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Characterization and pollen morphological comparison of ornamental plants in different environments

Caracterização e comparação morfológica polínica de plantas ornamentais em ambientes distintos

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ABSTRACT
There is a great variety in the shapes and sizes of pollen grains, which facilitates the morphological characterization, making the analysis of pollen morphology an effective and fundamental tool in the taxonomic systematization. However, the presence of intraspecific pollen variation exists. This study aimed to verify intraspecific palynological variations in pollen grains among three ornamental plant species growing in different locations: *Antigonon leptopus* (Polygonaceae), *Ixora chinensis* (Rubiaceae), *Pleroma mutabile* (Melastomataceae). Two samples of each species were collected in two different regions (Itapoa and Joinville, Santa Catarina State, Brazil). The buds of the studied species were collected during field trips, the anthers were removed, macerated and acetolyzed, before preparing slides that were analyzed by light microscopy. The grains of the species studied showed variability in size from one municipality to another, with the grains of the species located in Joinville showing larger measurements of both the equatorial diameter and the polar axis when compared to those of the grains of the species from Itapoa. These results are of great importance for expanding studies in systematics, ecology, and plant physiology, demonstrating how different environmental conditions can influence the adaptation of species.
Keywords: morphology, Palynology, pollen.

RESUMO
Existe uma grande variedade nas formas e nos tamanhos dos grãos de pólen, o que facilita a caracterização morfológica, tornando a análise da morfologia polínica uma ferramenta eficaz e fundamental na sistematização taxonômica. No entanto observa-se a presença de variação polínica intraespecífica. Este estudo teve como objetivo verificar as variações palinológicas intraespecíficas em grãos de pólen de três espécies de plantas ornamentais que crescem em diferentes localidades: *Antigonon leptopus* (Polygonaceae), *Ixora chinensis* (Rubiaceae) e *Pleroma mutabile* (Melastomataceae). Coletaram-se duas amostras de cada espécie em duas regiões distintas (Itapoá e Joinville, estado de Santa Catarina, Brasil). Os botões florais das espécies estudadas foram coletados durante expedições de campo, as anteras foram removidas, maceradas e acetolisadas, antes da preparação de lâminas, as quais foram analisadas por microscopia óptica. Os grãos das espécies examinadas apresentaram variabilidade de tamanho entre os municípios; os grãos das espécies localizadas em Joinville evidenciaram medidas maiores tanto do diâmetro equatorial quanto do eixo polar quando comparados aos grãos das espécies de Itapoá. Tais resultados são de grande importância para expandir os estudos em sistemática, ecologia e fisiologia vegetal, uma vez que demonstram como diferentes condições ambientais podem influenciar a adaptação das espécies.
Palavras-chave: morfologia, Palinologia, pólen.

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INTRODUCTION

There is a great variety in the shapes and sizes of pollen grains, which facilitates the morphological characterization of some taxa and, therefore, the analysis of pollen morphology becomes an effective and fundamental tool in the taxonomic systematization of phanerogams (ANDRADE, 2020). However, the presence of intraspecific pollen variation exists and is reported in several taxa (BOLNICK *et al.*, 2011). This situation can eventually make these characters display a range of variations, even overlapping with those of other species and hence restrict their perception as of taxonomic value (VIOLLE *et al.*, 2012). Pollen morphology, in addition to aiding taxonomy, can reveal information about plant-pollinator interactions and provide insights into the resilience of species in the face of climate change and environmental stressors (ALONSO-BLANCO *et al.*, 2009). Thus, the characterization and morphological comparison of pollen from plants of the same species, living in different environments, is of great importance for expanding studies in systematics, ecology, and plant physiology, allowing us to demonstrate how different environmental conditions can influence the adaptation of species. In this context, this study aimed to verify intraspecific variations in pollen grains among ornamental plant species growing in different locations, by observing the diverse morphological palynological characteristics.

MATERIAL AND METHODS

Field trips were conducted in the municipalities of Itapoa and Joinville, in the state of Santa Catarina (SC), southern Brazil. This state is entirely within the Atlantic rain forest biome. The two municipalities are 30.97 km apart in a straight line (figure 1).

Itapoa has an altitude of 18 m and is characterized as having a humid subtropical climate, characterized by mild winters and hot summers, according to Silveira *et al.* (2016). As the municipality is in a coastal area, the temperature is higher as average when compared to higher altitude inland regions in SC. According to Monteiro (1963), southern Brazil, including Itapoa, presents a mesothermal climate pattern with significant annual temperature range, due to the combination of geographical and climatic factors typical of the subtropical zone. Furthermore, the region's climate is influenced by tropical atmospheric systems, such as warm air masses, which intensify the heat during the summer (SILVEIRA *et al.*, 2019). Itapoa has a vegetation cover predominantly composed of coastal plain Atlantic rain forest and, in addition, typical beach vegetation formations, *restinga* (coastal dune scrubland), and mangroves are found but, in the mountainous areas, the denser and more diverse formations of submontane and montane forest stand out, harboring a rich biodiversity, including native tree species such as *Ocotea catharinensis* Mez. (Lauraceae), *Aspidosperma olivaceum* Müll. Arg. (Apocynaceae), *Ocotea odorifera* (Vellozo) Rohwer (Lauraceae), *Manilkara subsericea* (Mart.) Dubard (Sapotaceae), *Nectandra lanceolata* Nees & Mart. ex Nees (Lauraceae), and *Schizolobium parahyba* (Vell.) Blake (Fabaceae) (MELLO *et al.*, 2012).

Joinville, according to data presented by the Joinville Municipal Government (2020), has an altitude of 4 m, with a predominantly humid mesothermal climate, without a dry season, and among the ecosystems that occur in the Joinville region, the Dense Ombrophilous rain Forest (approximately 680 km^2) and its associated ecosystems stand out, with more than 60% coverage, notably the mangroves with 36 km^2.

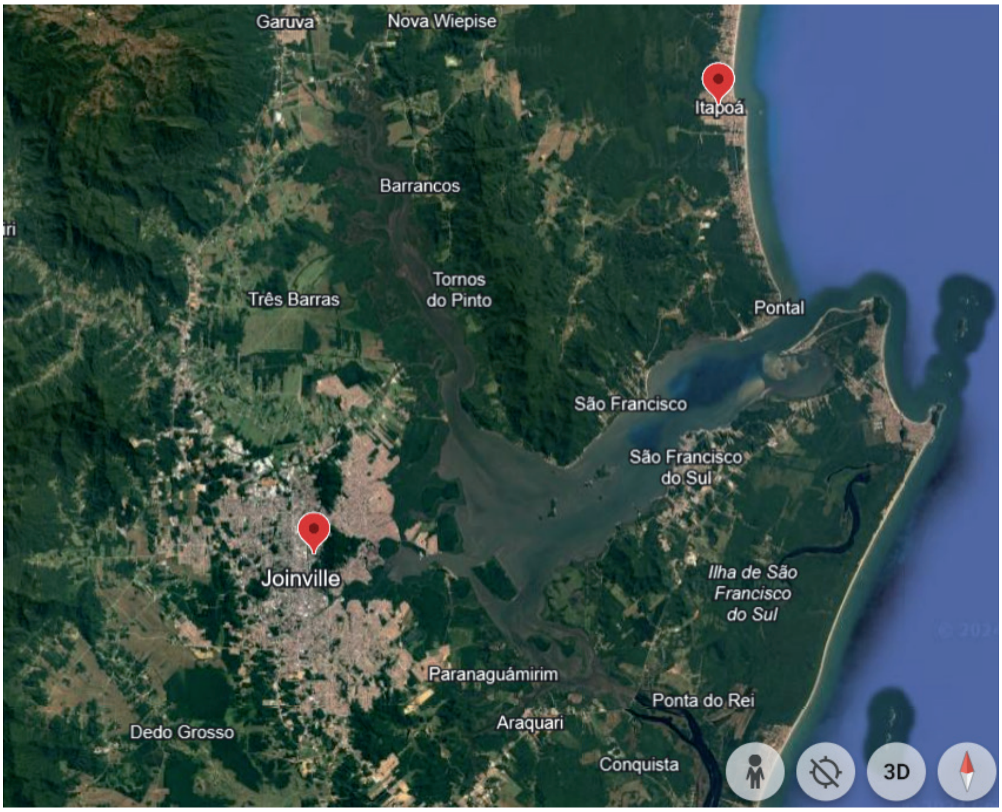

Figure 1 – Map of the studied locations. Source: Google Earth.

The following species were selected and collected: *Antigonon leptopus* Hook. & Arn. (Polygonaceae), *Ixora chinensis* Lam. (Rubiaceae) and *Pleroma mutabile* (Vell.) Triana (Melastomataceae), in both municipalities (figures 2 and 3).

Figure 2 – Images of the species studied in Itapoa, SC: 1A) *Antigonon leptopus*; 2A) *Ixora chinensis*; 3A) *Pleroma mutabile*. Source: primary.

Figure 3 – Images of the species studied in Joinville, SC: 1B) *Antigonon leptopus*; 2B) *Ixora chinensis*; 3B) *Pleroma mutabile*. Source: primary.

After the classification and identification of the plants, the unprecedented nature of pollen studies on the sampled taxa was investigated, and the collected specimens were prepared for herbarium preparation. Closed buds were removed from the species and preserved in acetic acid (JANSONIUS & McGREGOR, 1996). Just before acetolysis, the buds were opened, the anthers removed and macerated in a petri dish and then subjected to the acetolysis process (placed in acidic solutions and specific substances and subjected to centrifugation) (ERDTMANN, 1960). The acetolyzed pollen grains were mounted in glycerinated gelatin on thin light microscope slides (five slides per species) (FÆGRI & IVERSEN, 1989). The slides were deposited in the palynological collection of the Bee Laboratory (Label) at Univille.

The slides were observed under a light microscope (400x) using the "Dino-Eye Capture 2.0" software. The pollen grains on the slides were photographed, with 25 repetitions for each type of view (equatorial and polar), measuring the following parameters in micrometers (µm): equatorial diameter and polar axis, as well as exine thickness (BARTH & MELHEM, 1988). The grains were classified based on characteristics such as pollen unit, size, scope, symmetry, polarity, shape, apertures, and ornamentation (PUNT *et al.*, 2007). The measurements were tabulated (Microsoft Excel) to obtain the size range (Xmin-Xmax), the arithmetic mean, and the standard deviation of the arithmetic mean (MOORE *et al.*, 1991).

The data were compiled into tables, with the respective measurements of pollen grains in polar and equatorial views, exine thickness measurement, and other characteristics (pollen unit, size, scope, symmetry, polarity, shape, apertures, ornamentation) (SALGADO-LABORIAU, 2007).

Comparative tables were produced with the results from both municipalities, for the same species, three tables for each species, comparing the grain measurements between the municipalities (polar axis, equatorial diameter, and exine thickness).

Tables with descriptive statistics for each region were produced using Excel. A t-test was performed for the average measurements assuming different variances, and subsequently, box-plot graphs were made for each parameter analyzed.

RESULTS AND DISCUSSION

The numerical results are presented in tables 1, 2, 3 and 4, and the images of the pollen grains are in figure 4. All pollen grains are monads, isopolar, with radial symmetry.

The pollen grains of the ornamental species of Itapoa presented the following results: *Antigonon leptopus* (Polygonaceae), *Ixora chinensis* (Rubiaceae) and *Pleroma mutabile* (Melastomataceae) have 3-colporated grains with circular amb, and *P. mutabile* is also 3-pseudocolpate; the shape and thickness of the exine of these species are oblate-spheroidal (*A. leptopus* [P=50,12 µm; E=54,30 µm;

ex=1,92 µm] *I. chinensis* [P=25,78 µm; E=27,13 µm; ex=1,23 µm] *P. mutabile* [P=18,63 µm; E=20,24 µm; ex=1,43 µm]).

For the pollen grains of the species found in Joinville, the result were: *Antigonon leptopus, Ixora chinensis,* and *Pleroma mutabile* have 3-colporate grains with circular amb, and *Pleroma* is also 3-pseudocolpate; the shape and thickness of the exine in these species are oblate-spheroidal except *P. mutabile* that is suboblate (*A. leptopus* [P=60,08 µm; E=64,98; ex=2,91] *I. chinensis* [P=30,96 µm; E=31,74 µm; ex=0,97 µm] *P. mutabile* [P=21,56 µm; E=25,83 µm; ex=1,44 µm]).

Table 1 – Morphometric data of the pollen grains of the species studied in the municipality of Itapoa. Abbreviations: P/E=polar axis/equatorial diameter.

	P/E	**Shape**	**Exine (µm)**	**Apertures**
Antigonon leptopus	0,92	Oblate-spheroidal	1,92	3-colporate
Ixora chinensis	0,95	Oblate-sphroidal	1,23	3-colporate
Pleroma mutabile	0,92	Oblate-spheroidal	1,43	3-colporate 3-pseudocolpate

Table 2 – Morphometric data of the pollen grains of the species studied in the municipality of Joinville. Abbreviations: P/E=polar axis/equatorial diameter.

	P/E	**Shape**	**Exine (µm)**	**Apertures**
Antigonon leptopus	0,92	Oblate-spheroidal	2,91	3-colporate
Ixora chinensis	0,97	Oblate-spheroidal	2,48	3-colporate
Pleroma mutabile	0,83	Suboblate	1,44	3-colporate 3-pseudocolpate

Table 3 – Morphometric data of the pollen grains of the analyzed species from Itapoa.

	Ornamentation	**Âmb**	**Average polar axis (µm)**	**Average equatorial diameter (µm)**	**Size**
Antigonon leptopus	Reticulate	Circular	50,12	54,30	Large
Ixora chinensis	Microreticulate	Circular	25,78	27,13	Medium
Pleroma mutabile	Psilate	Subtriangular	18,63	20,24	Small

Table 4 – Morphometric data of the pollen grains of the analyzed species from Joinville.

	Ornamentation	**Âmb**	**Average polar axis (µm)**	**Average equatorial diameter (µm)**	**Size**
Antigonon leptopus	Reticulate	Circular	60,08	64,98	Large
Ixora chinensis	Microreticulate	Circular	30,96	31,74	Medium
Pleroma mutabile	Psilate	Subtriangular	21,56	25,83	Small

Figure 4 – Images of the pollen grains of the studied species: 1A) *A. leptopus* (sample from Itapoa); 2A) *I. chinensis* (sample from Itapoa); 3A) *P. mutabile* (sample from Itapoa); 1B) *A. leptopus* (sample from Joinville); 2B) *I. chinensis* (sample from Joinville); 3B) *P. mutabile* (sample from Joinville). Source: primary.

The statistical analysis of the average morphometric measurements of the pollen grains between the municipalities is in figures 5, 6 and 7.

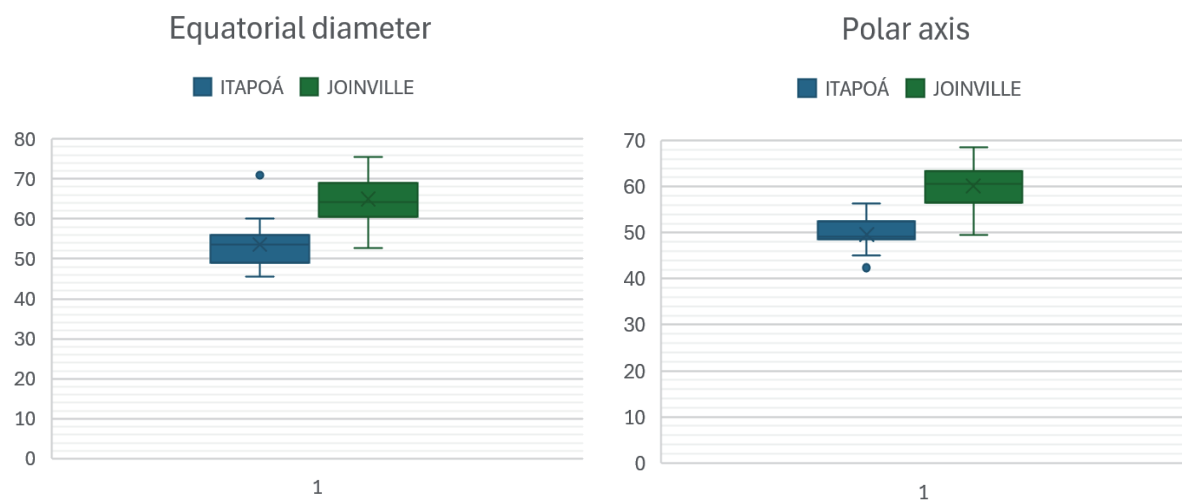

Figure 5 – Graphs 1 and 2: equatorial diameter and polar axis of the studied species *Antigonon leptopus*. Source: primary.

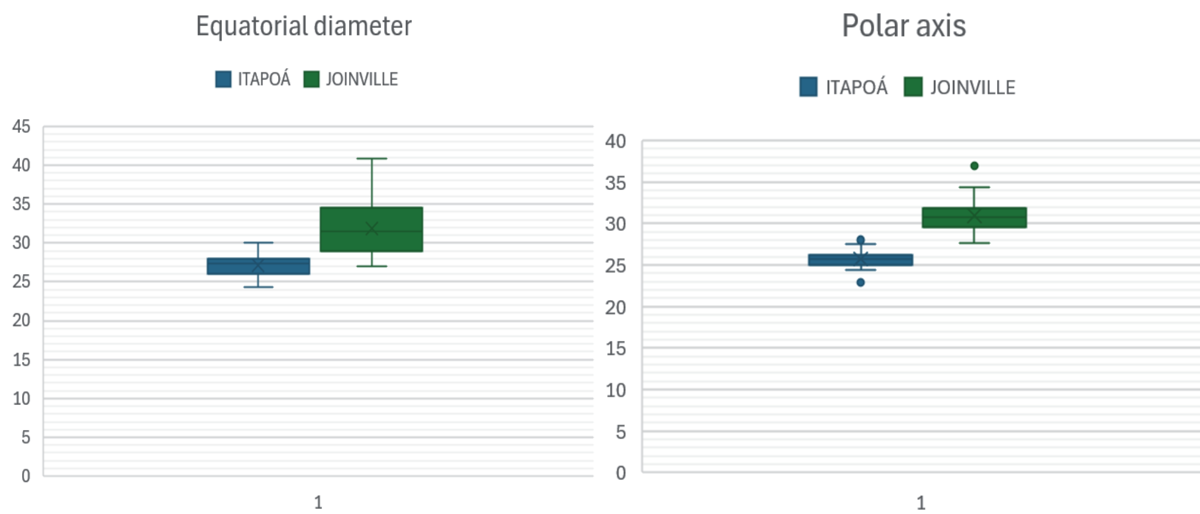

Figure 6 – Graphs 3 and 4: equatorial diameter and polar axis of the studied species *Ixora chinensis*. Source: primary.

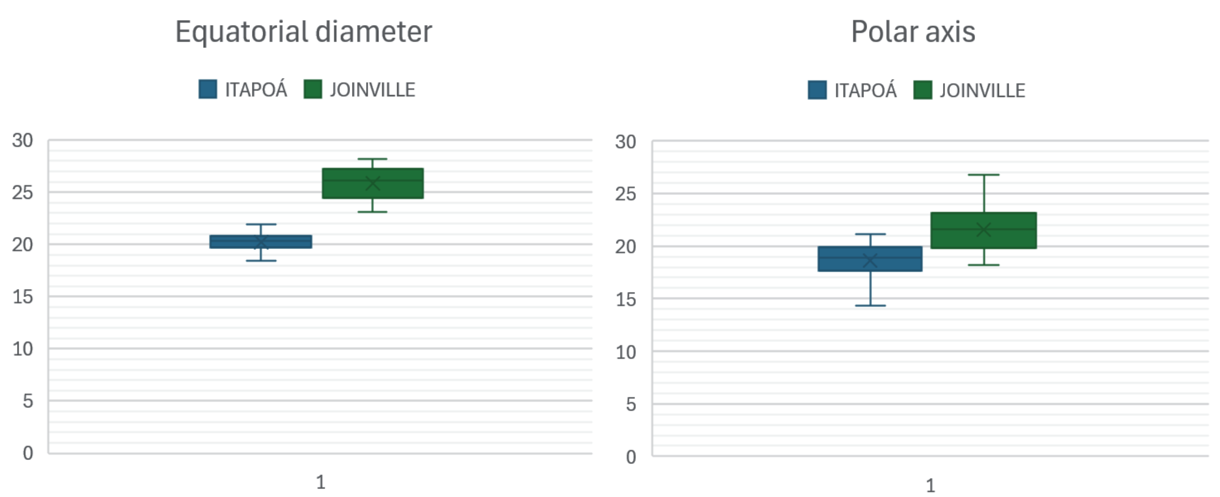

Figure 7 – Graphs 5 and 6: equatorial diameter and polar axis of the studied species *Pleroma mutabile*. Source: primary.

Statistical tests were performed to verify if the sample averages showed a significant difference between them. For this, a two-sample t-test was performed, assuming different variances. The test showed a significant difference between the analyzed data (figures 8 to 10).

1

	Itapoá	Joinville
Average	53,61732	64,98744
Variance	44,08069	30,94648
Observations	25	25
Hypothesis of the difference in averages	0	
Degrees of freedom	47	
Stat t	-6,56335	
P(T<=t) one-tailed test	1,88E-08	
Critical T- value one- tailed test	1,677927	
P(T<=t) two- tailed test	3,76E-08	
Critical T - value two- tailed test	2,011741	

2

	Itapoá	Joinville
Average	49,68128	60,08028
Variance	17,16865	22,80048
Observations	25	25
Hypothesis of the difference in averages	0	
Degrees of freedom	47	
Stat t	-8,22431	
P(T<=t) one-tailed test	5,9E-11	
Critical T- value one- tailed test	1,677927	
P(T<=t) two- tailed test	1,18E-10	
Critical T - value two- tailed test	2,011741	

Figure 8 – 1) t-test, equatorial diameter of *Antigonon leptopus*; 2) t-test, polar axis of *Antigonon leptopus*. Source: primary.

1

	ITAPOÁ	JOINVILLE
Average	27,0732	31,8536
Variance	2,12135	11,4872
Observations	25	25
Hypothesis of the difference in averages	0	
Degrees of freedom	33	
Stat t	-6,47924	
P(T<=t) one-tailed test	1,2E-07	
Critical T- value one- tailed test	1,69236	
P(T<=t) two- tailed test	2,4E-07	
Critical T - value two- tailed test	2,03452	

2

	ITAPOÁ	JOINVILLE
Average	25,7308	30,9367
Varience	1,331021	4,28394
Observations	25	25
Hypothesis of the difference in averages	0	
Degrees of freedom	38	
Stat t	-10,9847649	
P(T<=t) one-tailed test	1,1776E-13	
Critical T- value one- tailed test	1,68595446	
P(T<=t) two- tailed test	2,3552E-13	
Critical T - value two- tailed test	2,02439416	

Figure 9 – 1) t-test, equatorial diameter of *Ixora chinensis*; 2) t-test, polar axis of *Ixora chinensis*. Source: primary.

1

	itapoá	joinville
Average	20,2432	25,8365
Variance	0,64994	2,5249
Observations	25	25
Hypothesis of the difference in averages	0	
Degrees of freedom	36	
Stat t	-15,6956	
P(T<=t) one-tailed test	5,6E-18	
Critical T- value one- tailed test	1,6883	
P(T<=t) two- tailed test	1,1E-17	
Critical T - value two- tailed test	2,02809	

2

	Itapoá	Joinville
Average	18,6387	21,5663
Variance	2,88548	4,41636
Observations	25	25
Hypothesis of the difference in averages	0	
Degrees of freedom	46	
Stat t	-5,41716	
P(T<=t) one-tailed test	1,1E-06	
Critical T- value one- tailed test	1,67866	
P(T<=t) two- tailed test	2,1E-06	
Critical T - value two- tailed test	2,0129	

Figure 10 – 1) t-test, equatorial diameter of *Pleroma mutabile*; 2) t-test, polar axis of *Pleroma mutabile*. Source: primary.

Antigonon leptopus (Polygonaceae)

In the study of pollen from the species *Antigonon leptopus* (Polygonaceae) in different municipalities (Itapoa and Joinville), it is classified as large, tricolporate, oblate-spheroidal, and has a reticulate exine. However, there are differences in the average size of the equatorial diameter and the polar axis between the municipalities, with Itapoa presenting averages of 54,306 μm (equatorial diameter) and 50,125 μm (polar axis), while in Joinville these values were higher, with averages of

64,987 µm (equatorial diameter) and 60,080 µm (polar axis). The calculation of the P/E ratio, which reflects the overall shape of the pollen grain, also varied gradually between the samples from Itapoa (P/E = 0.923) and Joinville (P/E = 0.924), indicating a small difference in sphericity between the populations. Performing statistical tests, such as the boxplot analysis test, reinforces the existence of a significant difference in measurements between the two municipalities, as illustrated in the reference graphs (figures 5 to 7). The exine is also much thicker in Joinville (2,91 µm) than in Itapoa (1,92 µm). But, in a general way, the data obtained are consistent with the literature, such as Paul & Chowdhury (2020) and Heigl (2021), which describe the species' pollen as monadic, isopolar, radiosymmetric, with a reticulate or microreticulate exine.

The morphological variability observed in *A. leptopus* pollen grains between Itapoa and Joinville reinforces the importance of considering regional factors in the analysis of widely distributed species. The significant difference in the size of the equatorial diameter and the polar axis, as well as in the P/E ratio, suggests that these populations are subject to specific environmental or adaptive influences that shape their morphological characteristics. The larger average size of pollen grains in Joinville may be associated with environmental conditions more specific to growth or selection by pollinators that prefer larger grains. Skogsmyr & Lankinen (2002) suggest that pollen size may be correlated with greater vigor or pollen tube growth rates, which increases the chances of reproductive success. Furthermore, a small difference in the P/E ratio between Itapoa and Joinville, with more spherical grains in Itapoa, may reflect local adaptations to specific selective pressures, such as nutrient availability or variations in wind regime. Galen (2000) analyzed how environmental stresses, such as low water availability, can lead to changes in grain size, affecting the reproductive efficiency of plants. Furthermore, the variability suggests that *A. leptopus* exhibits phenotypic plasticity, which may be an adaptive advantage in heterogeneous environments. This plasticity may allow the species to respond to environmental changes or expand its geographic distribution, considering that *A. leptopus* is widely cultivated and naturalized in different tropical and subtropical regions.

Antigonon leptopus is native to the Pacific and Atlantic coastal plains of Mexico but also occurs as a roadside weed from southern Mexico to Central America, being widely introduced and invasive throughout tropical regions of the world, including in the south and eastern USA, the West Indies, South America, the Old World tropics of Asia and Africa and is utilized for its edible properties and also as an ornamental vine in warmer parts of the world (FREEMAN, 2022).

Ixora chinensis (Rubiaceae)

The analyzed species, in both municipalities, presents medium-sized, monad-shaped, tricolporate pollen grains with an oblate-spheroidal, circular shape and a microreticulate ornamented exine. The species found in Itapoa had an average equatorial diameter of 27.137 µm and an average polar axis diameter of 25.780 µm; the calculated ratio between the average polar axis diameter and the equatorial diameter (P/E) was 0.95. Regarding the species found in Joinville, it had an average equatorial diameter of 31.745 µm and an average polar axis diameter of 30.963 µm; the calculated ratio between the average polar axis diameter and the equatorial diameter (P/E) was 0.97. Both populations have pollen grains with an oblate-spheroidal shape and microreticulate ornamentation, characteristics common in the Rubiaceae family, establishing morphological conservation between the locations. The exine is much thicker in Joinville (2,48 µm) than in Itapoa (1,23 µm). The divergence in average sizes also occurred in the statistical tests, where it can be observed, in figure 6, that there is a discrepancy in the average grain values, in addition to the t-test presented in figure 9, which reveals that the averages have a statistically significant difference. Kuang *et al.* (2021) report for *I. chinensis* pollen grains of medium size, with microreticulate exine, tri-aperturate with long colpi, with orbicules (Ubisch bodies) with filaments but the main difference is in the shape thatis reported by the mentioned authors as being prolate, that is, very different.

An analysis of the morphology of the species' pollen grains in the Itapoa and Joinville municipalities reveals variations that may be related to environmental, climatic, or genetic differences. The differences in the P/E ratio, with values of 0.95 for Itapoa and 0.97 for Joinville, suggest that external factors may influence the development of pollen grains. This level of increase in the Joinville index may be associated with variations in climatic conditions, such as relative humidity, temperature, or altitude, which impact pollen development.

Ixora chinensis is native to the subtropical and tropical regions of Asia, specifically originating in southern China and Southeast Asia, including countries like Cambodia, Laos, Myanmar, the Philippines, Thailand, and Vietnam, being a popular evergreen shrub that grows wild in woodlands and shrublands, but it is also widely cultivated in gardens, parks, and along roadsides globally in tropical and subtropical areas worldwide (WU *et al.*, 2011).

Pleroma mutabile (Melastomataceae)

The studied species presents, in both municipalities, small, monad-shaped, subtriangular, tricolporate and tripseudocolpate grains, with a psilate exine. The species found in Itapoa presented an average equatorial diameter of 20.24 µm and an average polar axis diameter of 18.63 µm, while the species found in Joinville presented an average equatorial diameter of 25.83 µm and an average polar axis diameter of 21.56 µm; regarding the P/E ratio, Itapoa had 0.92 and Joinville 0.83; the shape differed between the regions, with the one found in Itapoa classified as oblate-spheroidal and the one found in Joinville as suboblate. The exine thickness is not very different from one place to the other (Joinville – 1,44 µm, Itapoa – 1,43 µm). According to Cruz-Barros *et al*. (2007), the pollen grains of *Pleroma mutabile* (named as *Tibouchina mutabilis* in their article) have a prolate-spheroidal shape (ours are oblate-spheroidal and suboblate), triangular amb (ours are subtriangular), colporate apertures without margins and operculate colpori (ours are 3-colporate, 3-pseudocolpate), rugulate exine (ours are psilate), medium size (ours are small). All samples analyzed by the mentioned authors are from the state of São Paulo, situated at 500 km at least.

These differences can be attributed to local environmental variations, such as humidity, temperature, and soil conditions. The study by Leitão-Filho *et al.* (1993), regarding the species *Tibouchina pulchra,* showed differences related to altitude that influence reproduction and fertilization, indicating ecological adaptations to local conditions, such as pollinator availability and environmental pressure, which impact the size and shape of pollen grains, indicating that phenotypic plasticity in pollen grains is frequently linked to adaptation to specific ecological conditions.

Pleroma mutabile (also known as *Tibouchina mutabilis*) is native to the Brazilian Atlantic rain Forest, primarily in the Serra do Mar region of states like São Paulo, Paraná, Rio de Janeiro, and Santa Catarina, an evergreen pioneer tree with an open crown, cultivated as an ornamental plant in Brazil and Australia (GUIMARÃES *et al*., 2019).

CONCLUSION

In short, the measurements carried out show that, in Joinville, the grains had larger measurements. The increase in polar diameter and area may be associated with an increase in exine thickness, since this acts as a protective layer for the pollen against solar radiation, desiccation, and pathogen attack (DOBRITSA *et al*., 2009).

The morphological studies of the species *Antigonon leptopus*, *Ixora chinensis*, and *Pleroma mutabile* done here revealed variations in pollen grains between the environments of the Itapoa and Joinville regions, highlighting the impact of, probably, environmental and ecological factors on pollen characteristics. Analysis of palynological parameters, such as equatorial diameter, polar axis, and P/E ratio, showed that these differences are statistically significant and specific, possibly associated with local variations in temperature, humidity, altitude, and pollinator availability. The phenotypic plasticity observed in the three analyzed species evidences the adaptive capacity of the plants to respond to heterogeneous environmental conditions, which may favor their survival and geographic expansion. In the case of *A. leptopus*, the discrepancy with literature reinforces the need for detailed regional studies to capture morphological nuances that reflect local adaptations. Similarly, the differences observed for *I. chinensis* and *P. mutabile* illustrate how specific selective pressures can shape morphological aspects related to reproductive efficiency.

There have been observations about intraspecific pollen variations associated with environmental factors. Thus, Koti *et al*. (2005) verified that exposing *Glycine max* (L.) Merr. to different day/night temperatures caused variations in exine sculpturing and pollen shape. Karabournioti *et al*. (2007) and Xiao *et al*. (2022) showed that the annual precipitation contributed to intraspecific variation of pollen in *Thymus capitatus* Hoffmanns & Link, and *Cathaya argyrophylla* Chun. & Kuang. respectively. Silva *et al*. (2019) analyzed morphological changes in physalis pollen grains (*Physalis peruviana* L. (Solanaceae)) grown under different light spectra and verified that plants grown in full sun and under white mesh treatments have pollen grains with a larger transverse diameter and area compared to the other treatments. Fatmi *et al*. (2020) assessed, in *Atriplex halimus* R.Br., that ten pollen shapes were produced as being due to gradient of different climatic zones. Complementarily, intraspecific variations associated with geographical locations (latitudes and longitudes) were observed in *Paris tetraphylla* A. Gray (KURITA *et al*., 1990) and *Sambucus nigra* L. (WRONSKA-PILAREK *et al*., 2020). Hasegawa *et al*. (2023), in *Weigela hortensis* C. A. Mey., noticed an increase in spine length and density with altitude and attributed this altitudinal variation in exine ornamentation to changes in pollinator's assemblages. It should also be reminded that the growing conditions of tropical ornamental plants, related to factors such as rainfall, humidity, temperature, and planting density, favor the occurrence of diseases that limit production, reduce flower quality and affect reproductive aspects (LINS & COELHO, 2004).

The importance of the observations made relies on the fact that pollen morphological characters are used in the taxonomy of angiosperms and are often engaged in resolving relationships at different taxonomic levels (EL GHAZALI, 2025). However, in taxa possessing intraspecific pollen morphological variations, palynological characters may fail to validate taxonomic relationships and it is therefore relevant to assess the intraspecific morphological variability of pollen, which will allow us to determine the taxonomic value of these characteristics. Pollinic variations broaden the knowledge about the intraspecific diversity of species and emphasize the relevance of integrative analyses in the study of biodiversity, highlighting the importance of ecological and regional studies to understand the relationship between morphology and adaptation to the environment. Environmental factors influence the phenotypic variation within species, play a key role in natural selection and determine evolutionary courses.

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