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Pollen morphology of Brazilian *Fevillea* (Cucurbitaceae)

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**Abstract**

Pollen morphology of the five Brazilian species of the genus *Fevillea* (*F. bahiensis*, *F. cordifolia*, *F. passiflora*, *F. pedatifolia*, and *F. trilobata*) was studied using light and scanning electron microscopy. Pollen in these species is generally stenopalynous, which is in agreement with observations in previous studies of other species. Pollen is shed in monads and characterised by being isopolar, radially symmetric, prolate, and striate. Pollen is of medium size, tricolporate, with long and narrow colpi, a circular endoaperture and a reduced polar area. The exine is up to 2 μm thick.

**Keywords:** Basal Cucurbitaceae, Brazil, pollen morphology

The family Cucurbitaceae comprises about 825 species in 118 genera, with a predominantly tropical distribution (90% of the species) in Africa, Central and South America and Southeast Asia (Jeffrey, 2005). Two subfamilies, Fevilleoideae (syn. Nhandiroboideae, Zanonioidae) and Cucurbitoideae, were traditionally recognised based on morphological (Jeffrey, 2005) and molecular (Schaefer et al., 2009) characters. This subdivision was also indicated by molecular data (Kocyan et al., 2007), although the support was not strong. However, the most recent molecular analyses failed to recognise this subdivision and instead identified five well-supported clades, one corresponding to the traditional subfamily Cucurbitoideae and four clades that include taxa traditionally assigned to the subfamily Fevilleoideae (Schaefer et al., 2009). One of the four clades corresponds to the tribe Fevilleeae of Bentham and Hooker (1867) and includes *Fevillea*, four other neotropical genera and one African genus. The three other clades correspond to the tribes Gomphogyneae of Bentham, Fevilleeae of Bentham and Hooker (1867), Zanonioidae of Blume (1826), and a clade including two Asian genera. Although these four clades do not constitute a monophyletic group, we refer to them here as the subfamily Fevilleoideae.

Members of Cucurbitaceae are well represented in Brazil. The revisions of selected Cucurbitaceae species by Campos (1962), Melhem (1966), and Salgado-Labouriau (1973) and the detailed study on pollen morphology of the genus *Cayaponia* by Barth et al. (2005) are the most important contributions to the current knowledge of the family in Brazil. A recent study by Van der Ham et al. (2010) focused particularly on the tectum ornamentation in the subfamily Fevilleoideae (*Fevillea* and related clades).

According to Erdtman (1952), Marticorena (1963), Jeffrey (1964, 1990), Aloyshina (1966), Shridhar and Singh (1990) and Khunwasi (1998), the pollen of Cucurbitaceae is eurypalynous, with considerable differences in grain shape, ornamentation pattern and position of apertures between the individual genera. However, within the tribes, subtribes and genera, the pollen is typically stenopalynous with only slight variation between pollen of the different species.
At family level, palynological data are therefore of great value as a taxonomical tool at the tribal or subtribal level.

The tribe Fevilleeae is placed close to the base of the Cucurbitaceae phylogenetic tree (Schaefer et al., 2009) and its members have morphologically rather similar pollen. Pollen morphology of the subfamily Fevilleoideae has been relatively well investigated (Aloyshina, 1966; Khunwasi, 1998; Lira et al., 1998; Van der Ham, 1999; de Wilde et al., 2007; Van der Ham et al., 2010).

The genus *Fevillea* is tropical American with six species of which four species, *F. bahiensis* G. L. Rob & Wunderlin, *F. cordifolia* L., *F. pedatifolia* (Cogn.) C. Jeffrey and *F. trilobata* L., are known from Brazil. According to Nee et al. (2009), studies of molecular phylogeny revealed the validity of the monotypic genus *Anisosperma* [A. *passiflora* (Vell.) S. Manso]. This work follows the circumscription of Robinson and Wunderlin (2005) to include *Anisosperma* in *Fevillea* and thus includes also *Fevillea passiflora* Vell.

Studies on pollen morphology of the genus have been published by Erdtman (1952), Marticorena (1963), Aloyshina (1966), Roubik and Moreno (1991), Khunwasi (1998) and Van der Ham et al. (2010). The present study is focused on pollen morphology of the Brazilian representatives of the genus *Fevillea* in order to contribute to the palynological knowledge of the Cucurbitaceae. It comprises preliminary results of a broad taxonomic revision of the basal cucurbitaceous lineages in Brazil.

**Materials and methods**

Pollen samples were obtained from herbarium specimens deposited in the following Brazilian and European herbaria: IAN (Embrapa Amazônia Oriental, Belém, Brazil), ICN (Instituto de Ciências Naturais, Porto Alegre, Brazil), INPA (Instituto Nacional de Pesquisas da Amazônia, Manaus, Brazil), IPA (Empresa Pernambucana de Pesquisa Agropecuária, Recife, Brazil), UB (Universidade de Brasília, Brasília, Brazil) and S (Swedish Museum of Natural History, Stockholm, Sweden). To document total morphological variability for each species, all the fertile species were analysed.

Pollen samples were processed following the acetolysis method described by Erdtman (1952). Permanent slides were mounted in glycerine jelly and stored at the Palynotheca of the Laboratory of Palynology of the Universidade Luterana do Brasil (ULBRA), Canoas, Brazil. Five slides of each herbarium specimen were prepared. General observations were made using a Leica DMLB optical microscope. Dimensions of grains were measured one week after acetylosis (cf. Salgado-Labouriau, 1973). Twenty-five pollen grains per species were measured in equatorial view for determination of the polar diameter (P), the equatorial diameter (E), the exine thickness and grain shape (P/E). Terminology follows Punt et al. (2007). Statistical analysis were conducted to obtain the means and standard deviations, and coefficient of variation were calculated and compared using the confidence interval (95%). Samples for scanning electron microscopy (SEM) were separated after acetolysis and conserved in 50% glycerol. Part of the suspended material was applied to a filter paper for drying and then transferred to the surface of metallic stubs previously covered with carbon tape. After drying, the samples were coated with gold/palladium and studied using a Phillips XL 20 scanning electron microscope.

**Results**

Pollen of the Brazilian genus *Fevillea* is stenopalous and can be characterised by the following general description: monads; isopolar, radially symmetric, prolate (subprolate in *F. pedatifolia*); medium size; striate, with reduced polar area; tricorate with long and narrow colpi; circular endopertum, exine up to 2 μm thick (Figure 1).

*Fevillea pedatifolia* had the smallest polar diameter (23.62 μm), while *F. cordifolia* has the largest pollen with an equatorial diameter of 31.06 μm. The smallest mean equatorial diameter was identified in *F. trilobata* (18.91 μm); the largest was in *F. cordifolia* (23.30 μm) (see Tables I and II).

**Discussion**

The Cucurbitaceae are a family of europalous pollen characterised by rather great variation in pollen morphology. However, at the level of tribes and subtribes, pollen morphology tends to be more consistent. According to our study, pollen in the genus *Fevillea* is stenopalous. Consequently, the pollen morphology of the Brazilian *Fevillea* is in full agreement with the reported profile for the other taxa traditionally included in the subfamily Fevilleoideae (Marticorena, 1963; Jeffrey, 1990; Kocyan et al., 2007). The presence of a striate exine is in the Cucurbitaceae an exclusive feature of this subfamily (Marticorena, 1963; Khunwasi, 1998; Van der Ham, 1999; de Wilde et al., 2007; Van der Ham et al., 2010). A reticulate exine occurs in *Bolbostemma*, some species of
Gerrardanthus, and also in Neoalsomitra suberosa (F. M. Bailey) Hutch (Kocyan et al., 2007). In addition, ambiguities in exine ornamentation pattern were documented for Alsomitra macrocarpa M. Roem with a reticulate pattern observed by Marticorena (1963) and a striate pattern observed by Khunwasi (1998). Van der Ham (1999) described the pattern as indistinctly rugulate. Generally, the morphological features of pollen examined by Marticorena (1963), Roubik and
Moreno (1991) and Khunwasi (1998) are confirmed by the present study. SEM observations revealed that exine striations are similar to those of *Sicydium*, *Neoalsomitra*, *Zanonia indica* (Lira et al., 1998; Van der Ham, 1999; de Wilde et al., 2007), which are formed by structures arranged in rings or in a spiral pattern. Lira et al. (1998) and de Wilde et al. (2007) also observed the existence of a granular membrane in the colpi of some *Sicydium* species, which is also seen in all species of *Fevillea* studied here (Figure 2).

A review of the relevant literature and the data provided in the present work suggest that pollen morphology alone cannot be used for distinction at the generic and specific level in the Fevilleeae and related clades traditionally included in the subfamily Fevilleoideae. Consequently, additional studies focusing on the search of morphological micro-characters using transmission electron microscopy (TEM) and exine stratification in this subfamily are necessary for a better understanding of intrafamilial relationships in the basal lineages of Cucurbitaceae.

**Acknowledgements**

We wish to thank the herbaria IAN (Embrapa Amazônia Oriental, Belém, Brazil), ICN (Instituto de Ciências Naturais, Porto Alegre, Brazil), INPA (Instituto Nacional de Pesquisas da Amazônia, Manaus, Brazil), IPA (Empresa Pernambucana de Pesquisa Agropecuária, Recife, Brazil), UB (Universidade de Brasília, Brasilia, Brazil) and S (Swedish Museum of Natural History, Stockholm, Sweden). This work is part of the Programa de Taxonomia (PROTAX/CNPq), process number 56.3949/2005-8.

**Specimens investigated**

*Fevillea bahiensis*. Brazil, Bahia, Santos 2307 (IPA).

*Fevillea cordifolia*. Brazil, Acre, Cid & Nelson 2596 (INPA); Pará, Tavares et al. 334 (INPA) and Rondônia, Prance et al. 6629 (INPA).

*Fevillea passiflora*. Brazil, Paraná, Dusén 13640 (S); Santa Catarina, Sobral et al. 9192 (ICN) and São Paulo, Costa 338 (ICN).

*Fevillea pedatifolia*. Brazil, Acre, Prance et al. 2936 (S) and Amazonas, Lemos Froés 20620 (IAN).

*Fevillea trilobata*. Brazil, Bahia, Bautista et al. 1686 (INPA); Santos s.n. (IPA 18218); Minas Gerais, Heringer 6393 (UB); Pernambuco, Pickel 524 (IPA) and Rio Grande do Sul, Hagelund s.n. (ICN 148453).

**Table I.** Polar diameter (P) measurements (in μm) of pollen grains of the Brazilian species of *Fevillea*.

<table>
<thead>
<tr>
<th>Species</th>
<th>Variation</th>
<th>$\bar{x}$ ± SD</th>
<th>CV (%)</th>
<th>IC 95 (%)</th>
<th>P/E</th>
<th>EX (μm)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Fevillea bahiensis</em></td>
<td>25.00–32.50</td>
<td>27.21 ± 2.37</td>
<td>8.74</td>
<td>26.30–28.12</td>
<td>1.35</td>
<td>&lt;2.0</td>
</tr>
<tr>
<td><em>Fevillea cordifolia</em></td>
<td>25.00–37.50</td>
<td>31.06 ± 2.69</td>
<td>0.67</td>
<td>30.85–31.27</td>
<td>1.33</td>
<td>&lt;2.0</td>
</tr>
<tr>
<td><em>Fevillea passiflora</em></td>
<td>22.50–39.00</td>
<td>29.99 ± 2.79</td>
<td>9.31</td>
<td>29.77–30.21</td>
<td>1.34</td>
<td>&lt;2.0</td>
</tr>
<tr>
<td><em>Fevillea pedatifolia</em></td>
<td>20.00–27.50</td>
<td>23.62 ± 2.34</td>
<td>4.34</td>
<td>23.40–23.84</td>
<td>1.19</td>
<td>&lt;2.0</td>
</tr>
<tr>
<td><em>Fevillea trilobata</em></td>
<td>22.50–39.00</td>
<td>27.94 ± 1.83</td>
<td>6.38</td>
<td>27.78–28.15</td>
<td>1.48</td>
<td>&lt;2.0</td>
</tr>
</tbody>
</table>

Arithmetic average ($\bar{x}$), standard deviation (SD), variability coefficient (CV), confidence interval (IC) and exine thickness (EX), ($n = 25$).

**Table II.** Equatorial diameter (E) measurements (μm) of pollen grains of the Brazilian species of *Fevillea*.

<table>
<thead>
<tr>
<th>Species</th>
<th>Variation</th>
<th>$\bar{x}$ ± SD</th>
<th>CV (%)</th>
<th>IC 95 (%)</th>
<th>Shape</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Fevillea cordifolia</em></td>
<td>15.00–30.00</td>
<td>23.30 ± 2.34</td>
<td>10.40</td>
<td>23.12–23.48</td>
<td>Prolate</td>
</tr>
<tr>
<td><em>Fevillea passiflora</em></td>
<td>12.50–32.50</td>
<td>22.43 ± 2.37</td>
<td>11.10</td>
<td>22.17–22.66</td>
<td>Prolate</td>
</tr>
<tr>
<td><em>Fevillea pedatifolia</em></td>
<td>17.50–22.50</td>
<td>19.73 ± 1.66</td>
<td>8.41</td>
<td>19.58–19.89</td>
<td>Subprolate</td>
</tr>
<tr>
<td><em>Fevillea trilobata</em></td>
<td>12.50–30.00</td>
<td>18.91 ± 0.26</td>
<td>1.37</td>
<td>18.45–19.17</td>
<td>Prolate</td>
</tr>
</tbody>
</table>

Arithmetic average ($\bar{x}$), standard deviation (SD), variability coefficient (CV), confidence interval (IC) and shape, ($n = 25$).

References


