., 2022c).

In relation to the fungal concentration existing in the collected dust, statistically significant differences were evident between the repositories analyzed possibly due to the diverse air circulation systems existing in them that favor differences in the environmental characteristics of each one. The R-13 characterized by have ventilation ducts, it has a lot of dust loaded with fungal propagules accumulates, but, inside the repository the sedimentation of the dust and the fungal load that it carries is low, probably due to the natural-cross ventilation that exists in this place. In addition, the inclination that these ducts have in the wall (approximately 45°) plays an important role in trapping dust that comes from outside. In a prior study carried out in ML, a much higher fungal concentration was obtained from the dust (6 x 10^5 CFU/g) (Molina and Borrego, 2014). Nevertheless, ML is a climate-controlled repository (with an average T of 22°C ± 2°C and an average RH of 58.5% ± 2%), which probably favors the permanence of high viable fungal concentrations in the settled dust, because this repository don't have continuous replaces of air with outdoor, while in R-13, due to having natural-cross ventilation, the mycobiota moves along with the dust (enters and leaves) is almost constantly replaced, which can sometimes be very low, particularly on very rainy days where it occurs the washing of the outdoor air (Sabariego et al., 2004), or it is exposed to intense solar irradiation where temperatures reach 45°C under the sun.

Although form the settled dust 7 and 13 genera were isolated in ML and R-13 respectively, the genera *Aspergillus*, *Cladosporium* and *Penicillium* prevailed, and they were previously reported as predominant in this niche (Shan et al., 2019; Andersen et al., 2021), and it has even been possible to visualize through scanning electron microscopy *Aspergillus* conidiophores developing on dust particles (Nastasi et al., 2020), indicating that members of this genus can grow at the expense of the nutrients present in this niche. On the other hand, it has been reported that the genera *Aspergillus*, *Cladosporium* and *Penicillium* are very abundant in the dust of houses located in Havana, Cuba (Sánchez-Espinosa et al., 2021), an example of what seems to be a common in the country capital. Besides, when comparing the rest of the obtained genera with preceding reports, it can be seen that the number of genera found in this study is greater. When the current results were compared with those obtained by Molina &Borrego (2015) and Borrego et al. (2022a) it is detected that six and five genera were isolated from ML on those occasions, respectively, with a predominance of *Aspergillus*, while now seven were isolated, with a prevalence of *Cladosporium* genus. Even if in 2015 and 2022 from ML *Chaetomium* and *Humicola* were isolated, on this occasion those genera were not detected and instead *Fusarium*, *Neurospora*, *Nigrospora*, *Talaromyces* and *Ulocladium* were found. Most of these genera were isolated from air, with the exception of *Talaromyces* and *Ulocladium*. The presence of *Fusarium*, *Nigrospora* and non-sporulating mycelia in the dust of Havana houses has been reported (Sánchez-Espinosa et al., 2021), which reveals that they are frequent taxa and propagules in the dust of the capital. In contrast, *Talaromyces* (*Penicillium* teleomorph) has been detected for the first time in dust collected at NARC, although this genus has been isolated from air from Provincial Historic Archive of Santiago de Cuba (PHASC) (Borrego et al., 2022b) and PHAVC (Borrego et al., 2022c) repositories, and species of this genus were also found from airborne of

Pakistani libraries (Hassan et al., 2021). Likewise, a few years ago this genus was detected on the surface of different types of documents kept at the NARC (Borrego et al., 2017). However, *Ulocladium*, a humidity indicator genus according to Codina et al. (2008) and Nastasi et al. (2020), was detected in the air of French archives (Roussel et al., 2012) and from a Brazilian library (Silva et al., 2021), but it had never been isolated before in a Cuban archive (air, dust or document), which is why it is the first record.

On the other hand, a greater diversity of genera was obtained from the dust collected in R-13. Among them *Aureobasidium*, *Botrytis*, *Drechslera*, *Paecilomyces*, *Pithomyces* and *Trichoderma* were new records for dust of the NARC and some of them such as *Botrytis*, *Drechslera* and *Pithomyces* have not even been detected in the air of NARC repositories (Borrego, 2023), which corroborates the novelty of the findings. Instead, *Trichoderma*, is a genus that has been previously isolated from the NARC repositories air (Borrego et al., 2017; Borrego, 2023), even from the air of ML (Molina and Borrego, 2014).

It should be highlighted that for the first time in this study, the dust accumulated in the ventilation ducts of a NARC repository characterized by having natural-cross ventilation was analyzed and the highest fungal concentration and the greatest diversity of genera were obtained. This high concentration and fungal diversity in the dust that settles in the ventilation ducts of the studied repository suggests that the outdoor environment is an important source of fungal contamination for indoor environments (Viegas et al., 2020; Hassan et al., 2021), and particularly for this repository, since precisely on that side of the building (south side), there are gardens with abundant vegetation and leafy trees, as well as avenues with high pedestrian and vehicular movement, which it favors a high movement of bioparticles (deposition/dispersion) and with it a greater diversity of fungal taxa (Pinzari, 2011; Xing and Brimblecombe, 2019). Probably, similar results could be obtained in other NARC repositories with analogous characteristics of ventilation if they were analyzed.

A total of 14 genera were detected both in the dust collected in R-13 and in the dust accumulated in the ventilation ducts, which represents a 46.7% coincidence. This indicates that almost half of the genera detected in the collected dust may come from dust accumulated in the ducts and that the air is responsible for introducing them into the indoor of R-13. Within the common genera, the aforementioned new records *Aureobasidium*, *Botrytis*, *Drechslera* and *Pithomyces* were found, as well as *Alternaria*, *Aspergillus*, *Botryotrichum*, *Cladosporium*, *Fusarium*, *Neurospora*, *Paecilomyces*, *Penicillium*, *Trichoderma* and *Rhodotorula*. On the other hand, the genera *Botryotrichum*, *Chaetomium*, *Neurospora*, *Ovulariopsis*, *Zygosporium*, *Candida* and *Rhodotorula* (which were obtained from the dust collected and/or dust accumulated in the ventilation ducts) are genera that were also detected in the air of R-13, demonstrating that they were introduced into the indoor environment by dust.

Dust constitutes a source of microorganisms in indoor environments (Osman et al., 2018) and in particular of fungi (Aleksic et al., 2017). The re-suspension of settled dust on the surfaces of objects and furniture is responsible for the increase in the microbial load in the air (Fröhlich-Nowoisky et al., 2009). On the other hand, the dust that penetrates in a premises is the transport element of particles (including bioparticles), nutrients and water (Viegas et al., 2020); In fact, dust is considered the greatest source of nutrients for fungi, hence dusty conditions intensify fungal contamination (Borrego et al., 2015; Borrego et al., 2022a). Likewise, it promotes a microenvironment on the surface of the materials (documents) that prevents the normal flow of air, so large surfaces covered with dust absorb a large amount of moisture, thus guaranteeing the conditions for the fungi it carries to begin to develop and degrade the supports that are part of the artworks, heritage collections and documents (Borrego, 2015; Awad et al., 2020; Abdel-Maksoud et al., 2022). Furthermore, it promotes the re-suspension of fungal propagules into the air of the repositories and their environmental dispersion when dusty documents are rubbed or handled, facilitating their inhalation as well as the volatile compounds product of fungal metabolism (Aleksic et al., 2017). Therefore, the dust is considered as the cause of a second source of contamination of valuable collections in archives, libraries and museums (Schneider, 2003; Skóra and Gutarowska, 2016; Shan et al., 2019) and for this, dust is considered an important ecological niche to carry out microbiological diagnosis in an indoor environment, since it is a reflection of the accumulation of fungal propagules (Borrego et al., 2021b); however despite this, it is not used as a reference to indicate the microbiological quality of an environment.

In relation to the species isolated from the dust, a marked predominance of those belonging to the genera *Aspergillus*, *Cladosporium* and *Penicillium* was also obtained, which coincides with findings previously found in the dust collected in NARC and PHAVC repositories (Molina &Borrego, 2014; Borrego et al., 2022a, c), as well as in an Italian archive (Maggi et al., 2000) and an Egyptian museum (Awad et al., 2020).

The dust collected in NARC repositories has not been a highly studied niche; therefore, there are not many references about the diversity of species that may exist. Nevertheless, for the first time, species of the genera *Aureobasidium*, *Botrytis*, *Botryotrichum*, *Chrysosporium*, *Cunninghamella*, *Drechslera*, *Epicoccum*, *Fusarium*, *Gliomastix*,

Mucor, *Ovulariopsis*, *Nigrospora*, *Paecilomyces*, *Pithomyces*, *Rhizopus*, *Syncephalastrum*, *Talaromyces*, *Trichoderma*, *Ulocladium* and *Wallemia* were isolated. However, representatives of the genera *Chrysosporium*, *Fusarium*, *Nigrospora*, *Rhizopus* and *Talaromyces* were detected in the dust collected in the PHAVC (Borrego et al., 2022c). Likewise, species of *Aureobasidium*, *Botrytis*, *Drechslera*, *Mucor*, *Paecilomyces*, *Pithomyces*, *Rhizopus*, *Syncephalastrum*, *Talaromyces*, *Trichoderma*, *Ulocladium* and *Wallemia* were detected in dust collected in archives and libraries in other countries (Maggi et al., 2000; Roussel et al., 2012; Skóra et al., 2015; Chary, 2017). On the other hand, the genera *Cunninghamella*, *Epicoccum*, *Fusarium*, *Gliomastix*, *Nigrospora*, *Pithomyces* and *Rhizopus* were isolated from the atmosphere of Havana (Almaguer et al., 2014; Sánchez et al., 2019; Díaz et al., 2020; Cruz et al., 2021), suggesting that their existence in dust may be due to their presence in the outdoor air. Although the genus *Cunninghamella* is quite rare in the indoor air of archives, libraries, and museums, it was detected in the air of a Mexican library (Moctezuma-Zárate et al., 2015) and recently in the indoor air of a naturally ventilated Colombian library (Camargo et al., 2022), likewise, species of this genus were detected on documents kept in Brazilian libraries (Leite-Jr et al., 2018) and in an Italian museum (Montemartini-Corte et al., 2003).

About the concentrations of fungi found on the surface of the evaluated documents (between 5 x 10 to 3.5 x 10^2 CFU/cm^2), it can be affirmed that these values turn out to be similar to those obtained in previous studies where different types of documents preserved in NARC (Borrego et al., 2018; Borrego et al., 2017) and they are lower than those found on Portuguese (Pinheiro, 2014) and Polish documents (Skóra et al., 2015), but they are higher than those found on documents sampled in the PHAVC (Borrego et al., 2022c) and from other countries (Zielińska-Jankiewicz et al., 2008; Niesler et al., 2010), demonstrating the presence of dirt and dust settled on them even though it is not visible to the naked eye, which could contribute to air cross-contamination (Wu et al., 2021).

The genera isolated from the documents belong to the phylum Ascomycota, which coincides with preceding studies (Savoldelli et al., 2021; Escudero-Leyva et al., 2023) and a marked predominance of species of the *Aspergillus* genus was obtained (7 in maps and 11 in NP) followed by *Penicillium* (4 in maps and 3 in NP). This behavior is consistent with prior reports both in Cuba (Borrego et al., 2015; Borrego et al., 2017; Borrego et al., 2018; Borrego et al., 2022c) and in other countries (Abdel-Maksoud et al., 2022; Saada et al., 2023) demonstrating the importance that *Aspergillus* species can have in the possible biodeterioration of paper documents.

Among the species isolated from documents were *A. flavus*, *A. niger*, *P. citrinum*, and *P.janthinellum* which have been previously isolated from documents conserved at NARC (Molina & Borrego, 2014; Borrego et al., 2017), other species not common to both types of documents were also previously detected; among them are *A. candidus*, *A. ornatus*, *Candida* sp., *C. cladosporioides*, *Paecilomyces variotii*, *P. citrinum*, *P. janczewskii*, as well as non-sporulating mycelia (Molina &Borrego, 2014; Borrego et al., 2015; Borrego et al., 2017). Likewise, some species detected in this study turned out to be new records found on the NARC documents, such is the case of *A. nidulans*, *A.penicillioides*, *A. restrictus*, *A. sydowii*, *C. sphaerospermum*, *P. aurantiogriseum*, *Torula* sp., and *Zygosporium* sp.

It should be noted that members of the genera *Trichophyton* and *Gliomastix* were isolated from documents for the second time, since the first report was made by Borrego et al. (2018). The same occurs with the genus *Talaromyces*, since previously the species *Talaromyces helicus* was isolated from a map preserved in ML (Borrego et al., 2012) and the species *Talaromyces flavus* was isolated from a photo conserved in the photo-library of the NARC (Borrego et al., 2017). Likewise, *Talaromyces* species have been isolated from documents in different supports preserved in archives from Poland (Skóra et al., 2015; Kwaśna et al., 2020), Greece (Karakasidou et al., 2018), Argentina (Borrego et al., 2018) and the Czech Republic (Branysova et al., 2021) as well as in an Egyptian museum (Saada et al., 2023). Representatives of *Gliomastix*genus were detected on NPs preserved in R-13 and in the dust collected in the ventilation ducts of R-13, revealing that this species was probably deposited on the documents after having entered the air of this repository together with the dust. Although this genus was not detected in the outdoor air in this study, in previous investigations made both in the indoor environment of a Cuban museum with natural ventilation (Cruz et al., 2021) and in the outdoor environment of Havana it was detected (Sánchez et al., 2019), indicating that its existence in the indoor environment may be due to the incidence of the outdoor environment. In addition, representatives of this genus have been isolated from the surface of books kept in the library of Istanbul, Turkey (Kadaifciler, 2017). Regarding the *Torula* and *Zygosporium* genera, their species have been isolated from the air or the dust of Cuban archives (Borrego &Molina, 2018; Borrego et al., 2022a, b, c) but never before from documents. However, other yeast species such as *Candida* sp. and *Rhodotorula* sp., have been isolated from documents of the NARC (Borrego et al., 2017; Borrego et al., 2018). In spite of, representatives of *Torula*genus have been isolated from audiovisual materials in the Czech Republic (Branysova et al., 2021).

It should be noted that non-sporulating and septate mycelia were again isolated from the surface of documents, a result that agrees with previous findings in Cuba (Borrego et al., 2017; 2018). These types of mycelia were also detected in the air and in dust, so it could be inferred that their existence on the documents comes from one or the two aforementioned niches.

In general, the species isolated in these three niches (air, dust and document surfaces) *A. flavus*, *A. niger*, *A. sydowii*, *A. versicolor*, *Chaetomium globosum*, *C. cladosporioides*, *C. herbarum*, *C.oxysporum*, *C. sphaerospermum*, *F. graminearum*, *Nigrospora sphaerica*, *Paecilomyces variotii*, *P. aurantiogriseum*, *P. chrysogenum* and *P. citrinum* were also detected in outdoor air (Almaguer & Rojas-Flores, 2013).

As *Aspergillus* is a genus that has a large number of allergenic, primary pathogenic and opportunistic species, as well as mycotoxin-producing species (de Hoog et al., 2000; Navale et al., 2021) that can be inhaled or come into contact with the skin (Viegas et al., 2018), there is a tendency to characterize the isolates until species in order to identify the risk they represent to the personnel health working in archives, libraries and museums. It is necessary to highlight that the most abundant species of this genus isolated from indoor air were *A. flavus*, *A. glaucus*, *A. japonicus*, *A. niger* and *A. restrictus* and they have been isolated in these environments both in Cuba (Rodríguez et al., 2014; Borrego &Perdomo, 2016; Molina & Borrego, 2016; Borrego & Molina , 2019; Borrego et al., 2020; Borrego & Molina, 2020; Borrego et al., 2021a; Borrego et al., 2022a, b, c) as in other countries (Leite-Jr. et al., 2018; Saada et al., 2020; Camargo et al., 2022). Likewise, the species *A. flavus*, *A. glaucus*, *A. niger*, *A. oryzae*, *A. restrictus*, *A. sydowii*, *A. tamarii*, *A. terreus*and *A. ustus* were previously found in the indoor air of Havana libraries and museums (Rojas et al., 2012; Borrego &Molina, 2019). On the other hand, the most abundantly species of this genus detected in the dust were *A. flavus*, *A. glaucus*, *A. niger*, *A. restrictus* and *A. versicolor* and they were previously isolated from the dust collected in NARC repositories (Borrego et al., 2022a), likewise,*A. flavus*, *A.niger*, and *A. restrictus* were also detected in the dust collected in other Cuban archive (HPAVC) (Borrego et al., 2022c), while the most species found in document surfaces were *A.flavus*, *A. glaucus*, *A. niger* and *A. penicillioides* which were previously detected on documents from NARC (Molina &Borrego, 2014; Borrego et al., 2017) and documents in different supports of other countries (Sakr et al., 2018; Purkrtova et al., 2022; Saada et al., 2023).Precisely the species *A. flavus*, *A. japonicus*, *A. niger*, *A. oryzae*, *A. sydowii*, *A. tamarii* and *A. terreus* have been found in the atmosphere of Havana (Almaguer et al., 2021), and perhaps for this reason they could have penetrated into the indoor environments through the existing ventilation or air conditioning systems in the repositories remaining in the air, next to dust or settling on documents.

For their part, the species *C. cladosporioides*, despite having been isolated from the air and dust of both studied repositories (abundant species), were only detected on the notarial protocols preserved in R-13 and were not found on any of the maps analyzed. This infers that the furniture (drawer horizontal flat file) where the maps are kept is playing an important protective role, while the existing open shelving in R-13 allows a greater deposition of fungal propagules on the documents.

It is known that the species *C. cladosporioides* and *C. sphaerospermum* produce mycotoxins (Al Hallak et al., 2023), and together with *C. herbarum* can cause severe allergic states in people (Sandoval-Denis et al., 2015; Luo et al., 2016) as well as superficial mycosis on the skin and nails (Lysková, 2007) and it is also an opportunistic pathogen (de Hoog et al., 2000; Ma et al., 2021). Likewise, that *Fusarium graminearum*, *Nigrospora sphaerica*, *P. aurantiogriseum*, *P.chrysogenum* and *P. citrinum* are opportunistic pathogens (de Hoog et al., 2000; Zinedine et al., 2007; Sham et al., 2021) and particularly *Fusarium graminearum*, *P. aurantiogriseum* and *P.citrinum* produce also mycotoxins (Frisvad and Samson, 2004) and cause superficial mycosis (Lysková, 2007), so that inhalation of their propagules or the simple contact with them for a long time, these species can be dangerous to health.

In Cuba it has been shown that personnel working in archives are exposed to biohazard circumstantially (Borrego et al., 2021b, 2022c; Herrera et al., 2021) and it has been suggested that the cheapest, ecological and sustainable way to mitigate this effect is using personal protective equipment for document handling.

About the common taxa detected in the studied niches, it could be seen that the total species similarity (QS_{TOTAL}) between IA and OA was medium (0.6), but in a previous study this coefficient was higher (0.8) (Borrego et al., 2022a). Likewise, the QS_{TOTAL} values obtained when comparing the AI with the CD varied from medium to high depending on the sampling analyzed, agreeing with the result previously reported by Borrego et al. (2022a). However, the behavior of QS_{TOTAL} had never before been analyzed to compare the indoor air with the document surface (IA-DS) and the document surface with collected dust (DS-CD), therefore these results cannot be compared with others previously obtained both in Cuba and in other countries. However, these values were very low and indicative that the surface of the documents provides little information to the total mycobiota of an indoor archival environment; therefore, it is a niche from which its study can be dispensed with.

When the QS are analyzed for each predominant genus in particular, it is observed that the specific QS for *Penicillium* species ($QS_{Pen.}$) were very low in all cases, demonstrating that the *Penicillium* species were few in all the niches evaluated; however, Borrego et al. (2022a) reported a $QS_{Pen.}$ high (0.7) when comparing AI with DC. The opposite occurred when IA-OA and IA-CD were compared for *Aspergillus* and *Cladosporium* species; in those cases, the QS fluctuated from medium to high values depending on the sampling, and showed a higher trend in relation to the report by Borrego et al. (2022a). Likewise, the behavior of $QS_{Asp.}$ and $QS_{Clad.}$ had never before been analyzed to compare the indoor air with the document surface (IA-DS) and the document surface with collected dust (DS-CD), therefore these results cannot be compared with others obtained previously. However, these values were very low, demonstrating once again that document surfaces contribute very little to environmental fungal diversity and that the greatest contribution and with a high interrelationship of species is provided by OA, IA and DC.
These results showed, on the one hand, that the outdoor air exerts a considerable influence on the indoor air of the premises, particularly for environments with natural ventilation, and on the other, that dust influences the species diversity of the indoor air of the premises; and that both airborne and dustborne mycobiota can influence mycobiota that can be detected on the documents surface. Likewise, it was demonstrated that the fungal diversity in these three ecological niches allows evaluating the environmental fungal quality and its possible impact on the conservation of heritage documents and the personnel health in the archives. On the other hand, it was evidenced that the fungal species diversity profile followed the following order: in the deposited dust > airborne > document surface.

CONCLUSIONS

The maximum concentrations of fungi were obtained in the season of little rain (770 CFU/m^3 in R-13 and 440 CFU/m^3in ML) and the indoor/outdoor (I/O) ratios in all cases were ≤ 1.0, demonstrating the good environmental quality of the repositories despite having different air circulation characteristics since R-13 is naturally ventilated, while ML is a climate-controlled repository. *Aspergillus* and *Cladosporium* genera were dominant in the indoor air of the repositories followed by *Chrysosporium* and *Candida*that were classified as common genera. In this niche four species were abundant (*A. flavus*, *A. niger*, *C. cladosporioides* and *C. oxysporum*) an all cases the I/O ratios were ≤ 1.0 evidencing that those species come from the outdoor air that penetrated in the repositories. The average dust loads collected in ML and R-13 were 11.7 and 31.6 mg/m^2/day, respectively. From dust, the predominant genera were *Cladosporium*, *Penicillium* and *Aspergillus*, but *Aureobasidium*, *Botrytis*, *Drechslera*, *Paecilomyces*, *Pithomyces* and *Trichoderma* were new records for the ARNAC´s dust; likewise, a high diversity of species was obtained. In the collected dust, 15 species belonging to five different genera were classified as abundant (*Aspergillus athecius, A. flavus, A. glaucus, A. niger, A. restrictus, A. versicolor, Cladosporium cladosporioides, C. herbarum, C. lignicola, C. sphaerospermum, Fusarium graminearum, Neurospora crassa, Penicillium aurantiogriseum, P. chrysogenum, P. citrinum*). Although the documents analyzed were apparently clean, fungi were isolated in concentrations that ranged between 5 x 10 and 3.5 x 10^2 CFU/cm^2. The species *A. flavus*, *A. glaucus*, *A. niger*, *P. janthinellum* and *P. raistrickii* were common to all the documents analyzed. Although the environments were not contaminated, species risky to human health were isolated. The species similarity (Sørensen similarity coefficient, QS) among the analyzed niches (indoor air, outdoor air, collected dust, document surface) showed that the QS_{TOTAL} obtained by comparing the species of the indoor air of the repositories with those of the dust collected, was high. The species diversity profile followed the order: settled dust > airborne > document surface.

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Sofía Borrego: Conceptualization, Acquisition of funds, Administration of the project, Formal analysis, Investigation, Methodology, Supervision, writing – original draft, Writing – review & editing, Visualization.
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The authors declare that there is no conflict of interest.