



Assessment of airborne *Curvularia* propagules in the atmosphere of Havana, Cuba

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Received: 5 August 2020 / Accepted: 3 November 2020
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Abstract Even though the presence of *Curvularia* conidia in the tropical air and the role in respiratory allergy symptoms or plant diseases incidence has been reported, traditional aerobiological investigations usually do not consider their profound study. An aeromycological study of the *Curvularia* propagules in Havana was conducted during the years 2017 and 2018. Monitoring was carried out by means of volumetric viable and non-viable methodologies. Three types of microscopically identifiable airborne spores (Lunata, Genuiculata and Maculans) were identified, according to their distinctive features (conidium shape, pigmentation, number and position of the septa), defined by optical microscopy with 400 × magnification. The spores of *Curvularia* showed prevalence during rainy months and were predominantly abundant through the daytime. The annual distribution showed the prevalence of the Lunata type, with a diurnal pattern and corroborated through the isolated species since all belonged to this morphological group. The average temperature and relative humidity, as well as the precipitations,

correlated positively with the concentrations. The wind speed showed a negative correlation, which indicates that the climatic conditions of the Havana influence the aerobiological dynamics of airborne *Curvularia* propagules.

Keywords Airborne fungi · Atmosphere · Spores · Genuiculata · Lunata · Maculans

1 Introduction

The atmosphere of tropical cities has a high fungal diversity, which can be reflected in the greater number of propagules collected in air samples (Menezes et al. 2004; Prasla et al. 2013). Out of the different methods available for the airborne fungi identification, direct microscopy and culture-based techniques are widely used. Many airborne fungal spores are often implicated in respiratory allergy symptoms or plant diseases incidence (Levetin et al. 2016). This is significant, in order to reduce outdoor contamination of the air, that badly affects atopic and non-atopic individuals, and to elude losses in urban crops. According to Esch and Codina (2017), some airborne species of *Curvularia* are associated with hypersensitivity diseases, such as asthma, allergic rhinitis and allergic bronchopulmonary mycoses. The exposure to this fungal genus has also been associated with allergic fungal sinusitis or allergic fungal rhinosinusitis, since it can be

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detected from sinus exudates from patients with these conditions (Singh 2019). Nowadays, *Curvularia* is responsible for a high skin prick test response in tropics, such as (26–32%) in Singapore (Chew et al. 2000). In temperate zones, Larenas-Linnerman et al. (2016) noted the frequency of skin prick test reaction to *Curvularia* was 3.8% after important flood in the USA (post-Katrina hurricane) and 18% in Finland among children with perennial and seasonal asthma.

The conidia of *Curvularia* are frequently observed in air samples, although they are rarely abundant in some aerobiological studies in temperate climates (Levetin et al. 2016). Some investigations noted that these propagules are more frequent and dense in the atmosphere of tropical or subtropical countries (Wu et al. 2007; Chen et al. 2014). The prevalence of airborne *Curvularia* spores in the atmosphere of Havana (Cuba) was reported for the first time by Quintero (1964), later by Arnold et al. (1987) and then in almost all the subsequent aerobiological investigations (Rojas et al. 2007; Almaguer et al. 2013; Almaguer and Rojas, 2013). However, many of these works were based on momentary sampling with viable methods and fundamentally providing data on the presence of the genus. The frequency of *Curvularia* in this neotropical city was only exceeded by *Cladosporium*, *Leptosphaeria*, *Coprinus*, *Aspergillus* and *Penicillium* (Almaguer and Rojas 2013; Almaguer et al. 2013, 2015). Even though the high occurrence of *Curvularia* conidia in the tropical air, traditional aerobiological investigations usually do not consider their study, but instead focuses on other conidia genera such as *Cladosporium*, among others.

The fungal genus *Curvularia* is represented by a large number of species distributed worldwide, usually characterized by cosmopolitan or pantropical distribution patterns. There are more than 130 species registered in Mycobank (<https://www.mycobank.org>) or Index Fungorum (<https://www.indexfungorum.org>) recognized as saprobes or pathogens, in association with plants, animals and humans (Sivanesan 1987; Hyde et al. 2017; Manamgoda et al. 2015; Tan et al. 2018). In Cuba, there is a great variety of substrates where can be found more than 20 species of this genus, among which the Poaceae stand out as the most numerous family of the phanerogamic flora (Mena 2004; Camino et al. 2006).

Curvularia genus produces curved or lunate shaped conidia, which can also be oblong, claviform,

ellipsoidal, fusiform, obovoid or piriform. Its color is a pale brown, to dark brown, often with the cells at the ends lighter than the rest and sometimes with dark bands on the septa. In some species, the conidial wall is smooth to verrucose and hilum is protuberant (Manamgoda et al. 2012; Ariyawansa et al. 2015). Previously, Boedijn (1933) divided *Curvularia* into three groups of species (Lunata Geniculata and Maculans) based on morphological features of conidia. The Lunata group was characterized by producing four-celled, more or less curved spores wherein one of the middle cells is inflated and darker. Species with conidia of five cells were placed in the Geniculata group, while the Maculans group was proposed for species with four cells, straight or more or less asymmetric with the central cells larger and darker than the final cells (Sivanesan 1987; Da Cunha 2014; Madrid et al. 2014). This morphological classification can be useful in the aerobiological monitoring based on a volumetric spore trap of the Hirst design (Hirst 1952) and direct counting of *Curvularia* conidia. From spores alone, it is not probable to differentiate species with certainty, but it is possible to distinguish at least these three types. It has been widely reported for *Cladosporium*, but also for *Coprinus* or *Leptosphaeria* (Aira et al. 2009, 2013; Hernández Trejo et al. 2012).

Continuous monitoring of the atmosphere allows determining the spore spectrum with a particular interest within the air, making it possible to compare their annual dynamics. To identify the most important types within a wide range of airborne fungi and to study their temporal patterns in an area of economic and social interest, the combination of quantitative and qualitative studies is appropriate. In this way, it contributes to the knowledge of the ecology of the fungi that are dispersed through the air.

This research aims to determine the presence and distribution of *Curvularia* spores in the atmosphere of the city of Havana during 2017 and 2018 by means of the combination of viable and non-viable methodology: a volumetric Hirst type sampler and a slit-to-agar impactor Aeroscope. The relationship between the concentrations of spores in the air and the main meteorological variables was also analyzed. This information could help us to issue alerts of higher high *Curvularia* spore concentrations and thus prevent the development of diseases in sensitized people and losses in urban crops.

2 Materials and methods

2.1 Sampling area

A two-year study was carried out in Havana, located on the northwestern coast of Cuba, at an altitude of 24 m above sea level. Havana is the capital and the biggest and most populated city in the country.

2.2 Meteorological conditions

The climate of Havana is subtropical, with a rainy season (May to October) and a dry season (November to April). The daily values of the meteorological variables were recorded at the meteorological station of Casablanca, located in Havana city and belonging to Cuban Institute of Meteorology (INSMET). The meteorological variables recorded were wind speed (km/h), mean temperature (°C), maximum temperature (°C), minimum temperature (°C), mean relative humidity (%), maximum relative humidity (%), minimum relative humidity (%) and rainfall (mm). The average annual temperature was 25.5 °C during both studied years, the total annual rainfall was 1131 and 1221.5 mm, in 2017 and 2018, respectively. The relative mean humidity reached 76.2%, during the first year and 76.9% in the second one.

2.3 Sampling and detection of airborne *Curvularia* propagules

The airborne fungus selected for this study was that of *Curvularia* because of its allergenic and phytopathogenic properties. The spores of *Curvularia* were counted by means of direct microscopy and collected from January 2017 to December 2018 by using a volumetric Hirst type sampler-Lanzoni VPPS 2000 (Hirst 1952). Sampling was carried out continuously (operational 24 h/day). The aerobiological station was located on the rooftop of the Biology Faculty of the University of Havana (23° 08' N and 82° 23' W) at 35 m above ground level. The Lanzoni sampler was calibrated for a 10 air liters per minute flow, and the spores impacted in a Melinex tape coated with a 2% silicone solution as an adhesive. The spore count was first performed in all the samples collected following the methodology proposed by Galán et al. (2007). The magnification used was 400X for counting. The aerobiological data considered were spore

count (spores), spore mean daily concentrations (spores/m³), monthly distribution (spores), presence (spores/m³), maximum daily concentration (spores/m³), mean concentration (spores/m³) and number of days over the annual daily spore mean concentration. To determine the *Curvularia* spores diversity in the atmosphere of the study area were selected the aerobiological samples recording an average daily concentration above the annual average value in each year. Furthermore, the following characteristics were measured in these samples: conidial shape, color, spore length, spore width and number of septa. Based on its morphology, the spore types within *Curvularia* were classified following the Boedijn (1933) groups: Lunata, Geniculata and Maculans. Morphological identification was performed by means an optical Nikon Optiphot II microscope (Nikon Manufacturing, Tokyo, Japan). The spore photographs, included in this study, were obtained with an HDCE-20(CCD) camera assembled to the microscope, using a 1000X lens and software IMI Video Capture.exe. With the aim of obtaining a model reflecting intradiurnal oscillations of the spore counts during the two years, the days on which conidia counts were higher than the mean of the sampling period were selected, excluding the days with precipitation (Nilsson and Persson, 1981; Aira et al. 2009). The resulting days were used to calculate the spore concentration every two hours, thereafter expressing the data as percentages.

Cultivable *Curvularia* propagules were collected weekly at 11:00 local time hours, using a slit-to-agar impactor aeroscope (Chirana, Czech Republic) with a suction volume of 29 air liters per minute flow, and malt extract agar as culture medium (Almaguer et al. 2013; Sánchez et al. 2019). The isolates selection was performed according the distinctive features proposed by Seifert et al. (2011): brown conidiophores, bearing conidia apically or on new sympodial growing points; dark conidia (porospores), typically curved with one of the central cells enlarged, more or less fusiform, end cells often lighter, three-celled to five-celled. The identification of the isolates was based on the criteria established by Ellis (1971), Ellis (1976), Mena (2004) and Manamgoda et al. (2012).

2.4 Statistical analysis

The Mann–Whitney U test was applied to verify differences in the percentage values of total spore of

each examined *Curvularia* type (Geniculata, Lunata and Maulans) between 2017 and 2018 years. Spearman's rank correlation was used to determine the strength of the relationship between the spore daily concentrations in the air on a given day and meteorological variables in the same day and during the previous 2 days. The significance was calculated for * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, by using Statistica software version 8.0. (Statistica, Tulsa, Okla.)

3 Results

3.1 Presence of airborne *Curvularia* spores

The samples collected by means of the non-viable methodology (Lanzoni VPPS 2000) registered 2498 spores of *Curvularia* genus in the atmosphere of Havana, 75.7% during the year 2017 and 24.3% during 2018. Spores counts were higher in the samples corresponding to the months from June to October, with a more marked presence during the first year Fig. 1. During these rainy seasons, *Curvularia* spores counts reached a mean of 160 spores every month in 2017 and 60 throughout 2018. The *Curvularia* spores presence in the atmosphere was registered in 296 days (81%) during 2017, with a mean ranging from 19–30 days per month. Lower values were observed in 2018 as their spores were detected in 230 days (63%). The highest daily *Curvularia* spores concentration was recorded in different seasons in both years, on September 27th in 2017 (65 spores/m³) and March 1st in 2018 (15 spores/m³). Although the mean of the daily concentrations was also different, in both years, a higher number of days with daily concentration over the annual average were observed from June to October Table 1.

With the aim to detect the *Curvularia* spore types present in the study area, the aerobiological samples corresponding to days with a daily spore concentration above the mean annual average were selected. It corresponded to days with daily spore concentration above 6 spores/m³ and 2 spores/m³ in the first and the second year studied, respectively. As a result, 202 samples were selected, distributed in 101 for each of the two study years. The detailed analysis of the spores in the aerobiological slides allowed the differentiation

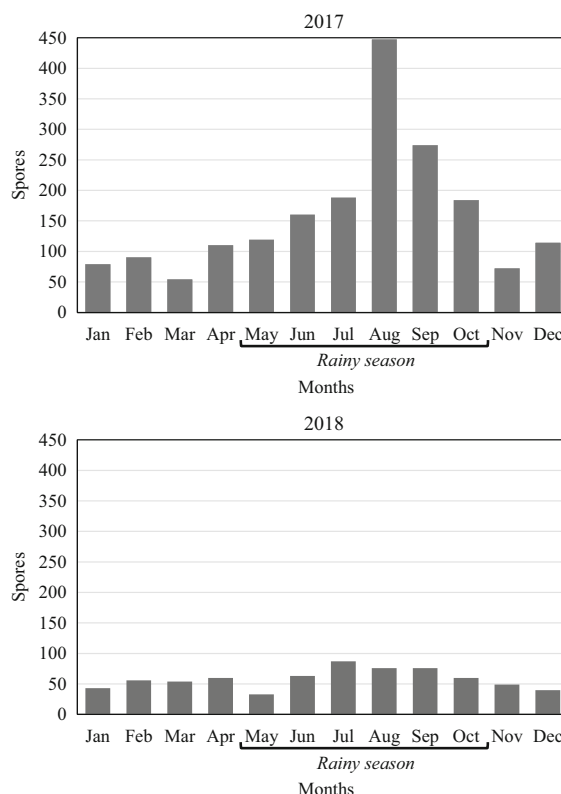


Fig. 1 Monthly distribution of airborne *Curvularia* spores trapped by Lanzoni VPPS and counted by direct microscopy during 2017 and 2018

of three types: Maculans, Lunata and Geniculata, based on its morphological features Fig. 2.

A total amount of 1789 of *Curvularia* spores was detected in the 202 selected samples of the two years of study in the outdoor air of Havana. The quantity of spores observed in the first year was 1298 spores, 1101 belonged to the Lunata spore type, 153 to Geniculata and 44 to Maculans. During the 2018 year, 491 spores were counted, belonging 409 to the Lunata spore type, 67 to Geniculata and 15 to Maculans. Quantitative data corresponding to counted spores were different between years based on marked differences detected via the U-Mann–Whitney (Z) test Table 2.

The annual distribution during both years revealed a prevalence of Lunata spores type (85% and 84% in 2017 and 2018, respectively) followed by the Geniculata spore type (12% and 14%) and in a reduced proportion the Maculans spore type (3%) Fig. 3. The main differences were detected within monthly percentages, as the Lunata spores oscillated from 74–90%

Table 1 Aerobiological data of airborne *Curvularia* spores in the atmosphere of Havana in 2017 and 2018 (presence, highest daily concentration, mean concentration and days over the annual mean)

	2017						2018						
	Presence		Highest daily concentration		Mean concentration (spores/m ³)	Days > Annual mean	Presence		Highest daily concentration		Mean concentration (spores/m ³)	Days > Annual mean	
	Days	%	(spores/m ³)	Date			Days	%	(spores/m ³)	Date			
Jan	23	6	13	06/01/2017	3	3	Jan	15	4	14	11/01/2018	1	5
Feb	22	6	17	21/02/2017	1	6	Feb	19	5	10	08/02/2018	5	9
Mar	22	6	9	30/03/2017	2	2	Mar	19	5	15	01/03/2018	2	6
Apr	25	7	30	26/04/2017	4	4	Apr	19	5	13	09/04/2018	2	6
May	25	7	16	23/05/2017	4	8	May	18	5	6	21/05/2018	1	3
Jun	29	8	21	04/06/2017	6	10	Jun	21	6	8	14/06/2018	2	10
Jul	29	8	23	27/07/2017	7	12	Jul	26	7	10	16/07/2018	3	15
Aug	30	8	46	01/08/2017	16	22	Aug	25	7	11	24/08/2018	3	13
Sep	19	5	65	27/09/2017	10	13	Sep	22	6	13	09/09/2018	3	13
Oct	23	6	25	30/09/2017	6	13	Oct	15	4	12	12/10/2018	2	7
Nov	27	7	15	22/11/2017	3	2	Nov	19	5	9	19/11/2018	2	8
Dec	23	6	24	20/12/2017	4	6	Dec	12	3	8	01/12/2018	1	6
Annual	297	81	65	27/09/2017	6	101	Annual	230	63	15	01/03/2018	2	101

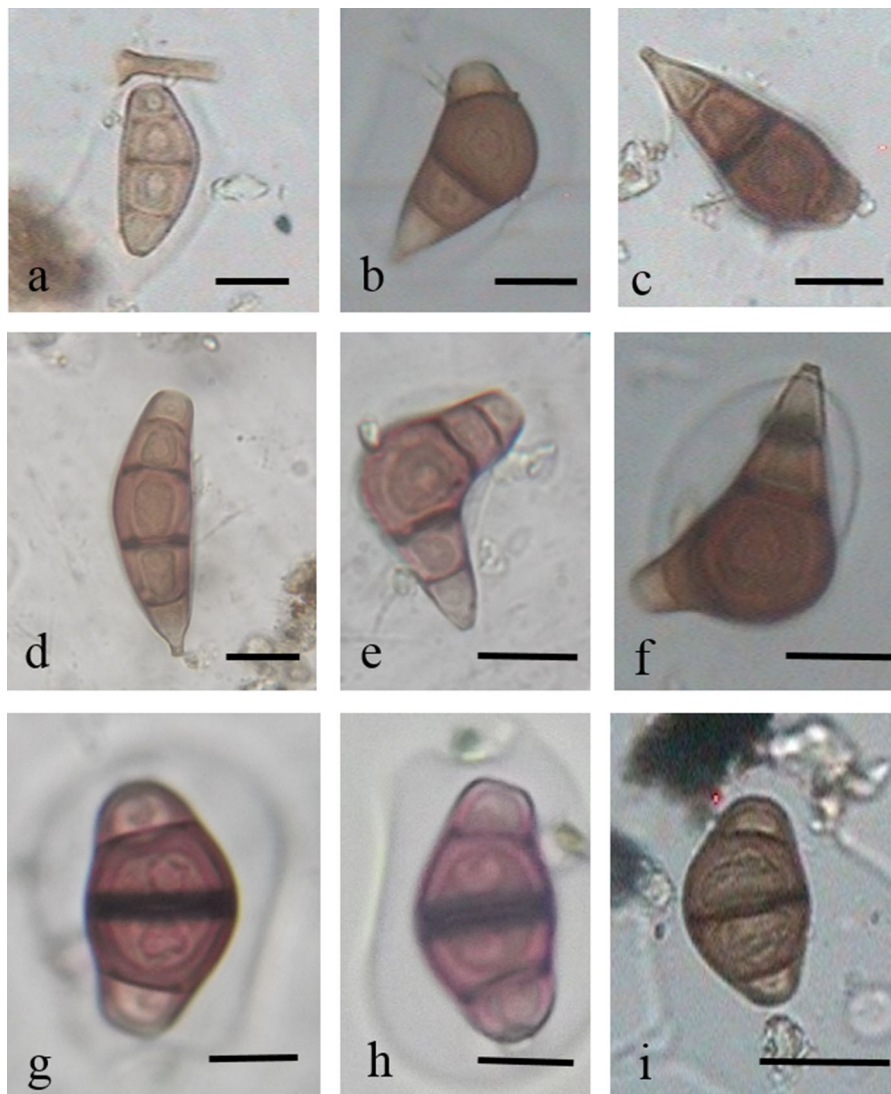


Fig. 2 Photomicrographs of airborne *Curvularia* spores types (a–c: Lunata, d–f: Geniculata, g–i: Maculans) trapped by Lanzoni VPPS 2000 in the atmosphere of Havana, Cuba. — Bars = 10 μm

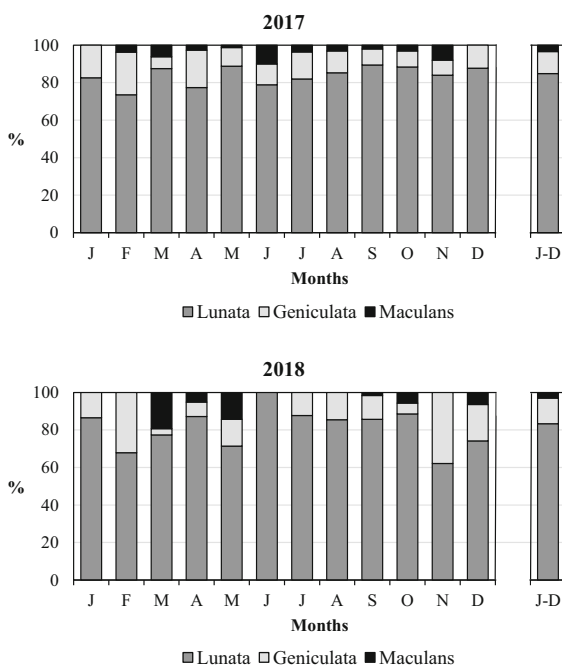
during 2017 and 62–100% during 2018, with the greatest percentages located during the rainy seasons (September and June, respectively). The Geniculata type representation fluctuated from 6–90% during 2017 and 0–38% during 2018. The percentages reached by Maculans did not exceed 10% and 19% in the first and second year, respectively, and even its presence was not detected in some months.

3.2 Daily and intradiurnal variations

Daily variations of the three spore types throughout the two years studied are presented in Fig. 4. During the first year, the highest concentrations were recorded on August 26th (41 spores/ m^3 —Lunata), April 26th (8 spores/ m^3 —Geniculata) and August 28th (4 spores/ m^3 —Maculans). In the second year, the daily concentrations registered for each spore type were lower, detecting the daily maximum peaks on January 11th (13 spores/ m^3 —Lunata and 5 spores/ m^3 —Geniculata) and March 1st (5 spores/ m^3 —Maculans). Although the

Table 2 Results of the U-Mann–Whitney (Z) test comparing mean spore concentrations and meteorology in 2017 and 2018. (* $p < 0.05$. ** $p < 0.01$. *** $p < 0.001$; ns. not significant)

Spores	2017	2018	U	Z	p-level	
<i>Curvularia</i>	5.2	1.9	44,235.000	7.855	0.000	***
Lunata	11.0	4.0	1475.500	8.669	0.000	***
Geniculata	1.5	0.7	3291.000	4.265	0.000	***
Maculans	0.5	0.1	3969.500	2.620	0.009	**
Meteorology						
Wind speed (km/h)	47.3	49.5	56,997.000	−3.203	0.001	**
T ^a min (°C)	22.3	22.2	64,697.500	0.672	0.501	n.s
T ^a med (°C)	25.5	25.3	63,243.500	1.182	0.237	n.s
T ^a max (°C)	29.5	29.3	65,015.000	0.560	0.575	n.s
RH min (%)	54.3	59.9	44,875.000	−7.630	0.000	***
RH med (%)	76.2	76.9	62,578.500	−1.416	0.157	n.s
RH max (%)	90.8	89.8	62,050.000	1.601	0.109	n.s
Rainfall (mm)	3.10	3.35	65,578.500	−0.362	0.717	n.s

**Fig. 3** Monthly percentages (%) of airborne *Curvularia* spore types (Lunata, Geniculata and Maculans) during 2017 and 2018

daily concentration values were much lower in the second year of study, in both years several days were detected with high concentrations that showed a dynamic fluctuation of these conidia in the atmosphere of Havana, being more evident in the Lunata type.

According to the diurnal behavior, *Curvularia* spores were predominantly abundant through the daytime. During the morning and the afternoon, the concentration progressively increased to reach a double peak pattern from 13–14 and 17–19 h (Fig. 5). Importantly, during the year of 2017 the sampling had a near normal diurnal variation of the spores, peaking at 18 hrs. The Lunata spore type showed a similar intradiurnal distribution, being also abundant during the daytime mainly after the noon. An analogous behavior was observed for the Geniculata spore type, but moderately different between years. While it was continuously distributed in 2017 along the daytime, with a maximum at 9–10 and 19–20 h, it is remarkable the restricted presence to 7–8 and 11–20 h during 2018. However, a double peak pattern was also indicated. The Maculans pattern also differed for the two study years, reaching higher percentages during nighttime in 2017 and through daytime in 2018.

3.3 Relationships with meteorological factors

In general, the significant correlation coefficients between the daily concentration and the meteorological variables oscillated from $r_s = 0.235$ to $r_s = 0.505$. Overall, higher significant Spearman coefficients were registered for temperature, mainly maximum temperature. In this sense, the maximum temperature of the same day, as well as the registered during the previous

days correlated positively. This association was also found for Lunata and Genuculata but only in the cases of the maximum temperature and the maximum temperature of the previous day. A similar result was observed for the relative humidity but with less significant Spearman coefficients. The maximum wind speed of the same day as well as the registered during the previous days negatively correlated with

Fig. 5 Intradiurnal distribution (%) of *Curvularia* and its spore types (Lunata, Genuculata and Maculans) in the atmosphere of Havana, Cuba, during 2017 and 2018

the *Curvularia* presence. Finally, only rainfall correlated with the Genuculata concentrations in the atmosphere (Table 3).

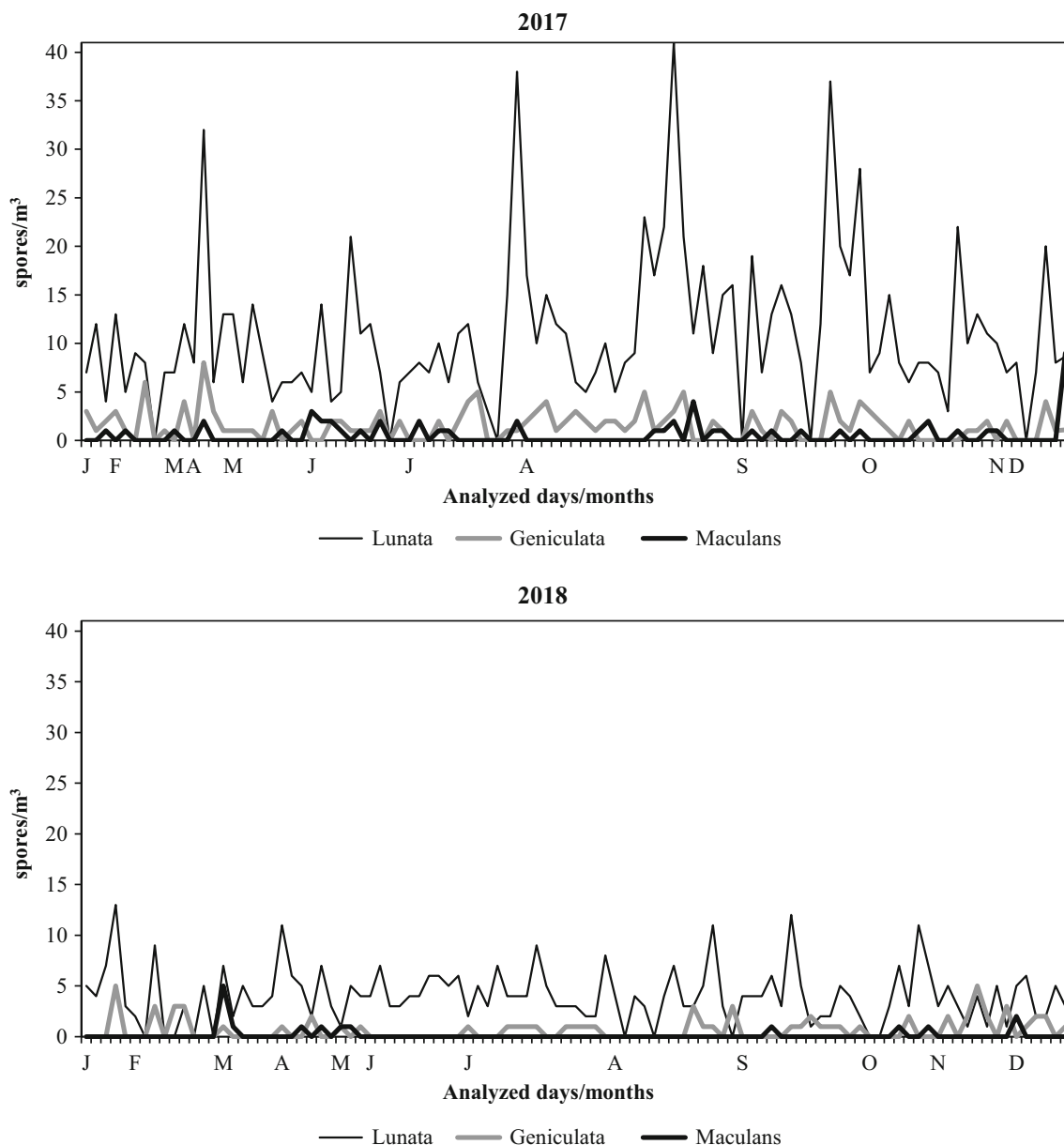


Fig. 4 Daily concentrations of *Curvularia* spore types (Lunata, Genuculata and Maculans) trapped by Lanzoni VPPS and counted by direct microscopy during 2017 and 2018

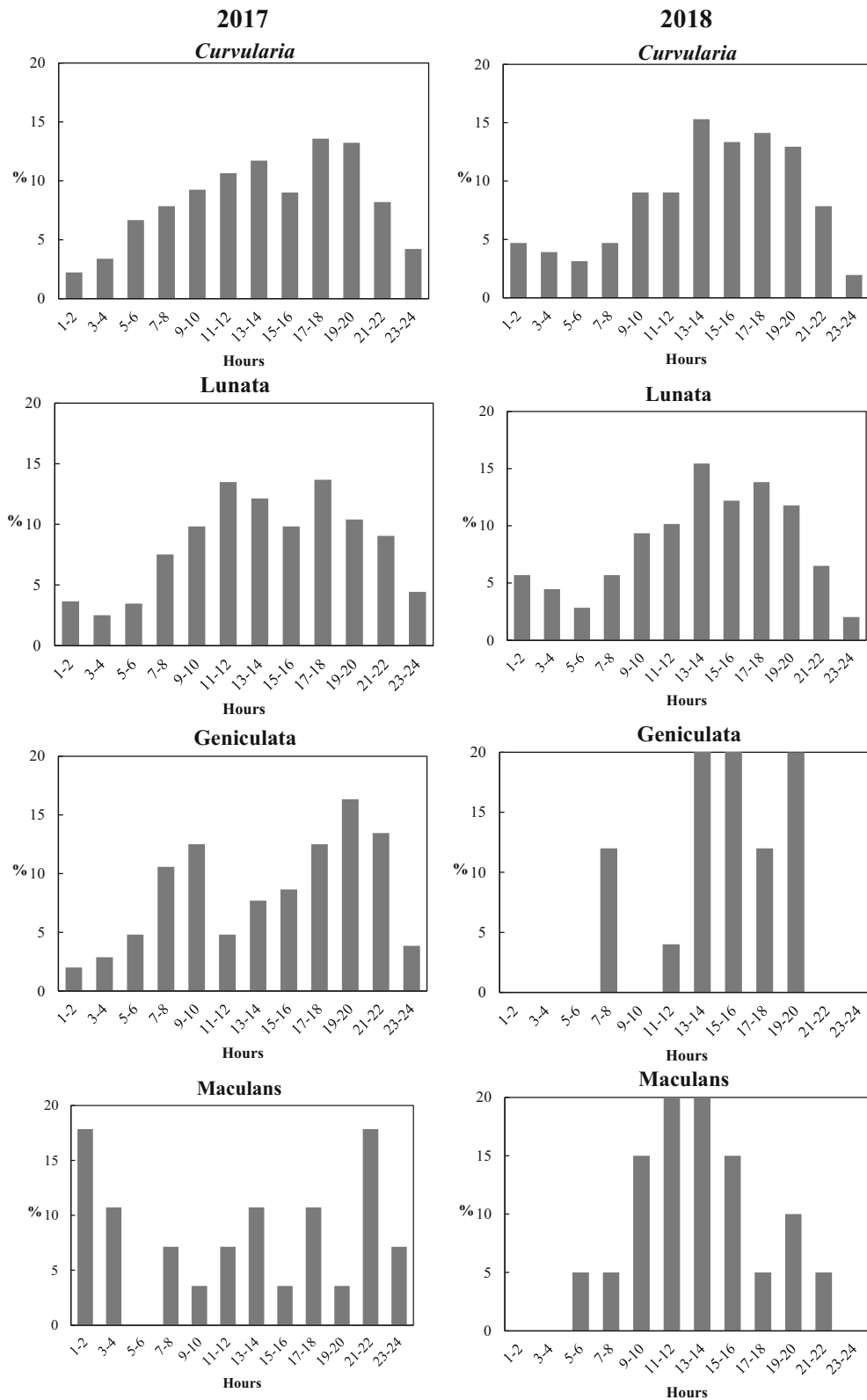


Table 3 Correlation between concentrations of *Curvularia* spore types and meteorological variables during both years (2017–2018). Spearman test significances (r_s) are marked in bold (* $p < 0.05$). n.s: no significative

Temperature	<i>Curvularia</i>	Lunata	Geniculata	Maculans
T ^a min	0.347*	0.110 n.s	0.100 n.s	0.022 n.s
T ^a min-1	0.302*	0.060 n.s	0.010 n.s	– 0.024 n.s
T ^a min-2	0.312*	0.110 n.s	0.006 n.s	– 0.014 n.s
T ^a med	0.426*	0.043*	0.120 n.s	– 0.005 n.s
T ^a med-1	0.336*	0.101 n.s	0.081 n.s	– 0.016 n.s
T ^a med-2	0.312*	0.120 n.s	0.010 n.s	– 0.040 n.s
T ^a max	0.505*	0.188*	0.238*	– 0.086 n.s
T ^a max-1	0.389*	0.145*	0.165*	0.019 n.s
T ^a max-2	0.302*	0.138 n.s	0.054 n.s	0.013 n.s
Relative Humidity	<i>Curvularia</i>	Lunata	Geniculata	Maculans
RH min	– 0.051 n.s	–0.228 n.s	–0.235 n.s	0.029 n.s
RH min-1	0.044 n.s	–0.152 n.s	–0.106 n.s	– 0.072 n.s
RH min-2	0.116*	–0.090 n.s	0.039 n.s	– 0.085 n.s
RH med	0.228*	0.087 n.s	–0.021 n.s	0.112 n.s
RH med-1	0.245*	0.112 n.s	0.048 n.s	0.060 n.s
RH med-2	0.219*	0.165*	0.178*	0.061 n.s
RH max	0.282*	0.285*	0.115 n.s	0.007 n.s
RH max-1	0.249*	0.205*	0.213*	0.006 n.s
RH max-2	0.168*	0.209*	0.209*	0.062 n.s
Rainfall	<i>Curvularia</i>	Lunata	Geniculata	Maculans
R	– 0.047 n.s	– 0.045 n.s	– 0.106 n.s	0.017 n.s
R-1	– 0.032 n.s	0.120 n.s	– 0.027 n.s	0.039 n.s
R-2	0.017 n.s	0.061 n.s	0.143*	0.019 n.s
Wind Speed	<i>Curvularia</i>	Lunata	Geniculata	Maculans
WS	– 0.166*	– 0.154*	– 0.082 n.s	– 0.118 n.s
WS-1	– 0.233*	– 0.136 n.s	– 0.113 n.s	– 0.056 n.s
WS-2	– 0.168*	– 0.209*	– 0.081 n.s	– 0.053 n.s

3.4 Airborne *Curvularia* species

Parallel to the non-viable characterization, fourteen isolates of *Curvularia* were obtained from the atmosphere of Havana during 2017 and thirteen in 2018, based on the cultivation of airborne propagules weekly collected. The isolates showed smooth, mostly three-septate conidia. The central septum was not thickened neither situated in the medial portion of the conidia. The morphological characteristics corresponded to *Curvularia aerea* (Bat. J. A. Lima and C. T. Vascon) Tsuda (9 isolates in 2017 and 7 isolates in 2018), *Curvularia pallescens* Boedijn (4 and 3 isolates,

respectively) and *Curvularia trifolii* (Kauffman) Boedijn (1 and 2 isolates, respectively). Moreover, during the second year was isolated the specie *Curvularia verruculosa* Tandon and Bilgrami ex M.B. Ellis.

4 Discussion

The *Curvularia* genus has a worldwide distribution with a wide host range with whom the fungus is associated as pathogen or saprobe (Marin-Felix et al. 2017). Most of their species are mainly graminicolous

causing diverse disease symptoms such as leaf spots, leaf blight and root rots, among others. Sivanesan (1987) identified some of the affected crops, such as maize (*Zea mays* L.), rice (*Oryza sativa* L.), sorghum (*Sorghum* spp.), sugar cane (*Saccharum officinarum* L.) and wheat (*Triticum aestivum* L.). Despite *Curvularia* affect grasses and grain crops, causing diseases of economic importance, Marin-Felix et al. (2017) reported the occurrence on this genus on *Actinidiaceae*, *Aizoaceae*, *Caricaceae*, *Convolvulaceae*, *Fabaceae*, *Iridaceae*, *Lythraceae*, *Lamiaceae*, *Oleaceae*, *Polygonaceae* and *Rubiaceae*. Therefore, the results of our study could be valuable to the urban agriculture located at the peripheral areas of Havana, considering the elevated presence of some crops belonging to the aforementioned families. According to Vázquez & Fernández (2007), it is important to contribute to the agroecological management of pests and diseases in Cuban urban agriculture. In addition, *Curvularia* plant pathogen species are associated with sugarcane seedlings, producing root rotteness and malformations affecting fertility and seedbed efficiency, which may be located in agricultural areas near Havana. *C. lunata*, *C. senegalensis* and *C. gudauskasii* represent the most common species affecting seed bank yields by up to 30% (Orberá et al. 2012). Martínez et al. (2009) reported *Curvularia* sp. as one of the principal fungi causing diseases in Triticales and wheat in San José de las Lajas, also proximate to Havana province.

Moreover, the study of airborne *Curvularia* dynamics in tropics is important for human health, mainly as consequence of its relationship with allergy episodes. In India, *C. lunata* was reported to be allergenically significant and its allergens were included in the skin prick tests (Agashe and Anand 1982; Larenas-Linnerman et al. 2016; Navpreet et al. 2017). Larenas-Linnerman et al. (2016) and Stewart and Robinson (2017) refer to the use extracts of *C. spicifera* and *C. lunata* for the diagnosis of fungal sensitization in patients with allergic conditions. The allergens of *C. lunata* involved in immune responses include Cur 1 1 (functional serine protease), Cur 1 2 (enolase), Cur 1 3 (cytochrome c), Cur 1 4 (vacuolar serine protease), Cur 1 sod (manganese superoxide dismutase), Cur 1 Trx (thioredoxin) and Cur 1 gst (glutathione S-transferase) (Williams et al. 2016). Since the past century, Agarwal et al. (1982) reported that *Curvularia* shares immunoglobulin E (IgE) cross-reactivity to other

conidial genera such as *Alternaria* and *Stemphylium*. Gupta et al. (2002) found allergenic cross-reactivity among *Curvularia lunata*, *Alternaria alternata* and *Epicoccum nigrum* based on a serological and molecular study in India. More recently, Levetin et al. (2016) suggested the same element for the *Bipolaris* complex, attributable to these fungi being closely related taxonomically. The pathogenicity of *Curvularia* is also important. *C. lunata* can cause infections associated with different classes of medical devices that is the reason for being considered an opportunistic pathogen for human (Levetin et al. 2016). McAleer et al. (1981) reported an allergic bronchopulmonary fungal disease caused by this species in Western Australia, where several *Curvularia* spp. were abundant.

Airborne fungi not only affect atopic individuals, they can also cause reactions in people not sensitized to fungal allergens and cause respiratory symptoms similar to those that occur in an allergic response. For this reason, currently in Havana, some projects are investigating sensitization to airborne fungi (Álvarez et al. 2020). Related to this, Barrios et al. (2019) analyzed the skin sensitivity to environmental fungi and the nasal mycobiota of patients with respiratory allergies, isolating *Curvularia* spp. from a large number of nasal samples from patients with asthma and/or rhinitis. Our study showed that the *Curvularia* airborne spores were mostly detected in the atmosphere of Havana during the rainy months. Some authors have also reported that *Curvularia* is part of the common aerobiota in other tropical regions with the high levels recorded during the rainy season. This seasonal incidence has been described in the metropolitan area of Monterrey city (Rocha et al. 2013), Florida (Codina et al. 2008), India (Singh et al. 1987; Roy and Bhattacharya 2016; Dey et al. 2019), Nigeria (Dransfield 1966) and Australia (Upsher and Griffiths 1973). The aforementioned studies proposed that this seasonal incidence was associated probably with new growth of grass. Conversely, Muñoz (1974) noted that *Curvularia* was mainly present in the summer months in Spain, while Menezes et al. (2004) reported a randomly presence of *Curvularia* throughout the year in Fortaleza City, Brazil.

Despite the absence of the correlation with the rainfall, we found an important positive correlation with temperature and relative humidity. These results suggest that in Havana, during the rainy periods (May–

October), the rains can have a washing effect and decrease the daily concentration of *Curvularia* spores. However, as during this season the temperatures are high it can also favor the release of spores or the establishment of conditions of high relative humidity that are related to the amount of *Curvularia* spores in the atmosphere.

Airborne fungal spores detected through direct microscopy sometimes could not be identified to species level, and some are placed in spore types or groups composed by various families, genera or species with similar morphology. According to their distinctive features (conidium shape, pigmentation, number and position of the septa), the spores of *Curvularia* recorded in Havana could be divided in the Boedijn (1933) groups and counted as three identifiable spore types: Lunata, Geniculata and Maculans. The conidia were typically curved and oblong, although some spores were claviform, ellipsoidal, fusiform, obovoid or piriform. The color was characteristically pale brown, to dark brown, often with the cells at the ends lighter than the rest, sometimes with dark bands on the septa (3–4 distoseptate). These observations are similar to conidial morphology reported by some authors (Sivanesan 1987; Grant 1990; Seifert et al. 2011; Manamgoda et al. 2012; Ariyawansa et al. 2015). The aforementioned features are recognizable under microscopic examination of aerobiological glass microscope slides at 400X magnification. Although was not used in the *Curvularia* spore types differentiation, hilum was protuberant in some spores.

The Lunata type included species with the following morphological features: more or less curved conidia, four-celled, with one of the intermediate cells enlarged and darker. Some members of the Lunata type were detected by means of viable method, related to the isolated species *C. aerea*, *C. pallescens*, *C. trifolii* and *C. verruculosa*. These species, identified through their cultivation, were found previously in Cuba in rice plants and grains (Neninger et al. 2003; Estrada and Sandoval 2004). *C. trifolii* has been isolated in Cuba from several host grasses, such as *Rottboellia cochinchinensis*, *Saccharum officinarum* and *Zea mays* (Mena 2004; Camino et al. 2006). These plants are commonly distributed nearby the sampling area, mainly in the agrarian zones located to the south or southwest, reason why this area could be the source of the inoculum. On the other hand, *C. pallescens* is

cosmopolitan and saprobe, while *C. aerea* is a plurivorous species that has been isolated from air, rice and other grasses in Havana and nearby agricultural areas (Mena 2004; Almaguer et al. 2012; Almaguer and Rojas 2013). Species of the other two *Curvularia* types, Geniculata and Maculans, were not detected through cultivation. However, studies conducted by Madrid et al. (2014) reported the Maculans group comprised the species *C. cesatii* (reassigned to *Endophragmiella cesatii* by Hughes, 1979), *C. intermedia*, *C. maculans* (currently classified synonym of *C. eragrostidis*) and *C. spicifera*. In the Geniculata group could be grouped *C. affinis*, *C. falcata* (synonymized by Sivanesan (1987) with *C. senegalensis*), *C. fallax* and *C. geniculata*.

Various species of *Curvularia* have morphological similarities, and to recognize species of this genus based absolutely on conidial features is not an easy task (Tan et al. 2018). This fact reinforces the importance to combine methodologies based on microscopically spores count with culturable sampling methodologies. This strategy of outdoor environments monitoring allows a more complete data on airborne fungal diversity. Fernández-Rodríguez et al. (2014) suggested that simultaneous combination overcomes the own limitations of each procedure. For example, the *C. aerea* isolates produced subcylindrical stromata in culture, which is an additional morphological characteristic for identification. According to Aira et al. (2009) parallel to aerobiological investigation, the corresponding biodiversity study should be taken into account in order to incorporate the relevant local species to a given spore type spectrum. It is indistinguishable if these species are native/introduced or their host source, based exclusively on morphological features. Kakde and Kakde (2012) reported the genus within the most abundant, with a prevalence of *C. lunata* in a market in India related to post-harvest diseases in vegetables. Cuba is a Caribbean tropical archipelago, and the *Curvularia* biological sources could be variable from other latitudes. Therefore, it will be important to combine aerobiological samplings to field studies related to fungi distribution on substrates or analysis of fungal populations. The singularity of our study is that for the first time, in a neotropical urban area of the Caribbean, the temporal variation of *Curvularia* spores in the air and three types of spores identifiable morphologically within this genus is reported, by means of non-viable and

viable sampling methodologies. There are some similar aerobiological investigations directed to an infrageneric level differentiation but considering other spore types and genus. For example, the report about *Cladosporium cladosporioides* and *Cladosporium herbarum* spore types based on conidia wall surface (Aira et al. 2012), four different types of *Coprinus* based on the measure and the morphological features of the spores (Aira et al. 2009) and the recognition of some types of *Leptosphaeria* by Hernández Trejo et al. (2012).

Our study reveals the Lunata spore type was the predominant, followed by Geniculata and Maculans. The viable study supports this result since all species isolated by culturing belonged to the Lunata type. Therefore, its high representation is observed, both by the non-viable method that allows continuous monitoring 24 h a day and by the punctual method of weekly monitoring of cultivable airborne propagules. In addition, previous studies conducted in this same region have reported the prevalence of species in this type but using culture-based methodologies (Almaguer and Rojas 2013). The predominance of Lunata (as spore type or isolated species) is related to the broad representation in substrates and the diversity of species within widely recorded in tropical and sub-tropical climates (Ellis 1971, 1976) and in Cuba (Mena 2004). In other tropical countries such as India, *C. lunata* is described as the most frequent species detected in culture by Arora and Jain (2003) in Bikaner and also at Raipur by Tiwari et al. (2006) and Tiwari (2014).

The Lunata spore type was mainly represented in the atmosphere of Havana during the rainy months (May, September and October), and the highest concentrations were recorded within the same season (August). Inversely, the Geniculata spore type predominated in the dry months (January, February and April) with the most elevated levels recorded in April. The seasonality of Maculans was not clear with maximum fluctuations along the rainy and dry seasons (March, June and November) with the highest levels recorded in August. The spore daily concentration values were much lower in the second year of study, with many concentrations peaks that show a dynamic fluctuation of these conidia in the atmosphere of Havana (being more evident for the Lunata spore type). Some of these peaks were located during the rainy seasons mainly in the year 2017. Roy and

Bhattacharya (2016) reported the same seasonality in two outdoor environments of West Bengal, India, and detected a high prevalence during summer and monsoon season. Youssef et al. (1988) recovered propagules of *Curvularia* for a 12-month period in the atmosphere of Cairo, Egypt, with largest colonies isolated in April due to the detection of *C. lunata*. Rao et al. (2009) also noted the importance of the *Curvularia* spores in the atmosphere of Karachi, Pakistan, but with the prevalence of *C. clavata*. Although this specie was not isolated in culture in the preset study, morphologically belongs to spore type Lunata (Boedijn 1933).

The intradiurnal distribution of the *Curvularia* spores clearly follows a pattern with higher concentrations during the day, with a tendency to a double peak pattern. Rocha Estrada et al. (2013) in Mexico detected the same behavior. The intradiurnal pattern of the Lunata spore type was similar to *Curvularia*, but the double peak was more suitable. However, the pattern of Geniculata spore type similarity was less evident. This could be related to the proportions of the spores and the contribution to the total spores of the genus. The Maculans spore type did not show a marked distinctive pattern in the both years.

The classical studies of Ingold (1965), Gregory (1978) and Lacey (1981) in the past century informed the spore release in fungi and airborne circadian rhythms. Meredith (1962) observed a rapid increase in airborne *Curvularia* spores after dawn culminating in a maximum at or about noon in Jamaica. A similar behavior was detected in the air over paddy fields in India (Sreeramulu and Seshavataram 1962) and Australia (Upsher and Griffiths 1973). Haines et al. (1999) reported *Curvularia* as fungi with midday pattern, which have a maximum level of spores in the daytime of higher wind speed, atmospheric turbulence and temperature. Meredith (1963) described that spores of *C. geniculata* and *C. lunata* were violently discharged into the air during a dry or drying atmosphere. This process was associated with the rapid appearance of gas bubbles in the conidial cells often following some movement of the spore, pivoted at the attachment point. Upsher (1975) referred to some spores were also seen to become detached by twisting of the conidiphores. However, the aforementioned investigation also described airborne *Curvularia* circadian variations as a double peak pattern. Probably this pattern is a modified midday pattern due to the dilution of spore

plumes, since discrete peaks occurred between the ends of the morning and before the noon. Therefore, it can be inferred that the environmental factors influence in a similar way on the spore release mechanisms of the three spore types and it could be reflected in the intradiurnal variation of *Curvularia* spores in Havana.

The distribution pattern was similar to the observed for other airborne conidial genera widely researched, such as *Cladosporium*, *Alternaria*, *Helminthosporium* or *Periconia*, whose spores are dispersed in the atmosphere during the late morning and the evening (Ingold 1965; Gregory 1978; Lacey 1981; Haines et al. 1999; Aira et al. 2013). Intradiurnal fluctuations of these dry spores are related to conidia discharge under environmental circumstances of increasing airflow and decreasing relative air humidity (Stepalska and Wołek, J. 2009). Besides, Haines et al. (1999) reported maximum atmospheric concentration could occur at the same time to peak release for short distances nearby to the source of inoculum. There is evidenced the importance to combine aerobiological samplings to fields studies related to fungi distribution on substrates, taking into consideration the broad representation of *Curvularia* in substrates and the diversity of species within widely recorded in Cuba.

In general, fungal spore release reveals circadian rhythms correlated with meteorological conditions acting locally (Haines et al. 1999; Stepalska and Wołek, J. 2009). Correlations analysis indicated the meteorological conditions of the previous days also might have influenced the formation and release of *Curvularia* spores in Havana since it correlated to daily concentrations. High temperatures, relative humidity, wind speed and precipitation allow the conidia release to the atmosphere. Soo et al. (1998) reported similar significance of these environmental factors on increases of *Curvularia* spore concentrations.

5 Conclusion

The temporal variation in the air of three identifiable morphologically types of *Curvularia* spores is reported for the first time, in a neotropical city of the Caribbean. The most abundant *Curvularia* spore type was Lunata followed by Geniculata and Maculans. Spores of Lunata and Geniculata presented the same intradiurnal distribution patterns, with a tendency to a

double peak pattern, whereas the spores of the Maculans did not show a distinctive pattern. The influence of the main weather variables on spore concentration was evident, in rainy season the spore concentrations was higher in both study years, mainly in 2017. This fact was supported by the correlation test results as significant and positive associations were recorded with temperature, relative humidity and rainfall. Our findings can be a tool to issue alerts in case of an increase on *Curvularia* spore concentrations and thus to prevent the development of diseases in sensitized people in Havana. In addition, it contributes to regulate the phytosanitary treatments application during the moments with higher airborne spore concentrations and is useful to manage the crops located the urban agriculture zones at the peripheral areas of Havana.

Acknowledgements This work was part of the activities of the Cuban National Project: PNH: P211LH007-017 “Caracterización aeromicológica de la atmósfera de La Habana: su impacto en la salud y la agricultura.” The authors thank to Osvaldo Cuesta, Javier Bolufé and Pedro Roura of the Institute of Meteorology (INSMET) for access to meteorological data.

Author contributions M.A., L.D.: Research concept and design, Collection and/or assembly of data, Data analysis and interpretation, Writing the article, Critical revision of the article, Final approval of article., M.F.-G.: Data analysis and interpretation, Writing the article, Critical revision of the article, Final approval of article, and S.S.: Collection and/or assembly of data.

Funding This investigation was part of the Cuban National Project P211LH007-017 “Caracterización aeromicológica de la atmósfera de La Habana: su impacto en la salud y la agricultura”.

Compliance with ethical standards

Conflicts of interest The authors declare that they have no conflict of interest.

References

- Agarwal, M. K., Jones, R. T., & Yunginger, J. W. (1982). Shared allergenic and antigenic determinants in *Alternaria* and *Stemphylium* extracts. *J Allergy Clin Immunol*, 70, 437–444.
- Agashe, S. N., & Anand, P. (1982). Immediate type hypersensitivity to common pollen and molds in Bangalore city. *Aspects Allergy Appl Immunol*, 15, 49–52.

- Aira, M. J., Rodríguez-Rajo, F. J., Castro, M., & Jato, V. (2009). Characterization of *Coprinus* spores in the NW of the Iberian Peninsula Identification and count in aerobiological samples. *Cryptogamie. Mycologie*, 30(1), 57–66.
- Aira, M. J., Rodríguez-Rajo, F. J., Fernández-González, M., Seijo, C., Elvira-Rendueles, B., Abreu, I., & Tormo, R. (2013). Spatial and temporal distribution of *Alternaria* spores in the Iberian Peninsula atmosphere and meteorological relationships. *International Journal of Biometeorology*, 57(2), 265–274. <https://doi.org/10.1007/s00484-012-0550-x>
- Aira, M. J., Rodríguez-Rajo, F. J., Fernández-González, M., Seijo, C., Elvira-Rendueles, B., Gutiérrez-Bustillo, M., et al. (2012). *Cladosporium* airborne spore incidence in the environmental quality of the Iberian Peninsula. *Grana*, 51(4), 293–304. <https://doi.org/10.1080/00173134.2012.717636>
- Almaguer, M., Aira, M. J., Rodríguez-Rajo, F. J., & Rojas, T. I. (2013). Study of airborne fungus spores by viable and non-viable methods in Havana. Cuba. *Grana*, 52(4), 289–298. <https://doi.org/10.1080/00173134.2013.829869>
- Almaguer, M., Aira, M. J., Rodríguez-Rajo, F. J., & Rojas, T. I. (2015). Thirty four identifiable airborne fungal spores at Havana. Cuba. *Annals Agric Environ Med*, 22(2), 220–225. <https://doi.org/10.5604/12321966.1152068>
- Almaguer, M., & Rojas, T. I. (2013). Aeromicota viable de la atmósfera de La Habana Cuba. *Nova Acta Cientifica Compostelana*, 20, 35–45.
- Almaguer, M., Rojas, T. I., Rodríguez-Rajo, F. J., & Aira, M. J. (2012). Airborne fungal succession in a rice field of Cuba. *European Journal of Plant Pathology*, 133(2), 473–482. <https://doi.org/10.1007/s10658-011-9921-0>
- Álvarez, M., Castro, R., Leyva, Y., Lopez, B., Rodríguez, J., Rojas, T., Piera, O., & Labrada, A. (2020). Sensibilización a hongos anemófilos en trabajadores del Archivo y Biblioteca de la Universidad de La Habana. *Archivos del Hospital Universitario "General Calixto García"*, 8(2), 159–172.
- Ariyawansa, H. A., Thambugala, K. M., Manamgoda, D. S., Jayawardena, R., Camporesi, E., Boonmee, S., et al. (2015). Towards a natural classification and backbone tree for Pleosporaceae. *Fung Divers*, 71, 85–139. <https://doi.org/10.1007/s13225-015-0323-z>
- Arnold, G. R., Guerra, A. G., & Rodríguez de La Rosa, N. (1987). Presencia de hongos del aire del INIFAT. *Reporte de Investigación del Instituto de Investigaciones Fundamentales en Agricultura Tropical*, 43, 1–6.
- Arora, A., & Jain, V. K. (2003). Fungal air spora of Bikaner. *Indian j aerobiol*, 16(1), 1–9.
- Barrios, O. H., Rodríguez, I. P., Heredia, L. O., Torres, N. R., Vidal, L. G., Ayala, Y. M., & Luna, L. L. (2019). Sensibilidad cutánea a hongos ambientales y estudio de la microbiota nasal de pacientes con alergias respiratoria. *Investigaciones Médicoquirúrgicas*, 11(2), 1–10.
- Boedijn, K. B. (1933). Über einige phragmosporen Dematiazen. *Bulletin du Jardin Botanique de Buitenzorg*, 13, 120–134.
- Camino M, Mena J, Minter D (2006) Fungi of Cuba. www.cybertruffle.org.uk/cubafung
- Chen, B., Jasmine, C., Wu, C., Kim, H., Honda, Y., & Leon, Y. (2014). High ambient *Cladosporium* spores were associated with reduced lung function in schoolchildren in a longitudinal study. *Science of the Total Environment*, 481, 370–376. <https://doi.org/10.1016/j.scitotenv.2014.01.078>
- Chew, F. T., Lim, S. H., Shang, H. S., Dahlia, D. S., Goh, D. Y. T., Lee, B. W., et al. (2000). Evaluation of the allergenicity of tropical pollen and airborne spores in Singapore. *Allergy*, 55, 340–347. <https://doi.org/10.1034/j.1398-9995.2000.00308.x>
- Codina, R., Fox, R. W., Lockey, R. F., De Marco, P., & Bagg, A. (2008). Typical levels of airborne fungal spores in houses without obvious moisture problems during a rainy season in Florida. *J Invest Allergol Clin Immunol*, 18, 156–162.
- Da Cunha K (2014) Phenotypic and molecular characterization of clinical isolates of bipolaris, curvularia, exserohilum and pithomyces (Doctoral dissertation, Universitat Rovira i Virgili). <https://www.tdx.cat/handle/10803/279271#page=1>
- Dey, D., Ghosa, K., & Bhattacharya, S. G. (2019). Aerial fungal spectrum of Kolkata. India. along with their allergenic impact on the public health: a quantitative and qualitative evaluation. *Aerobiologia*, 35(1), 15–25. <https://doi.org/10.1007/s10453-018-9534-6>
- Dransfield, M. (1966). The fungal air spora at Samaru, northern Nigeria. *Trans British Mycol Soc*, 41(1), 121–132.
- Ellis, M. B. (1971). *Dematiaceous Hyphomycetes*. CMI: Commonwealth Mycological Institute. Kew. Survey. England.
- Ellis, M. B. (1976). *More Dematiaceous Hyphomycetes*. England: Commonwealth Mycological Institute. Kew.
- Esch, R. E., & Codina, R. (2017). Fungal raw materials used to produce allergen extracts. *Annals Allergy, Asthma Immunol*, 118(4), 399–405. <https://doi.org/10.1016/j.anai.2016.05.012>
- Estrada, G., & Sandoval, I. (2004). Patogenicidad de especies de *Curvularia* en arroz. *Fitosanidad*, 8(4), 23–26.
- Fernández-Rodríguez, S., Tormo-Molina, R., Maya-Manzano, J. M., Silva-Palacios, I., & Gonzalo-Garjón, Á. (2014). Outdoor airborne fungi captured by viable and non-viable methods. *Fungal Ecol*, 7, 16–26. <https://doi.org/10.1016/j.funeco.2013.11.004>
- Galán, C., González, P. C., Teno, P. A., & Vilches, E. D. (2007). *Manual de Calidad y Gestión de la Red Española de Aerobiología*. España: Servicio de Publicaciones. Universidad de Córdoba.
- Grant, E. (1990). *Sampling and identifying allergenic pollens and molds*. USA: Blewstone Press. San Antonio.
- Gregory, P. H. (1978). Distribution of airborne pollen and spores and their long distance transport. *Pure and Applied Geophysics*, 116(2–3), 309–315.
- Gupta, R., Singh, B. P., Sridhara, S., Gaur, S. N., Kumar, R., Chaudhary, V. K., & Arora, N. (2002). Allergenic cross-reactivity of *Curvularia lunata* with other airborne fungal species. *Allergy*, 57, 636–640. <https://doi.org/10.1034/j.1398-9995.2002.03331.x>
- Haines, J., Escamilla, B., Muilenberg, M., Gallup, J., & Levett, E. (1999). *Mycology of the air: a workshop manual for sampling and identifying airborne fungus spores*. New York State Museum, New York: Pan-American Aerobiology Association Tucson AZ.
- Hernández Trejo, F. H., Rodríguez, A. F. M., Molina, R. T., & Palacios, I. S. (2012). Airborne ascospores in Mérida (SW Spain) and the effect of rain and other meteorological

- parameters on their concentration. *Aerobiologia*, 28(1), 13–26. <https://doi.org/10.1007/s10453-011-9207-1>
- Hirst, J. M. (1952). An automatic volumetric spore trap. *Annals Appl Biol*, 39(2), 257–265.
- Hughes, S. J. (1979). *Fungi Can.*, 150, 1–2.
- Hyde, K. D., Norphanphou, C., Abreu, V. P., et al. (2017). Fungal diversity notes 603–708: taxonomic and phylogenetic notes on genera and species. *Fungal Divers*, 87, 1–235. <https://doi.org/10.1007/s13225-017-0391-3>
- Ingold, C. T. (1965). *Spore liberation*. Oxford: Clarendon Press, Oxford.
- Kakde, U., & Kakde, H. (2012). Incidence of post-harvest disease and airborne fungal spores in a vegetable market. *Acta Botanica Croatica*, 71(1), 147–157.
- Lacey, J. (1981). The aerobiology of conidial fungi. In G. T. Cole & B. Kendrick (Eds.), *Biology of conidial fungi* (pp. 373–416). New York: Academic Press, INC.
- LarenasLinnemann, D., Baxi, S., Phipatanakul, W., Portnoy, J. M., Barnes, C., Grimes, C., & Scott, J. (2016). Clinical evaluation and management of patients with suspected fungus sensitivity. *J Allergy Clin Immunol Pract*, 4(3), 405–414. <https://doi.org/10.1016/j.jaip.2015.10.015>
- Levetin, E., Horner, W. E., Scott, J. A., & Workgroup, E. A. (2016). Taxonomy of allergenic fungi. *J Allergy Clin Immunol In Pract*, 4(3), 375–385.
- Madrid, H., da Cunha, K. C., Gené, J., Dijksterhuis, J., Cano, J., Sutton, D. A., et al. (2014). Novel *Curvularia* species from clinical specimens. *Persoonia*, 33, 48–60.
- Manamgoda, D. S., Cai, L., McKenzie, E. H. C., Crous, P. W., Madrid, H., Chuksatirote, E., et al. (2012). A phylogenetic and taxonomic re-evaluation of the *Bipolaris-Cochliobolus-Curvularia* complex. *Fungal Divers*, 56, 131–144. <https://doi.org/10.1007/s13225-012-0189-2>
- Manamgoda, D. S., Rossman, A. Y., Castlebury, L. A., Chuksatirote, E., & Hyde, K. D. (2015). A taxonomic and phylogenetic re-appraisal of the genus *Curvularia* (Pleosporaceae) human and plant pathogens. *Phytotaxa*, 212(3), 175–198.
- Marin-Felix, Y., Groenewald, J. Z., Cai, L., Chen, Q., Marincowitz, S., Barnes, I., et al. (2017). Genera of phytopathogenic fungi: GOPHY 1. *Studies in Mycology*, 86, 99–216. <https://doi.org/10.1016/j.simyco.2017.04.002>
- Martínez, B., Pérez, S., & Plana, R. (2009). Principal fungal diseases in Triticales and wheat in San José de las Lajas—Havana province. *Revista de Protección Vegetal*, 24(3), 195.
- McAleer, R., Kroenert, D. B., Elder, J. L., & Froudast, J. H. (1981). Allergic bronchopulmonary disease caused by *Curvularia lunata* and *Drechslera hawaiiensis*. *Thorax*, 36, 338–344.
- Mena J (2004) Taxonomía del complejo *Bipolaris. Curvularia. Drechslera* y *Exserohilum* en Cuba. Phd Thesis. Instituto de Ecología y Sistemática. CITMA. La Habana. Cuba
- Menezes, E. A., Trindade, E. C. P., Costa, M. M., Freire, C. C. F., Cavalcante, M. S., & Cunha, F. A. (2004). Airborne fungi isolated from Fortaleza city state of Ceará Brazil. *Revista do Instituto de Medicina Tropical de São Paulo S Paulo*, 46(3), 133–137.
- Meredith, D. S. (1962). Some components of the air spora in Jamaican banana plantations. *Annals Appl Biol*, 50, 577–594. <https://doi.org/10.1111/j.1744-7348.1962.tb06050.x>
- Meredith, D. S. (1963). Violent spora release in some Fungi Imperfecti. *Ann Bot N S*, 27(105), 39–47.
- Muñoz, E. (1974). *Mapa micológico de España*. L Cheminova: Española S.A.
- Navpreet, K. G., Kumar, K., & Navneet, K. (2017). Risk assessment of outdoor airborne biological allergens in India: a review. *J Entomol Zool Stud*, 5(4), 1069–1075.
- Neninger, L. H., Hidalgo, E., Barrios, L., & Pueyo, M. (2003). Hongos presentes en semillas de arroz (*Oryza sativa* L.) en Cuba. *Fitosanidad*, 7(3), 7–11.
- Nilsson, S., & Person, S. (1981). Three pollen spectra in the Stock- holm region (Sweden). 1973–1980. *Grana*, 20, 179–182.
- Orberá, T., Giro, Z. G., Serrat Díaz, M., & Rodríguez, S. (2012). In vitro growth inhibition of *Curvularia gudauskasii* by *Bacillus subtilis*. *Annals Microbiol*, 62, 545–551. <https://doi.org/10.1007/s13213-011-0290-x>
- Prasla, I., Duman, K., Ciochetto, Z., Burman, A., Mahon, A., Park, S., & Bassford, J. (2013). Significant heterogeneity in airborne mold quantities on the caribbean island Of St kitts health implications and impact on food preservation. *Virol Mycol*, 3(1), 1–5.
- Quintero JM (1964) Atmospheric fungi. PhD Thesis. University of Puerto Rico
- Rao, T. A., Shaikh, A. H., & Ahmed, M. (2009). Airborne fungal flora of Karachi. *Pakistan Journal of Botany*, 41(3), 1421–1428.
- Rocha Estrada, A., Alvarado Vázquez, M. A., Gutiérrez Reyes, R., Salcedo Martínez, S. M., & Moreno Limón, S. (2013). Variación temporal de esporas de *Alternaria*, *Cladosporium*, *Coprinus*, *Curvularia* y *Venturia* en el aire del área metropolitana de monterrey nuevo león México. *Revista Internacional de Contaminación Ambiental*, 29(2), 155–165.
- Rojas, T. I., Llanes, N., Benítez, M., Aira, M. J., & Malagón, H. (2007). El género *Aspergillus* en la atmósfera de La Habana (Cuba). *Boletín Micológico*, 22, 41–46.
- Roy, S., & Bhattacharya, K. (2016). Seasonal variation of airborne fungal load in two outdoor environments of Farakka West Bengal India. *Journal of Palynology*, 52, 27–36.
- Sánchez, K. C., Almaguer, M., Pérez, I., Flores, T. I., & Aira, M. J. (2019). Diversidad fúngica en la atmósfera de La Habana (Cuba) durante tres períodos poco lluviosos. *Revista Internacional de Contaminación Ambiental*, 35(1), 137–150.
- Seifert, K., Morgan-Jones, G., Gams, W., & Kendrick, B. (2011). *The genera of hyphomycetes*. The Netherlands: CBS-KNAW Fungal Biodiversity Centre. Utrecht.
- Singh, B. P., Singh, A. B., Nair, P. K., & Gangal, S. V. (1987). Survey of airborne pollen and fungal spores at Dehra Dun. *India. Ann. Allergy*, 59, 229–234.
- Singh, V. (2019). Fungal rhinosinusitis: unravelling the disease spectrum. *Journal of Oral and Maxillofacial Surgery*, 18(2), 164–179.
- Sivanesan, A. (1987). Graminicolous species of *Bipolaris*, *Curvularia*, *Drechslera*. *Exserohilum teleomorphs*. *Myc. Papers*, 158, 1–261.

- Soo, H. L., Chew, F., Dahlia, B. M. D., Tiang, W. T., Wah, B., & Koon, T. T. (1998). Outdoor airborne fungal spores in Singapore. *Grana*, 37(4), 246–252.
- Sreeramulu, T., & Seshavatham, V. (1962). Spore content of the air over paddy fields. I. Changes in a field near Pentapadur from 21st September to 31st December 1957. *Indian Phytopathol*, 15, 61–74.
- Stepalska, D., & Wołek, J. (2009). Intradurnal periodicity of fungal spore concentrations (*Alternaria*, *Botrytis*, *Cladosporium*, *Didymella*, *Ganoderma*) in Cracow. *Poland. Aerobiologia*, 25(4), 333–340. <https://doi.org/10.1007/s10453-009-9137-3>
- Stewart, G. A., & Robinson, C. (2017). Indoor and Outdoor Allergens and Pollutants. In S. T. Holgatem & A. Sheik (Eds.), *O’HHeir R* (pp. 73–116). Netherlandsm (pp: Middleton’s Allergy Essentials. Elsevier.
- Tan, Y. P., Crous, P. W., & Shivas, R. G. (2018). Cryptic species of *Curvularia* in the culture collection of the queensland plant pathology herbarium. *MycKeys*, 35, 1–25. <https://doi.org/10.3897/mycokeys.35.25665>
- Tiwari, K. L. (2014). Study of Airspora during celebration of Rajyotsava. *B F A I J*, 6(1), 53–56.
- Tiwari, K. L., Jadhav, S. K., & Kunjam, S. (2006). Aeromycoflora of slum area of Raipur. *Adv plant sci*, 19(2), 387–390.
- Upsher, F. J. (1975). Fungal biological flora 3 *Curvularia* Boedijn. *Int. Biodet. Bull*, 11(1), 24–30.
- Upsher, F. J., & Griffiths, D. A. (1973). The air spora at a site in tropical Queensland, Australia. *Trans British Mycol Soc*, 61(3), 537–544.
- Vázquez, L. L., & Fernández, E. (2007). Manejo agroecológico de plagas y enfermedades en la agricultura urbana. Estudio de caso Ciudad de La Habana. *Cuba. Agroecología*, 2, 1–11.
- Williams, P. B., Barner, Ch. S., & Portnoy, J. M. (2016). Innate and adaptative immune response to fungal products and allergens. *J Allergy Clin Immunol Pract*, 4(3), 386–395.
- Wu, Y. H., Chan, C. C., Rao, C. Y., Lee, C. T., Hsu, H., Chiu, Y. H., & Chao, H. J. (2007). Characteristics, determinants and spatial variations of ambient fungal levels in the subtropical Taipei metropolis. *Atmospheric Environment*, 41(12), 2500–2509. <https://doi.org/10.1016/j.atmosenv.2006.11.035>
- Youssef, A., Youssef, D., & Karam El-din, A. (1988). Airborne spores of opportunistic fungi in the atmosphere of Cairo. *Egypt. Grana*, 27(1), 89–92.