

# Influence of Climatic Parameters on the Seasonal Dynamics of *Cladosporium* Spores in Three Agricultural Areas of Madagascar: Ambatondrazaka, Amparafaravola and Betafo

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**Abstract:** *This study analyzes the influence of climatic parameters on the dynamics of Cladosporium spores in Ambatondrazaka, Amparafaravola, and Betafo (Madagascar). Aeropalynological monitoring (April 1st – September 30th) was conducted using Hirst-type volumetric traps. Identification, based on conidial morphology, allowed for the calculation of the Seasonal Fungus Index (SFI) and daily concentrations.*

*Peak concentrations occurred in July and August, reaching maximum values of 2,295 spores m<sup>-3</sup> in Ambatondrazaka, 206 spores m<sup>-3</sup> in Betafo, and 139 spores m<sup>-3</sup> in Amparafaravola. The sensitization threshold (> 100 spores m<sup>-3</sup>) was frequently exceeded, particularly in Ambatondrazaka where it was surpassed for 58 days, highlighting a significant allergenic risk. Statistical analysis shows that temperature is the primary controlling factor ( $r = 0.795$ ;  $p < 0.05$ ), while precipitation exerts a wash-out effect on atmospheric concentrations. These findings are essential for developing an aeroallergens early warning system in Madagascar..*

**Keywords:** *Cladosporium · Aerobiology · Climatic parameters · Seasonal dynamics · Aeroallergen · Biological pollution*

## I. INTRODUCTION

The genus *Cladosporium* Nees ex Wallroth belongs to the phylum Ascomycota ([www.mycobank.org](http://www.mycobank.org)) and includes cosmopolitan fungi of the Dothideomycetes class, widely present in both natural and agricultural environments. These species are characterized by a high capacity to adapt to environmental conditions and represent a major component of the bioaerosphere. Their development is favored by moderate temperatures, with an optimum generally ranging between 22 and 28°C, while temperatures near 0°C severely limit growth and spore production (Hjelmroos 1993). Under experimental conditions, sporulation is observed between 8 and 24°C, and conidia maturation occurs less than 24 hours after formation (Mamgain et al. 2013).

*Cladosporium* spores are among the primary fungal aeroallergens in the atmosphere. Their inhalation is frequently associated with the onset of respiratory allergic manifestations, particularly in atopic individuals. Symptoms can appear once concentrations exceed 80 spores m<sup>-3</sup>, while levels above 300 spores m<sup>-3</sup> are generally linked to severe asthma attacks (Gravesen 1979; Black et al. 2000; Downs et al. 2001; Rapiejko et al. 2004).



The seasonal dynamics of *Cladosporium* spores are strongly influenced by climatic conditions. In temperate regions, concentrations increase progressively in the spring, reach a maximum in summer, and decrease in autumn (Hjelmroos 1993; Nikkels et al. 1996; Stepalska et al. 1999; Kasprzyk et al. 2004). In Mediterranean regions, bimodal profiles have been described, with successive peaks at the beginning and end of the hot season (Celenk et al. 2007; Recio et al. 2012; Aira et al. 2013). These variations reflect the combined influence of climatic factors, plant phenology, and agricultural practices.

Among meteorological parameters, air temperature, relative humidity, precipitation, sunshine, and wind speed play a decisive role in the processes of sporulation, release, and dispersion of spores. Numerous studies have highlighted positive correlations with temperature and solar radiation, and negative correlations with high humidity and precipitation (Grinn-Gofron & Rapiejko 2009; Oliveira et al. 2009; Escuredo et al. 2011; Sabariego et al. 2012). Intense rainy episodes or prolonged periods of high humidity can temporarily limit atmospheric concentrations, while hot and dry conditions favor their increase (Hjelmroos 1993; Peternel et al. 2004; Recio et al. 2012).

In Madagascar, agricultural zones exhibit great bioclimatic diversity due to altitudinal gradients and regional contrasts. The regions of Ambatondrazaka and Amparafaravola, located in the Alaotra basin, are characterized by a subhumid tropical climate favorable to irrigated crops, while the Betafo region, located in the Central Highlands, features a cooler and more contrasted high-altitude tropical climate. These climatic differences, associated with distinct agricultural systems, provide a particularly suitable framework for studying the influence of meteorological parameters on the dynamics of fungal spores.

Despite the importance of these factors, the relationships between climatic conditions and *Cladosporium* spore concentrations remain poorly documented in tropical agricultural zones, and no study to date has comparatively analyzed these interactions in the main agricultural basins of Madagascar.

The objective of the present study is therefore to analyze the influence of temperature, precipitation, relative humidity, and wind speed on the seasonal dynamics of *Cladosporium* spores in three agricultural areas of Madagascar—Ambatondrazaka, Amparafaravola, and Betafo—in order to identify the dominant climatic parameters controlling daily and seasonal fluctuations in airborne concentrations.

## **II. MATERIALS AND METHODS**

### **II.1. Geographical and Bioclimatic Description of the Sites**

Aeropalynological monitoring was conducted in three contrasting agricultural areas of Madagascar: Ambatondrazaka, Amparafaravola, and Betafo (Fig. 1). These sites are distributed along a marked latitudinal and altitudinal gradient. Amparafaravola (to the North) and Betafo (to the South) are the most distant locations, separated by approximately 270 km.

In terms of topography and climate, the study compares two distinct ecosystems:

- The Lake Alaotra Basin (Ambatondrazaka and Amparafaravola): Situated at an average altitude of 750 m, this area benefits from a subhumid tropical lowland climate. It is characterized by high average annual temperatures (approximately 23.5°C to 23.8°C) and a long growing season (230 to 240 days).
- The Central Highlands (Betafo): Located at an altitude of approximately 1,400 m, this site presents a significantly cooler high-altitude tropical climate. The average annual temperature is about 18.2°C, representing a thermal difference of more than 5°C compared to the lowland sites, which reduces the growing season to approximately 200 days.

These thermal and rainfall gradients (Table 1) directly influence the phenology of local crops (irrigated rice farming in Alaotra versus high-altitude polyculture in Betafo) and, consequently, the fungal sporulation dynamics of *Cladosporium* (Hjelmroos, 1993).



## II.2. Aerobiological Monitoring

Monitoring was conducted from April 1st to September 30th, 2023. Spore sampling was performed using Hirst-type volumetric traps (Lanzoni, Italy), installed in open agricultural environments at a height of 18 to 22 meters above the ground to ensure the representativeness of the atmospheric air.

The standardized protocol was applied as follows:

- Sampling: Air is continuously aspirated at a flow rate of 10 L min<sup>-1</sup>. Particles are impacted onto an adhesive tape (Melinex) fixed to a rotating drum moving at a speed of 2 mm h<sup>-1</sup>.
- Preparation: Each week, the tape is cut into seven 48 mm segments, each representing 24 hours of sampling.
- Analysis: The segments are mounted on glass slides and examined under an optical microscope (×400 magnification) according to the method described by Frenguelli (2003).

## II.3. Identification and Analysis Parameters

Spores of the genus *Cladosporium* were identified based on their morphological characteristics (club-like shape, dark pigmentation, and the presence of transverse and longitudinal septa). The data were processed to determine:

- The average daily concentration (spores m<sup>-3</sup>).
- The Seasonal Fungus Index (SFI), corresponding to the total cumulative sum of spores over the study period.

## II.4. Statistical Analysis

The normality of the distributions was verified using the Shapiro–Wilk test. Due to the non-normal distribution of the concentration data, spatial comparisons between the three sites were performed using the non-parametric Kruskal–Wallis test, followed by Dunn’s post-hoc test.

The relationships between spore concentrations and meteorological parameters (temperature, precipitation, humidity, and wind) were analyzed using correlation coefficients ( $p \leq 0.05$ ).

Fig. 1: The location of three monitoring sites in central and eastern Madagascar used in this study



Table 1 : Climatic and environmental characteristics of the aeropolynological monitoring sites in Madagascar

Monitoring site	Trap height above ground (m)	Geographic coordinates	Site altitude (m)	Climate type	Mean annual temperature (°C)	Mean annual rainfall (mm)
Ambatondrazaka	20	S 17° 50' / E 48° 26'	750	Humid tropical lowland	23.8	1,250
Amparafaravola	18	S 17° 35' / E	760	Sub-humid	23.5	1,200



		48° 13'		tropical		
Betafo	22	S 19° 50' / E 46° 52'	1,400	Tropical high-altitude (cooler)	18.2	1,300

### II.5. Statistical Analysis – Climate–Spore Relationships

The Shapiro–Wilk test showed that the spore concentrations were not normally distributed, and the Brown–Forsythe test demonstrated that the variances were not homogeneous. Therefore, non-parametric tests were used for subsequent statistical analyses (Shapiro et al., 1968; Brown & Forsythe, 1974).

The strength and direction of the correlations between daily meteorological parameters and daily spore concentrations were evaluated using Spearman’s correlation coefficients. Several parameters were included in this analysis:

- Temperature (T, °C)
- Relative air humidity (H, %)
- Precipitation (PP, mm)
- Wind speed (WS, m s<sup>-1</sup>)

Statistical hypotheses were tested with a significance threshold of  $p \leq 0.05$ .



Fig 2 : Cladosporium spores: 1 : light microscope; 2 : scanning microscope

## III. RESULTS

### III.1. Seasonal dynamics and spore concentrations

Aeropalynological monitoring conducted from 1 April to 30 September 2023 showed that seasonal spore peaks were observed mainly in July and August. In Ambatondrazaka, a rapid increase was noted from mid-July, peaking in August before a sharp decline. In Amparafaravola, the seasonal pattern was bimodal, with a marked decrease in August followed by a second, lower peak in September. In Betafo, the dynamics showed strong instability after the peak, with irregular fluctuations.

The maximum concentrations recorded were 2,295 spores m<sup>-3</sup> in Ambatondrazaka (2 August), 139 spores m<sup>-3</sup> in Amparafaravola (15 August), and 206 spores m<sup>-3</sup> in Betafo (10 August).

Table 2– Seasonal dynamics of airborne Cladosporium spores in three agricultural areas of Madagascar

Site	June	July	August	September	Max spores/m <sup>3</sup>	Day of maximum	No. days $\geq$ 80 spores/m <sup>3</sup>
Ambatondrazaka	949	11,662	19,337	1,883	2,295	2 August	58



Amparafaravola	26	608	1,037	126	139	15 August	8
Betafo	67	731	2,358	69	206	10 August	13

From a public health perspective, the threshold of 80 spores  $m^{-3}$  was exceeded for 58 days in Ambatondrazaka, 13 days in Betafo, and 8 days in Amparafaravola. Concentrations exceeding 300 spores  $m^{-3}$ , which are associated with severe health effects, were recorded for up to 29 days in the most contaminated areas.

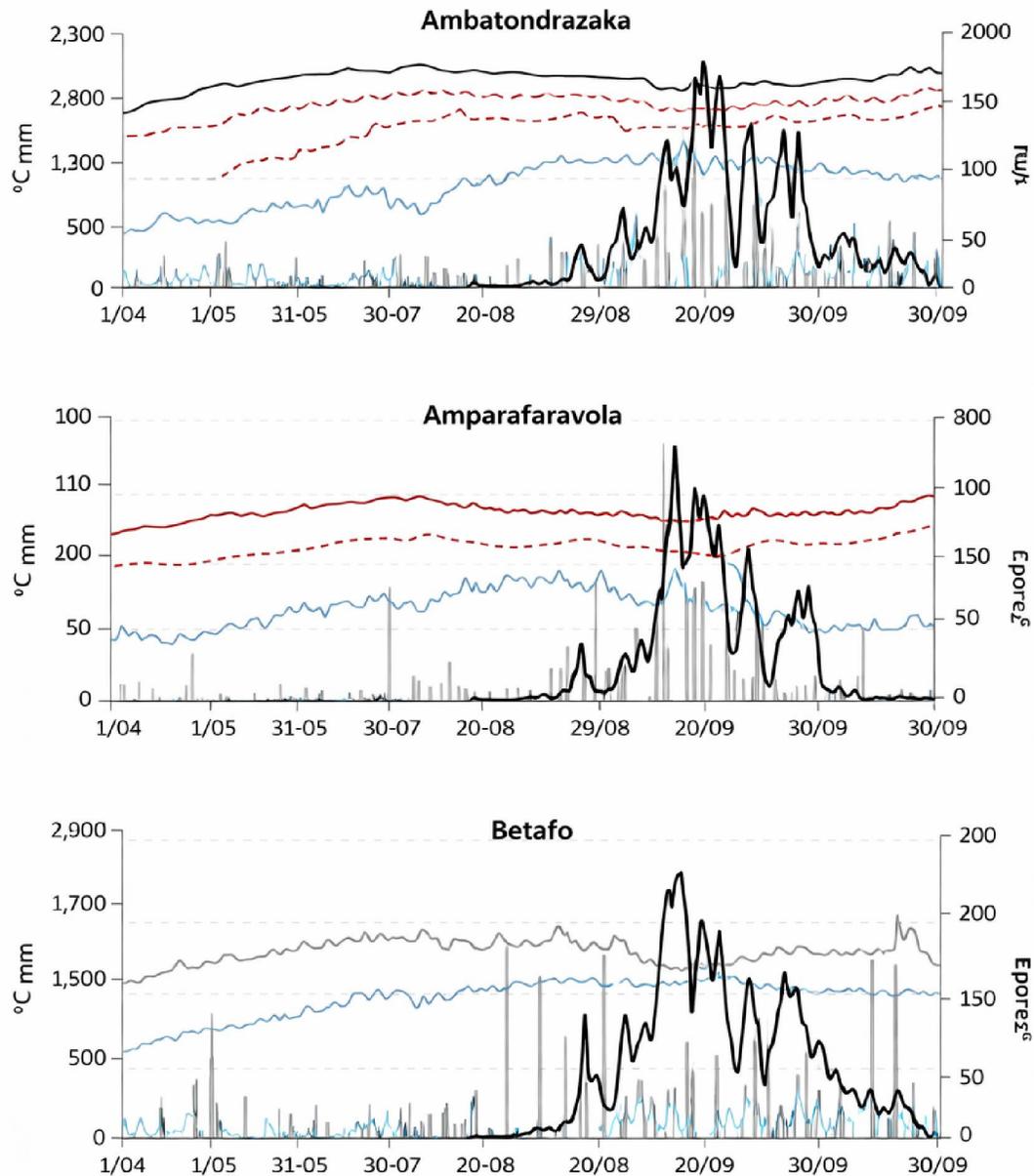


Fig. 3. Daily variations in Cladosporium spore concentrations (black line) and climatic parameters (mean temperature, minimum temperature, precipitation, humidity, and wind) in the air of three agricultural areas of Madagascar (Ambatondrazaka, Amparafaravola, and Betafo).



### III.2. Influence of climatic parameters

Statistical analysis (Shapiro–Wilk and Brown–Forsythe tests) led to the use of non-parametric methods (Spearman correlation).

- Thermal factor : Temperature is the main driver of the increase in spore concentrations.
  - o In Ambatondrazaka, mean temperature is the dominant factor ( $r = 0.795$ ).
  - o In Amparafaravola, minimum temperature controls sporulation ( $r = 0.795$ ).
  - o In Betafo, the influence is more moderate, with maximum temperature ( $r = 0.442$ ) and mean temperature ( $r = 0.376$ ).
- Humidity, precipitation, and wind: These parameters generally show negative correlations.
  - o In Amparafaravola, humidity ( $r = -0.290$ ) and precipitation ( $r = -0.404$ ) significantly limit spore presence. Only heavy rainfall causes a sharp decrease.
  - o Wind plays a secondary or marginal role in the dispersion or dilution of spore concentrations in Amparafaravola ( $r = -0.232$ ), as well as in Ambatondrazaka and Betafo.

Table 3 – Correlations between daily Cladosporium spore concentrations and climatic parameters in three agricultural areas of Madagascar

Site	Mean temperature (r)	Minimum temperature (r)	Maximum temperature (r)	Relative humidity (r)	Precipitation (r)	Wind speed (r)
Ambatondrazaka	0.795	NS	NS	NS	NS	NS
Amparafaravola	NS	0.795	NS	-0.290	-0.404	-0.232
Betafo	0.376	NS	0.442	NS	NS	NS

Note: NS = not statistically significant correlation ( $p > 0.05$ )



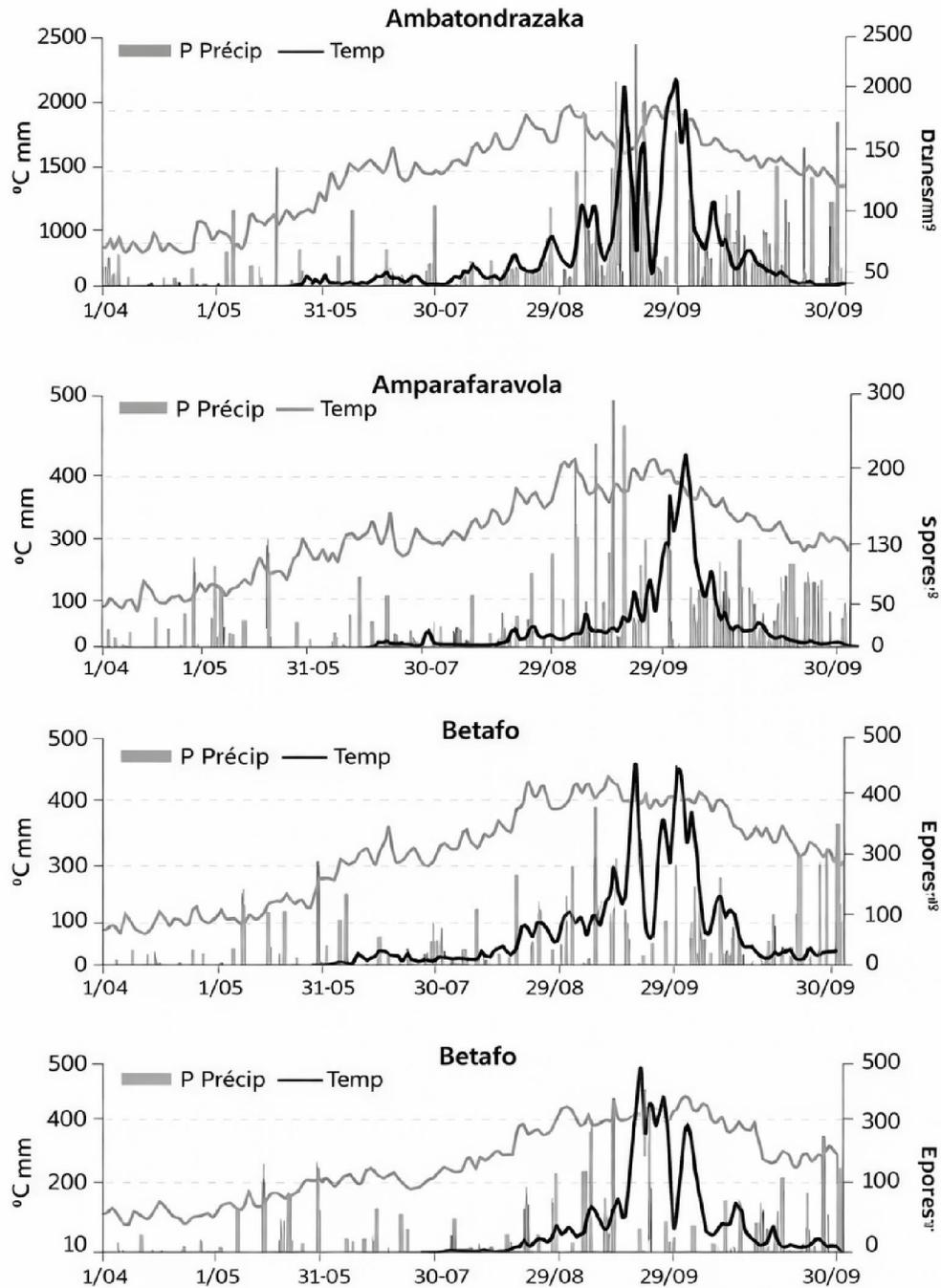


Fig. 4. Patterns of precipitation (PP, bars), mean temperature (grey line), and daily Cladosporium spore concentrations (black line) in the air of three agricultural areas of Madagascar in 2023 (Ambatondrazaka, Amparafaravola, and Betafo).



#### **IV. DISCUSSION**

The results highlight the predominant role of temperature in regulating *Cladosporium* spore concentrations in Madagascar. The strong positive correlations observed in Ambatondrazaka and Amparafaravola confirm that rising temperatures promote fungal development and spore release, in agreement with previous European studies. However, clear geographical disparities emerge:

- Lowland areas (Ambatondrazaka and Amparafaravola): Located at approximately 750 m above sea level and characterized by a tropical subhumid climate, these sites show a strong dependence on thermal conditions.
- High-altitude area (Betafo): At 1,400 m above sea level, the cooler climate appears to attenuate the direct impact of temperature.

The absence of a dominant controlling parameter suggests the influence of local factors such as specific agricultural practices, altitude, and microclimatic conditions. The negative influence of precipitation and humidity, particularly pronounced in Amparafaravola, can be explained by atmospheric washout and the limitation of spore release from wet vegetative surfaces.

As for wind, its moderate effect suggests that mechanical dispersion is often counterbalanced by atmospheric dilution. The July–August period represents a critical phase for public health risk. The high number of days exceeding the allergenic threshold—especially in Ambatondrazaka (58 days)—underscores the importance of integrating climatic parameters into early warning systems to prevent allergic diseases in tropical agricultural environments.

##### **IV.1. Predominance of the thermal factor**

Temperature proved to be the main driver of increased *Cladosporium* spore concentrations across all sites, with the notable exception of Betafo. However, the strength of this relationship varies according to geographical location. In Ambatondrazaka, mean temperature exerts the strongest influence, with a high correlation coefficient ( $r = 0.795$ ;  $p < 0.05$ ), indicating a decisive role of this parameter in regulating atmospheric concentrations. In Amparafaravola, minimum temperature appears to be the primary factor controlling sporulation, also with a high correlation coefficient ( $r = 0.795$ ;  $p < 0.05$ ), suggesting a particular sensitivity of spore emissions to nocturnal thermal conditions. In Betafo, the thermal influence is clearly more moderate. The coefficients observed for maximum temperature ( $r = 0.442$ ) and mean temperature ( $r = 0.376$ ) remain low to moderate, reflecting a weaker control of temperature over spore dynamics at this high-altitude site.

##### **IV.2. Role of humidity, precipitation, and wind**

In contrast to temperature, the other climatic parameters mainly exhibit negative correlations with spore concentrations, indicating washout or dispersal effects of particles in the atmosphere.

In Amparafaravola, relative humidity ( $r = -0.290$ ) and precipitation ( $r = -0.404$ ) significantly limit the presence of spores in the air. Only heavy rainfall leads to a sharp decrease in concentrations. Wind speed also shows a weak negative correlation ( $r = -0.232$ ), suggesting a secondary role in spore dispersion. In Ambatondrazaka and Betafo, the influence of wind remains marginal. It acts only in a limited manner on reducing concentrations during episodes of maximum gusts and does not constitute a dominant controlling factor.

##### **IV.3. Seasonal dynamics and concentration peaks**

The sporulation calendar closely follows seasonal variations, with concentration peaks mainly observed in July and August. In Ambatondrazaka, a rapid increase in concentrations is observed from mid-July, peaking in August before a sharp decline at the end of the season. In Amparafaravola, the seasonal profile is bimodal, characterized by a pronounced dip in August—probably related to local precipitation—followed by a second, less intense peak in September. In Betafo, the dynamics are marked by strong post-peak instability, with highly irregular daily fluctuations. This behavior suggests that the measured climatic parameters do not exert systematic control and that other local factors may contribute to regulating spore concentrations.



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