



Title	Pollen morphology and infrageneric classification of Alstroemeria L. (Alstroemeriaceae)
Author(s)	Sarwar, Abul Khayer Mohammad Golam; Hoshino, Yoichiro; Araki, Hajime
Citation	Grana, 49(4), 227-242 https://doi.org/10.1080/00173134.2010.529503
Issue Date	2010-12
Doc URL	http://hdl.handle.net/2115/47914
Rights	This is an electronic version of an article published in Grana, Volume 49, Issue 4 December 2010 , pages 227-242 . Grana is available online at: http://www.informaworld.com/smpp/
Type	article (author version)
File Information	sarwar.pdf



[Instructions for use](#)

Pollen morphology and infrageneric classification of *Alstroemeria* L. (*Alstroemeriaceae*)

ABUL KHAYER MOHAMMAD GOLAM SARWAR^{1,2}, YOICHIRO HOSHINO² AND
HAJIME ARAKI²

¹*Department of Crop Botany, Bangladesh Agricultural University, Mymensingh, Bangladesh,*

²*Field Science Center for Northern Biosphere, Hokkaido University, Sapporo, Japan*

Abstract

Pollen morphology of 53 taxa of the South American genus *Alstroemeria* was examined using light and scanning electron microscopy (LM and SEM, respectively) or only with SEM in search of new characters that might contribute to infrageneric classification of this genus. The *Alstroemeria* species are stenopalynous, characterised by monosulcate and large monads with striate-reticulate exine sculpture. Pollen with auriculae-like structures at each end of the sulcus is reported for the first time in *Alstroemeria*. Based on exine sculpture, the studied taxa were divided into two major groups; one with striate-reticulate exine (Type I) and the other with finely to coarsely rugulate-psilate exine (Type II). Type I exine sculpture can be further divided into two sub-types based on the width of muri. Although species from Brazil show relatively closer positions in principal component analysis of quantitative data, agglomerative hierarchical clustering of palynological features do not support a subdivision of this genus into a Brazilian and a Chilean species group. The infrageneric classification of *Alstroemeria* can be reliably achieved by combined analyses of morphological, palynological and molecular data from larger number of specimens of all species.

Keywords: *Alstroemeria, Brazilian species group, Chilean species group, non-acetolysed pollen, exine sculpture, infrageneric classification*

A. K. M. Golam Sarwar, Department of Crop Botany, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh. E-mail: gsshameem@gmail.com

(Received 29 March 2010; accepted XX October 2010)

The genus *Alstroemeria*, one of the largest genera of the family Alstroemeriaceae, consists of c. 60 species. The number of genera in Alstroemeriaceae varies from two to five (Aker & Healy, 1990; Bayer, 1998; Aagensen & Sanso, 2003; Hofreiter, 2007; Hoshino, 2008). The circumscription and taxonomic relationships between the two largest and closely related genera, *Alstroemeria* and *Bomarea*, have also been controversial, as some species have been published under both generic names with different epithets (Sanso & Xifreda, 2001).

Alstroemeria is a South American endemic with two centers of distribution, one in Chile (extending into contiguous Peru, Bolivia and Argentina) and the second throughout the eastern third of Brazil and contiguous Paraguay and Argentina (Uphof, 1952; Bayer, 1987; Aker & Healy, 1990).

Alstroemeria comprises perennial herbs that are erect and rhizomatous with storage roots. The leaves are sessile, simple, entire, generally resupinate, and either concentrate towards the stem apex, distributed along the stem, or as rosette on short stem. Blades can be membranous, chartaceous or leathery; linear, lanceolate, elliptic, or oblong; and either glabrous on both faces or papillose on the adaxial face. The inflorescence is terminal as an umbel-like, simple or branched cyme of two to many flowers or rarely the flowers may be solitary, wrapped within the leaf-like bracts. Flowers are zygomorphic and bisexual, with six free and petaloid tepals in two whorls. The tepals may be similar or different, the inner often

shorter, patterned and basally narrowed into a canaliculated base with necteriferous ducts.

The stamens are six, free, and in two whorls. The anthers are pseudobasifix, with longitudinal introse dehiscence. The ovary is inferior, syncarpous, tricarpellate, and trilocular with axile placentation; the style is apical, with three stigmas, and many anatropous ovules. The fruit is a dehiscent, locucidal capsule, and the seeds are globose, without a sarcotesta. Several species and hybrids of some *Alstroemeria* are cultivated commercially for their ornamental value as pot plants as well as cut flowers. The starchy storage roots of several species of *Alstroemeria* are edible (Bayer, 1998).

Erdtman (1952) described the pollen grains of *Alstroemeria* as “1-sulcate, large (longest axis 75–100 µm), usually distinctly plane-convex with sulcus on the convex part of grains, sexine striate (the proximal pole often forms a centre for striation). Sexine is thicker than nexine.” Previous studies of pollen morphology of *Alstroemeria* were mainly based on light microscopy (LM) and/or scanning electron microscopy (SEM) studies of selected taxa, or incorporated into regional pollen flora (Heusser, 1971; Schulze, 1978; Kosenko, 1994; Rudall et al., 2000; Sanso et al., 2005; Hofreiter, 2006). Transmission electron microscopy (TEM) has also been employed in a few studies (Sanso & Xifreda, 2001; Furness & Rudall, 2003). The pollen morphology of some taxa, viz. *A. brasiliensis*, *A. haemantha* and *A. pelegrina*, has been described more than once, but little is known about palynological features over the whole genus and their systematic/taxonomic implications.

Hitherto, *Alstroemeria* has not been formally divided into subgenera, but few attempts have been made to present an infrageneric classification of the genus. Baker (1888) divided *Alstroemeria* into Brazilian and Chilean species groups using mostly quantitative characters from leaf and umbel. The Brazilian species were subdivided further according to the leaf structure: thin leaves or rigid, strongly nerved leaves, but no formal divisions were made and, Bayer (1987), who revised the Chilean species, noted that a subdivision is premature until the Brazilian species can be included in the analysis. Ravenna (2000) proposed the section *Andesine* based on floral characters, including species from the Andean region and Patagonia, but it does not agree with the provisions of ICBN (Aagesen & Sanso, 2003, p. 59). The differences in floral morphology (Meerow & Tombolato, 1996; Meerow et al., 1999), chromosome morphology and C-banding pattern (Buitendijk & Ramanna, 1996) may support the division of *Alstroemeria* species into two groups based on geographic distributions; the Brazilian and the Chilean taxa. However, molecular phylogenetic studies of *Alstroemeria* failed to clarify unambiguously the position and relationships among the studied taxa (Dubouzet et al., 1998; Han et al., 2000; Aagesen & Sanso, 2003; Aros et al., 2006). Here, we therefore present a general pollen survey on *Alstroemeria* based on both light and scanning electron microscopy to search for new characters that could add information pertinent to infrageneric classification of this genus.

Materials and methods

Pollen morphology of 53 taxa (50, out of ca. 60, species, 2 sub-specific taxa and an interspecific hybrid) of *Alstroemeria* was examined with LM and SEM or only with SEM (see ‘List of specimens investigated’). Mature anthers from both dried herbarium specimens (K, MO, MOL, NY, SAPS and USM) and live plants cultivated in the glasshouses of Field Science Center of Northern Biosphere (FSC) of Hokkaido University and National Agricultural Research Centre for Hokkaido Region (NARCHR), Sapporo and Miyake Nursery Ltd., Mobera, Japan, were collected and used to perform the analyses. Abbreviations of the herbarium names are according to the Index Herbariorum (Holmgren et al., 1990).

Before pollen was extracted, fresh anthers were fixed in FAA and anthers from herbarium specimens were rehydrated in acetic acid for more than 24 hours. Pollen studies, both with LM and SEM, were carried out on mature non-acetolysed pollen, with the intention of preserving the integrity of the intine. The pollen was dehydrated in an ethanol series and mounted in silicone oil (viscosity 3000 cs) for LM observations. The following pollen dimensions were measured: polar length (P), equatorial diameter (E) and exine thickness (ET); the P/E ratio was also calculated. The measurements were done under a microscope (Primo Star, Carl Zeiss) with an image analyser, Digital Sight Camera (DS-5M, Nikon). The arithmetic mean, standard deviation and the maximum and minimum values were calculated

after (at least) 30 measurements for each parameter except in *Alstroemeria virdiflora* (Table 1) using the XLSTAT 2009.3 program. The number of pollen grains measured in *A. virdiflora* was less than 10, so no standard deviation was calculated for this species. Principal component analysis (PCA) and agglomerative hierarchical clustering (AHC) were also conducted using the same program to visualise the relationships among the studied species based on pollen data. A dendrogram was built by AHC. Pollen slides of all collections are deposited in the Hokkaido University Museum, Sapporo. Pollen size and shape classes follow Erdtman (1952) and descriptive terminology follows Punt et al. (1994, 2007).

For SEM, pollen grains were dehydrated through an ethanol series, critical point dried with a Hitachi HCP-2 critical point drying apparatus, mounted on aluminum stubs and sputter coated with Gold-Palladium by a Hitachi E102 ion sputter. Subsequently, the prepared pollen was examined and photographed with a Jeol JSM-5310LV scanning electron microscope operated at 15kv. The SEM micrographs of equatorial exine sculpture of similar position were used for the purpose of description and comparison.

Results

Based on LM and SEM observations, pollen grains are monads, large, heteropolar, bilateral; usually distinctly equatorially plano-convex or biconvex, amb elliptical; monosulcate, sulcus

on the convex part of the grains, distinct, long, straight, wide at the equator, narrow near the poles; auriculae-like structure at each end of the sulcus was observed in some taxa under both LM and SEM (Figures 1, 2, Table 1); P: 59.27–75.19 μm , E: 33.80–43.85 μm , P/E: 1.50–1.94, prolate in shape, ET: 1.35–2.70 μm (Table 1).

The exine is tectate or semi-tectate, sculpture commonly striate-reticulate (Type I; Figures 2, 3, 4A–L), with the sculpture elements radiating from the proximal pole. Although a continuous variation is observed in the exine sculpture, two sub-types can be distinguished within the Type I (striae-reticulate) exine sculpture based on the width of muri as follows:

Type Ia: Width of muri is (generally) smaller than 1 μm (Figures 3, 4, 5A–B). Exine sculpture of this sub-type is very common and observed in *Alstroemeria achirae*, *A. amazonica*, *A. andina* ssp. *venustula*, *A. angustifolia*, *A. aurea*, *A. cunha*, *A. diluta* ssp. *diluta*, *A. filipendula*, *A. gardnerii*, *A. guyana*, *A. haemantha*, *A. hookeri*, *A. hookeri* ssp. *recumbens*, *A. hygrophila*, *A. insignis*, *A. kingii*, *A. leporina*, *A. lineatiflora*, *A. monticola*, *A. orchidioides*, *A. paupercula*, *A. pelegrina*, *A. pelegrina* var. *alba*, *A. plantaginea*, *A. polyphylla*, *A. psittacina*, *A. pulchella*, *A. pulchra*, *A. pygmaea*, *A. revoluta*, *A. sellowiana*, *A. stenopetala*, *A. versicolor*, *A. viridiflora* and *A. werdermannii*.

Type Ib: Width of muri is (generally) equal or larger than 1 μm (Figure 5C–I) as observed in *Alstroemeria amabilis*, *A. exerens*, *A. foliosa*, *A. garaventae*, *A. presliana*, *A. umbellata* and *A. zoellneri*.

A few species, viz. *Alstroemeria inodora*, *A. isabellana*, *A. ligtu*, *A. magnifica* var. *magenta*, *A. pallida*, *A. violacea* and *Alstroemeria* hybrid, are also observed with intermediate type of exine sculpture (Type Ia/b; Figures 5J–O, 6A).

Completely striate sculpture is not observed, but sometimes finely to coarsely rugulate-psilate, viz. *Alstroemeria apertiflora*, *A. longistyla*, *A. orchidioides*, *A. spathulata* and *A. versicolor* (Type II; Figure 6B–F), were seen. The striate muri are arranged in a parallel to the sub-parallel pattern. In some species small interconnecting muri are apparent between the striae. The structure results in elongate perforations, and gives a semi-tectate exine structure (e.g., Figure 3H). The exine sculpture along the sulci is similar to that appearing at the equatorial position, but has a relatively flat surface.

In principal component analysis (PCA) using the LM characters, the first and second principal components explain 79.73% of the variance of the sample, 43.94% for the first component – Factor 1; and 35.79% for the second – Factor 2 (Table 2). The Chilean taxa were relatively widely distributed on the principal component plot compared to the Brazilian taxa (Figure 7). Among the species, *Alstroemeria hookeri* and *A. magnifica* var. *magenta* showed the highest values in the first and second component, respectively, and *A. andina* spp. *venustula* and *A. polyphylla*, respectively, showed the lowest values in the first and second component (Figure 7). All four of these taxa are of Chilean origin. The Brazilian taxa were situated at nearer positions (positioned within the open circle in Figure 7), although some of

the Chilean taxa were also included there.

In agglomerative hierarchical clustering (AHC) using quantitative characters, the taxa studied were distributed in three major clusters (Figure 8). Although all of these three clusters included both the Brazilian and Chilean species, Cluster 1 (Brown) mainly comprises Chilean species, but also *Alstroemeria isabellana*, the only Brazilian species included here, and Cluster 3 (Pink) mainly comprises Brazilian species as well as two Chilean taxa, *A. ligtu* and *A. pallida*. Moreover, when we add the exine sculpture data for the AHC analysis, there was only a change in the position of *A. violacea* from Cluster 2 (Green) to Cluster 3 (figure not shown).

Discussion

Variations in pollen morphological characters

All the *Alstroemeria* species examined are characterised by monad, monosulcate and large pollen with striate-reticulate exine sculpture indicating that the genus *Alstroemeria* is a closely related entity. This is in agreement with previous reports (Erdtman, 1952; Heusser, 1971; Schulze, 1978; Kosenko, 1994; Rudall et al., 2000; Sanso et al., 2005; Hofreiter, 2006). However, there are significant differences in the value of quantitative palynological characters that may to some extent be related to differences in the preparation of pollen grains

(Schols et al., 2004), as well as the mounting media (Meltssov et al., 2008 and references therein). We have used non-acetolysed pollen grains mounted in silicone oil for our measurements; however, all other researchers, except Sanso and Xifreda (2001), used acetolysed pollen for their observations and measurements. Although the majority of palynologists use the acetolysis method of Erdtman (1960) or the slightly modified method of Reitsma (1969) for the preparation of pollen grains for LM and SEM observations, these methods themselves also affect the size of pollen grains. The increase of pollen size after acetolysis varies among genera, sometimes even among species. In most instances, acetolysing for three minutes or more does not affect the size significantly (Reitsma, 1969) or between 6 and 30% (Schols et al., 2004). Pollen grains mounted in silicon oil were also smaller than grains mounted in glycerine (Faegri & Iversen, 1975; Meltssov et al., 2008).

Previously, auriculate pollen has been reported only for a few *Bomarea* species (Elsik & Thanikaimoni, 1970; Neuendorf, 1977; Schulze, 1978). We have for the first time observed the presence of auriculae-like structures at each end of the sulcus also in some *Alstroemeria* species (Figures 1, 2, Table 1). However, the majority of species of *Alstroemeria* as well as *Bomarea* have non-auriculate pollen (Table 1; Sanso & Xifreda, 2001).

No correlation between ploidy level and palynological features of *Alstroemeria* has been found. *Alstroemeria ligtu* is tetraploid and all other *Alstroemeria* species are diploids (Tsuchiya et al., 1987), and the pollen grains of *A. ligtu* are similar or sometime smaller than

other diploid species (Table 1). Among the Chilean species, the smallest pollen grains of *A. polypyphylla* may represent its exceptional geographical distribution. *Alstroemeria polypyphylla*, as well as *A. kingii*, is adapted and endemic to the Atacama Desert, one of the driest parts of the world (Hofreiter, 2007). Water availability may affect the pollen morphology, and reduced pollen size resulting from dry soil conditions has been suggested by Stanley and Linskens (1974), related to a general depression of synthesis and growth during onset of meiosis.

Although we did not conduct any correlation studies between floral and pollen morphological characters, there might be a positive correlation between floral diameter and pollen grain size in *Alstroemeria*. For example, *A. revoluta* has tiny flowers with a diameter of about 1 cm while others such as *A. magnifica* and *A. pelegrina*, have large, showy flowers with a diameter of up 7 cm (Sanso et al., 2005). Among the taxa studied, *A. pelegrina* also produces the largest pollen grains (75.19 μm) while *A. revoluta* produces relatively smaller grains (64.35 μm ; Table 1). The largest values of E, P/E and ET are observed in different taxa, viz. *A. hookeri*, *A. andina* ssp. *venustula* and *A. magnifica* var. *magenta*, respectively (Table 1).

Pollen of *Alstroemeria* was described as monosulcate (Kosenko, 1994), but his SEM illustration of *A. pallida* shows possible opercula, although this requires confirmation by TEM. No *Alstroemeria* species with operculate pollen has been observed in this and other

previous studies (Sanso & Xifreda, 2001; Furness & Rudall, 2003; Sanso et al., 2005; Hofreiter, 2006). Furness & Rudall (2003, figs. 2B–D) described *Alstroemeria* pollen as monosulcate with insulae on the aperture membrane; the exine is semitectate columellate in the non-apertural area and solid adjacent to the aperture. Although no difference has been observed in the exine of two *Alstroemeria* species previously studied (Sanso & Xifreda, 2001; Furness & Rudall, 2003), the detail study of exine structure by TEM may find some new characteristics those might be important for both infrageneric classification and phylogenetic studies of *Alstroemeria* and *Alstroemeriaceae* as well. Therefore, it might be an interesting topic for future studies.

Kosenko (1994) has also reported striate exine sculpture in four *Alstroemeria* species. Our and other previous studies (Sanso & Xifreda, 2001; Sanso et al., 2005; Hofreiter, 2006) did not observe any species with striate exine sculpture, though finely to coarsely reticulate-psilate exine sculpture has been observed in rare cases (Figure 6B–F).

The striate-reticulate exine sculpture of pollen might represent an entomophilous (or insect pollinated) mode of pollination in *Alstroemeria* (Dahgren & Clifford, 1982; Aker & Healy, 1990; Bayer, 1998; Hofreiter, 2007). A general relationship between pollen morphology and the pollen vectors of anemophilous and entomophilous species has been known since along ago (Wodehouse, 1935).

Taxonomic significance of palynological features

Inclusion of the monotypic genus *Schickendantzia* in *Alstroemeria* and, in consequence, the resurrection of *A. pygmaea*, has recently been proposed (Sanso & Xifreda, 1999). In morphological, anatomical and karyotype studies, *A. pygmaea* falls clearly within the variation range of *Alstroemeria* (Sanso, 1996, 2002; Sanso & Xifreda, 1999, 2001). The exine sculpture of *A. pygmaea* (Figure 4A) also shares interesting similarities with the *Alstroemeria* pollen examined in the present study (Figures 3, 4, 5A, B) and in previous studies (Kosenko, 1994) that supports inclusion of *A. pygmaea* in *Alstroemeria*.

The pollen morphological data do not support the division of *Alstroemeria* species into two distinct groups, i.e., the Brazilian and Chilean, corresponding to the geographical distribution. Little or no significant differences were observed in all quantitative characters (P, E, P/E ratio and ET) between the Brazilian (P: 59.27–71.24 µm, E: 34.37–39.37 µm, P/E: 1.69–1.92 ET: 1.35–2.31 µm) and the Chilean (P: 61.75–75.19 µm, E: 33.80–43.85 µm, P/E: 1.50–1.94, ET: 1.50–2.70 µm) species groups in either this (Table 1) or the previous studies of *Alstroemeria* pollen (Schulze, 1978; Kosenko, 1994). The smallest pollen grains in the genus are produced by the Brazilian species. Exceptions are *A. virdiflora* and *A. orchidioides* that produce larger pollen. *Alstroemeria orchidioides* also diverges from the other Brazilian species in several exceptional morphological features. For example, it is the only Brazilian

species that produces white flowers, and the hysteranthus condition of *A. orchidioides* flower is very unique, unknown elsewhere in the genus (Meerow et al., 1999, p. 443). The number of pollen grains available from herbarium specimens of the Brazilian taxa was fewer than that of the Chilean taxa. Therefore quantitative characters of some of the Brazilian species (and a few Chilean species) could not be measured in LM (Table 1).

The exine sculpture and other qualitative characters also did not show any distinct difference between these two geographic groups (Figures 3–6, Table 1). The exine sculpture has emerged as one of the valuable characteristics for distinguishing between two closely related genera of the Alstroemeriaceae, *Alstroemeria* and *Bomarea* (Sanso & Xifreda, 2001). Width and arrangement of muri (Sanso & Xifreda, 2001, p. 1067) are found to be useful characteristics at the subgeneric level. Exine sculptures of the taxa from northern Chile are consistently similar, striate-reticulate and muri width less than 1 µm (Type Ia; Figures 3B, D–F, I–K, N, 4C). However, a wide range of variations has been observed in the exine sculpture of Brazilian and central Chilean taxa. In *Alstroemeria*, the exine sculpture units (muri) show a tendency to increase in width towards the reticulate-psilate exine sculpture, or vice versa. The striate-reticulate pollen exine of *Alstroemeria* might be a potential apomorphy in the family Alstroemeriaceae (Aagesen & Sanso, 2003, figure 3).

The dispersion of the Chilean species of *Alstroemeria* on the principal component plot reflects a wider palynological diversity than in the Brazilian species (Figure 7). In PCA, there

are distinct positions of *Alstroemeria hookeri*, *A. magnifica* var. *magenta*, *A. andina* ssp. *venustula* and *A. polypylla*, which may represent their exceptional palynological features, i.e. the highest values of equatorial diameter, exine thickness and P/E, and the lowest value of P/E, respectively (Table 1). It is also noteworthy that the polar length of pollen grains did not directly affect the position of any taxon on the principal component plot; on the contrary, the P/E ratio had a strong influence. The position of the *Alstroemeria* hybrid, within the Chilean taxa, may indicate to the fact that it is an outcome of crossing between two taxa of Chilean origin (Figure 7; I. Miyake, personal communication).

In the AHC dendrogram, Cluster 1, the largest cluster in terms of species number, comprises mainly taxa from central Chile (Hofreiter, 2007) and only one Brazilian species, *Alstroemeria isabellana* (Figure 8). *Alstroemeria aurea* has a wide geographical spread and its ecotypes are believed to be the ancestor of all other Chilean species (Han et al., 2000). Although not found in Brazil, *A. aurea* plants grow on both sides of the Andean Mountains in Argentina, supporting the possibility that *A. aurea* ecotypes were also ancestors of the Brazilian species (Han et al., 2000). Based on *rbcL* data, Aagesen and Sanso (2003, figure 1C) also concluded that *A. aurea*, among other trans-Andean species, may have shared a common ancestor with the Brazilian species. The close similarity between *A. aurea* and *A. isabellana* as observed in the palynological features may support this supposition (Figures 3C, 5O, 8, Table 1). *Alstroemeria isabellana* extensively inhabits also in the mountain marshes

and rough pastures near Portalegre (Rio Grande do Sul), in central and southern Brazil, Argentina, Paraguay and Uruguay (Aker & Healy, 1990; Sanso et al., 2005). The members of Cluster 1 produce commonly large-sized pollen grains (larger than 70.0 µm) except *A. garaventae* and *A. hookeri* (Table 1). Cluster 3, the smallest cluster in terms of species number, is mainly composed of Brazilian species as well as the Chilean taxa *A. ligtu* and *A. pallida*. The inclusion of these two Chilean species might be due to their smaller pollen grains; members of this cluster produce pollen grains of 67.0–69.0 µm in polar length (Table 1).

Based on RAPD markers, Dubouzet et al. (1998) placed the Brazilian species *Alstroemeria inodora* and *A. brasiliensis* in the same group as the Chilean species *A. ligtu*. In their recent study on the estimation of genetic diversity of *Alstroemeria*, Aros et al. (2006) indicated that the Brazilian species *A. psittacina* appeared to have the closest association with both *A. ligtu* ssp. *ligtu* and *A. ligtu* ssp. *incarnata*. Thus, these species were clustered together and could not be separated in a group with other than Chilean species. Even using the morphological descriptors, *A. garaventae* and *A. psittacina* were clustered in a subgroup located further away from the rest of the wild *Alstroemeria* species (Aros et al., 2006). Thus, neither method was able to differentiate *A. psittacina* and other Brazilian species in a separate cluster from the Chilean species group. Moreover, some of the Chilean species showed wide cross-compatibility with not only Chilean but Brazilian species as well (Shinoda & Murata,

2003; Bridgen et al., 2009). *Alstroemeria exserens* and *A. spathulata* showed the lowest genetic distance and were assigned as sister species in an isolated group from the rest of the accessions (Aros et al., 2006). The close relationship between these two taxa is also supported by the palynological features (Figures 5C, 6B, 7, 8, Table 1).

Conclusions

Until now, all attempts to establish an infrageneric classification of *Alstroemeria* have been controversial. Molecular phylogenetic studies of *Alstroemeria* were based mainly on Chilean species, and only a few Brazilian species were included in these studies (Dubouzet et al., 1998; Han et al., 2000; Aagesen & Sanso, 2003; Aros et al., 2006). These studies, moreover, failed to clearly identify the position and relationships among the studied taxa. Here, we have demonstrated that pollen morphological features do not support the division of *Alstroemeria* species into a Brazilian and a Chilean species group corresponding to geographical distribution and the combined analyses of morphological, palynological and molecular data from a larger number of both species and specimens may be useful to identify relationships among the species as well as infrageneric classification of *Alstroemeria*.

Acknowledgements

The authors wish to express their sincere thanks to the directors and curators of the following herbaria: K, MO, MOL, NY, SAPS and USM; to Dr. Koichi Shinoda (NARCHR, Japan) and Mr. Isamu Miyake (Miyake Nursery Ltd., Japan) for allowing us to examine and/or for sending specimens on loan to sample polliniferous material. A. Sarwar is especially grateful to the Japan Society for the Promotion of Science (JSPS) for the Postdoctoral Fellowship for Foreign Researcher during the period of this study. We are also indebted to Prof. Dr. Else Marie Friis and an anonymous reviewer for their helpful comments and suggestions for the improvement of this manuscript.

Specimens investigated

Alstroemeria achirae Munoz-Schick & Brinck – Japan: Chiba, Mobara, cult. Miyake Nursery Ltd., 23.05.2009.

Sarwar & Miyake 09/007 (SAPS) (LM, SEM)

A. amabilis Assis – Brazil: Serra do Quiriri, Rio dos Alemães, Santa Catarina, 14.1.1998, Ribas, Corderio & Barbosa 2252 (MO04760573) (LM, SEM)

A. amazonica Ducke – Brazil: Rondônia, Municipio de Ariquemes, Mineracao Mibrasa, 15.05.1982. Teixeira et al. 470 (MO3581619) (LM, SEM); Venezuela: Prov. Estado Bolivar, Aristeguieta 6583 (NY) (SEM)

A. andina Phil. ssp. *venustula* (Phil.) Bayer – Chile: IV region, Cordillera Dona Ana, Cancha de sky, 06.01.1998. Squeo 88006 (MO4389533) (LM, SEM); Chile: Coquimbo, Elqui, 16.12.1940. Wagenknecht 18123 (MO1294087) (SEM)

A. angustifolia Hook. – Chile: Prov. Valparaiso, Leguna Penuelas, 07.01.1987. Zöllner 15005 (MO3902149)

(SEM)

A. apertiflora Bak. – Paraguay: Depto. A. Parana, Rva. Tati Yupi, Sabana, Compo alto, 27.09.1979. ITAIPU

Binacional 503 (MO3606580) (LM, SEM); Brazil: Santa Catarina. Campo Alegre. Lower fazenda of Ernesto Scheide, 11.12.1956. Smith & Klein 8560 (NY00545614) (SEM)

A. aurea Graham – Chile: Prov. Cautín, Lake Budi., 01.01.1974. Zöllner 8765 (MO2392560) (LM, SEM)

A. brasiliensis Spreng. – Brazil: Minas Gerais, Vel Rio de Janeiro, no date. 1841-1847. Widegren s.n.

(MO4310568) (SEM)

A. cunha Vell. – Brazil: Parque Estadual de Campinhos, Parana, 11.11.1998. Silva & Abe 2624 (MO04765898)

(LM, SEM); Brazil: Flora de Serra dos Orgaos-Correge, 15.07.1940. Brande 16375 (NY00545636) (SEM)

A. diluta Bayer ssp. *diluta* – Japan: Chiba, Mobera, cult. Miyake Nursery Ltd., 23.05.2009. Sarwar & Miyake 09/008 (SAPS) (LM, SEM)

A. exserens Meyen – Chile: Santiago, 16.01.1991. Teillier et al. 2338 (MO4028687) (SEM); Chile: Biobio, Nuble. 18.01.1986. Pederson 14272 (MO4297484) (LM, SEM)

A. filipendula Scub. ex Schenk. – Brazil: Minas Gerais, no date, Martiun s.n. (MO2148619) (SEM)

A. foliosa Mart. – Brazil: Goiás, 17.01.1972. Irwin et al. 34465 (MO2279228) (LM, SEM); Brazil: Prov. Pedra Branca, Minas. Caldas, Hoehne 2882 (NY00910729) (SEM)

A. garaventae Bayer – Japan: Hokkaido, Sapporo, cult. NARCHR, Acc. No. 1661, 04.06.2009. Sarwar & Hoshino 09/028 (SAPS) (LM, SEM)

A. gardneri Baker – Brazil: Brasilia, Mata do rio Tinco, 31.3.1978. Heringer et al. 429 (K) (LM, SEM); Brazil: Goiás, 14.05.1973. Anderson, Hill, dos Santos & Souza 10230 (MO2783259) (SEM)

A. gayana Phil. – Japan: Hokkaido, Sapporo, Hokkaido university campus, cult. FSC, 15.04.2009. Sarwar &

Hoshino 09/032 (SAPS) (LM, SEM)

A. haemantha Ruiz & Pav. – Chile: El Salto, near Valparaiso, 20.11.1965. Meyer 9317 (MO2171359) (LM, SEM)

A. hookeri Lodd. – Japan: Hokkaido, Sapporo, cult. NARCHR, Acc. No. 1295, 04.06.2009. Sarwar & Hoshino 09/24 (SAPS) (LM, SEM)

A. hookeri Lodd. ssp. *recumbens* (Herb.) Bayer – Chile: Atacama, Huasco, 26.10.1938. Worth & Morrison 16223 (MO1575405) (LM, SEM)

A. hygrophila Meerow, Tombolato & Meyer – Brazil: Distrito Federal, 11.10.1978. Heringer et al. 656 (MO3199009) (LM, SEM)

A. inodora Herb. – Brazil: Pará. 04.11.1998. Hatschbach & Silva 68303 (MO04760572) (SEM); Brazil: Rio de Janeiro. Serra dos Orgaos, Parque Nacional. Above Abrigo II, 08.03.1966. Hunt 6489 (NY00545625) (LM, SEM)

A. insignis Kraezlin – Brazil: S. Paulo, Alto de Serra, no day.12.1912. Hoehne 1207 (NY00545627) (LM, SEM)

A. isabellana Herb. – Brazil: Santa Catarina, 24.10.1957. Reitz & Klein 5358 (K) (LM, SEM); Brazil: Pará, 24.11.1980. Hatschbach 43366 (MO3584821) (LM, SEM)

A. kingii Phil. – Chile: Atacama, 02.11.1991. Muñoz et al. 2954 (MO4317596) (LM, SEM)

A. leporina Bayer & Grau – Chile: Coquimbo, 27.10.1991. Muñoz et al. 2679 (MO4317593) (LM, SEM)

A. ligtu L. – Chile: Valparaiso, 17.12.1973. Zöllner 8115 (MO2294521) (LM, SEM)

A. lineatiflora Ruiz & Pav. – Peru: Prov. Recuay, Dpto. Ancash, Carretera Pativilca-Conococha, 27.5.1970. Lopez et al. 7611 (MO2698083) (LM, SEM)

A. longistyla Schenk – Brazil: Distrito Federal, 09.09.1965, Irwin et al. 8137 (NY00910712) (SEM)

A. magnifica Herb. var. *magenta* (Bayer) Muñoz-Schick – Japan: Hokkaido, Sapporo, Hokkaido university campus, cult. FSC, 15.04.2009. Sarwar & Hoshino 09/033 (SAPS) (LM, SEM)

A. monticola Mart. – Brazil: Bahia, Mun. Abaira, 22.01.1992, Hind & Queiroz 50949 (NY00684885) (LM, SEM)

A. orchidioides Meerow, Tombolato & Meyer – Brazil: Distrito Federal, Corrego Landium, N. Brasilia, 09.09.1996. Irwin et al. 18125 (NY00910710) (LM, SEM)

A. pallida Graham – Chile: Santiago, Parque Nacional El Morado, Cordillera De Los Andes, 16.01.1991. Teillier et al. 2339 (MO4028686) (LM, SEM)

A. paupercula Phil. – Chile: Antofagasta, 05.12.1987. Dillon & Teillier 5083 (MO04894342) (LM, SEM)

A. pelegrina Ruiz & Pav. – Japan: Hokkaido, Sapporo, Hokkaido university campus, cult. FSC, 15.04.2009. Sarwar & Hoshino 09/034 (SAPS) (LM, SEM)

A. pelegrina Ruiz & Pav. fo. *alba* Voss – Japan: Hokkaido, Sapporo, Hokkaido university campus, cult. FSC, 15.04.2009. Sarwar & Hoshino 09/035 (SAPS) (LM, SEM)

A. plantaginea Mart. – Brazil: Minas Gerais, Sera do Espinhaco, near summit of Sera da Piedade, 13.01.1971. Irwin et al. 30246 (NY00910707) (LM, SEM)

A. polyphylla Phil. – Chile: Atacama, Huasco, 24.10.1938. Worth & Morrison 16195 (MO5257733) (LM, SEM)

A. presliana Herb. – Japan: Hokkaido, Sapporo, cult. NARCHR, Acc. No. 1687. 26.06.09. Sarwar & Hoshino 09/030 (SAPS) (SEM)

A. psittacina Lehm. – Argentina: Corrientes, Paso de los Libres. 18.11.1973. Lourteig et al. 2721 (MO2321119) (SEM)

A. pulchella L.f. – Brazil: Georgia, cult. Tifton, 25.06.1963. Comte 5115 (MO3180737) (SEM)

A. pulchra Sims – Chile: Valparaiso, 24.10.1976. Zöllner 9220 (MO2415686) (SEM)

A. pygmaea Herb. – Peru: Cusco, 12.12.2004. Farfán et al. 550 (MO6063958) (SEM)

A. revoluta Ruiz & Pav. – Japan: Chiba, Mobera, cult. Miyake Nursery Ltd., 23.05.2009. Sarwar & Miyake 09/013 (SAPS) (LM, SEM)

A. sellowiana Seub. – Brazil: Pará, 15.1.1950. Hatschbach 1876 (MO3148447) (SEM)

A. spathulata Presl. – Chile: Valparaiso, Quillota, 17.11.1991. Billiet & Jadin 5616 (MO4222542) (LM, SEM)

A. stenopetala Schenk – Brazil: Goiás, 20.03.1982. Heringer 18284 (MO3012976) (SEM)

A. umbellata Meyen – Chile: Santiago, Parque Nacional El Morado, Cordillera De Los Andes, 29.12.1990.

Teillier & González 2341 (MO4028685) (LM, SEM)

A. versicolor Ruiz & Pav. – Chile: Valparaiso, 17.12.1973. Zöllner 8113 (MO2294516) (LM, SEM)

A. violacea Phil. – Japan: Chiba, Mobera, cult. Miyake Nursery Ltd., 24.04.2009. Sarwar & Miyake 09/006

(SAPS) (LM, SEM); Peru: Depto. de Arequipa, Atico, 08.11.1947. Ferreyra 2515 (MOL00007301)

(SEM)

A. viridiflora Warm. – Brazil: Minacu, Mata ciliar-Cerradao-transicão, 28.01.1997. Assis et al. 368

(NY00910706) (LM, SEM)

A. werdermannii Bayer – Chile: Región III, Camino Carrizal Bajo a Huasco, desde Los Pozos hasta Tres Playitas,

2.11.1991. Muñoz et al. 2951 (MO4317594) (SEM)

A. zoellneri Bayer – Chile: Valparaiso. 8.12.1991. Zöllner 18162 (MO04665801) (LM, SEM)

Alstroemeria hybrid (Simple rose) – Japan: Chiba, Mobera, cult. Miyake Nursery Ltd., 04.06.2009. Sarwar &

Miyake 09/019 (SAPS) (LM, SEM)

References

Aagesen, L. & Sanso, M. (2003). The phylogeny of the Alstroemeriaceae based on morphology, *rps* 16 intron and *rbcL* sequence data. *Systematic Botany*, 28, 47–69.

Aker, S. & Healy, W. (1990). The phytogeography of the genus *Alstroemeria*. *Herbertia*, 46, 76–87.

Aros, D., Meneses, C. & Infante, R. (2006). Genetic diversity of wild species and cultivated varieties of

- Alstroemeria* estimated through morphological descriptors and RAPD markers. *Scientia Horticulturae*, 108, 86–90.
- Baker, J. G. (1888). *Handbook of the Amaryllideae including Alstroemerieae and Agaveae*. London: Gorge Bell.
- Bayer, E. (1987). Die Gattung *Alstroemeria* in Chile. *Mitteilungen der Botanischen Staatsammlung München*, 24, 1–362. (in German)
- Bayer, E. (1998). Alstroemeriaceae. In K. Kubitzki (Ed.), *Families and genera of vascular plants 3. Flowering plants. Monocotyledons: Lilianae (except Orchidaceae)* (pp. 79–82). Berlin: Springer-Verlag.
- Bridgen, M., Kollman, E. & Lu, C. (2009). Interspecific hybridization of *Alstroemeria* for the development of new ornamental plants. *Acta Horticulturae*, 836, 73–78.
- Buitendijk, J. H. & Ramanna, M. S. (1996). Giemsa C-banded karyotypes of eight species of *Alstroemeria* L. and some of their hybrids. *Annals of Botany*, 78, 449–457.
- Dahgren, R. M. T. & Clifford, H. T. (1982). *Monocotyledons - A comparative study*. New York: Acad. Press.
- Dubouzet, J., Murata, N. & Shinoda, K. (1998). Relationships among some cultivated species and varieties of *Alstroemeria* L. based on RAPD analysis. *Scientia Horticulturae*, 73, 37–44.
- Elsik, W. C. & Thanikaimoni, G. (1970). *Bomarea lyncina* Herb. (Amaryllidaceae) and *Auriculiidites* Elsik. *Pollen Spores*, 12, 177–180.
- Erdtman, G. (1952). *Pollen morphology and plant taxonomy - Angiosperms*. Leiden: E. J. Brill.
- Erdtman, G. (1960). The acetolysis method - a revised description. *Svensk Bot. Tidskr.*, 54, 561–564.
- Faegri, K. & Iversen, J. (1975). Textbook of pollen analysis, (3rd ed. Faegri, K.). Copenhagen: Scandinavian University Books.
- Furness, C. A. & Rudall, P. J. (2003). Apertures with lids: distribution and significance of operculate pollen in monocotyledons. *International Journal of Plant Sciences*, 164, 835–854.
- Han, T., de Jeu, M., Van Eck, H. & Jacobsen, E. (2000). Genetic diversity of Chilean and Brazilian

- Alstroemeria* species assessed by AFLP analysis. *Heredity*, 84, 564–569.
- Heusser, C. J. (1971). *Pollen and spores of Chile*. Tucson: Univ. Arizona Press,
- Hofreiter, A. (2006). *Lenontochir*: A synonym of *Bomarea* (Alstroemeriaceae)? *Harvard Papers in Botany*, 11, 53–60.
- Hofreiter, A. (2007). Biogeography and ecology of the Alstroemeriaceae-Luzuriagaceae clade in high-mountain regions of Central and South America. *Harvard Par. Bot.*, 12, 259–284.
- Holmgren, P. K., Holmgren, N. H. & Barnett, L. C. (Eds). (1990). *Index herbariorum. Part I: The herbaria of the world* (8th Ed.). New York: New York Botanical Garden.
- Hoshino, Y. (2008). Advances in *Alstroemeria* biotechnology. In J. A. T. da Silva (Ed.). *Floriculture, ornamental and plant biotechnology: advances and topical issues*, Vol. 5, pp. 540–547. Middlesex: Global Science Books.
- Kosenko, V. N. (1994). Pollen morphology of the family Alstroemeriaceae. *Botanischesky Zhurnal*, 79, 1–8 (in Russian).
- Meerow, A. W. & Tombolato, A. F. C. (1996). The *Alstroemeria* of Itatiaia. *Herbertia*, 51, 14–21.
- Meerow, A. W., Tombolato, A. F. C. & Meyer, F. (1999). Two new species of *Alstroemeria* L. (Alstroemeriaceae) from Brazil. *Brittonia*, 51, 439–444.
- Meltsov, V., Poska, A. & Saar, M. (2008). Pollen size in *Carex*: the effect of different chemical treatments and mounting media. *Grana*, 47, 220–233.
- Neuendorf, M. (1977). *Pardinae*, a new section of *Bomarea* (Alstroemeriaceae). *Botanical Notiser*, 130, 55–60.
- Punt, W., Blackmore, S., Nilsson, S. & Le Thomas, A. (1994). *Glossary of pollen and spore terminology*. Utrecht: LPP Foundation.
- Punt W, Hoen P. P., Blackmore, S., Nilsson, S. & Le Thomas, A. (2007). Glossary of pollen and spore terminology. *Review of Palaeobotany and Palynology*, 143, 1–81.

- Ravenna, P. (2000). New or interesting Alstroemeriaceae I. *Onira*, 4, 33–46.
- Reitsma, T. (1969). Size modification of recent pollen grains under different treatments. *Review of Palaeobotany and Palynology*, 9, 175–202.
- Rudall, P. J., Stobart, K. L., Hong, W. -P., Conran, J. G., Furness, C. A., Kite, G. C. & Chase, M. W. (2000). Consider the Lilies: systematics of Liliales. In Wilson, K. L. & Morrison, D. A. (Eds). *Monocots: systematics and evolution* (pp. 347–358). Melbourne: CSIRO.
- Sanso, A. M. (1996). El género *Alstroemeria* (Alstroemeriaceae) en Argentina. *Darwiniana*, 34, 349–382.
- Sanso, A. M. (2002). Chromosome studies in Andean taxa of *Alstroemeria* (Alstroemeriaceae). *Botanical Journal of the Linnean Society*, 138, 451–459.
- Sanso, A. M. & Xifreda, C. C. (1999). The synonymy of *Schickendantzia* with *Alstroemeria* (Alstroemeriaceae). *Systematics and Geography of Plants*, 68, 315–323.
- Sanso, A. M. & Xifreda, C. C. (2001). Generic delimitation between *Alstroemeria* and *Bomarea* (Alstroemeriaceae). *Annals of Botany*, 88, 1057–1069.
- Sanso, A. M., Assis, M. C. & Xifreda, C. C. (2005). *Alstroemeria*: a charming genus. *Acta Horticulturae* 683, 63–77.
- Schols, P., Es, K., d'Hondt, C., Merckx, V., Smets, E. & Huysmans, S. (2004). A new enzyme-based method for the treatment of fragile pollen grains collected from herbarium material. *Taxon*, 53, 777–782.
- Schulze, W. (1978). Beiträge zur Taxonomie der Liliifloren III. Alstroemeriaceae. *Wissenschaftliche Zeitschrift der Friedrich-Schiller-Universität Jena/Thüringen. Mathematisch-naturwissenschaftliche Reihe*, 27, 79–85. (in German)
- Shinoda, K. & Murata, N. (2003). Cross-compatibility in interspecific hybridization of fifteen *Alstroemeria* species. *Journal of the Japanese Society of Horticultural Science*, 72, 557–561. (in Japanese with English summary)

- Stanley, R. G. & Linskens, H. F. (1974). *Pollen: biology, biochemistry, management*. Berlin: Springer-Verlag.
- Tsuchiya, T., Hang, A., Healy Jr., W. E. & Hughes, H. (1987). Chromosome studies in the genus *Alstroemeria*. I. Chromosome numbers in 10 cultivars. *Botanical Gazette*, 148, 519–524.
- Uphof, T. C. J. (1952). A review of the genus *Alstroemeria*. *Plant Life (Herbertia)*, 8, 37–53.
- Wodehouse, R. P. (1935). *Pollen grains. Their structure, identification and significance in science and medicine*. New York: McGraw Hill.

Table 1. Variation in pollen characters of *Alstroemeria*; showing mean value and standard deviation (maximum – minimum values in parentheses). Taxa are arranged alphabetically within the group. (A) Pollen grains auriculate; n.d. Not discernible.

Name of Taxa	Polar length (P)	Equatorial diameter (E)	P/E	Exine thickness (ET)	Exine sculpture	Fig. No.
Chilean species group						
<i>A. achirae</i> (A)	70.54±2.20 (77.07 - 66.41)	43.18±1.75 (45.97 - 38.82)	1.63	1.71±0.11 (1.90 - 1.50)	Type Ia	3A
<i>A. andina</i> ssp. <i>venustula</i>	65.74±4.80 (74.93 - 56.81)	33.80±3.13 (43.21 - 28.62)	1.94	1.96±0.15 (2.28 - 1.54)	Type Ia	3B
<i>A. angustifolia</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	-
<i>A. aurea</i>	71.23±2.66 (75.13 - 64.34)	39.22±3.58 (51.02 - 32.81)	1.82	1.48±0.14 (1.90 - 1.30)	Type Ia	3C
<i>A. diluta</i> ssp. <i>diluta</i>	63.65±3.08 (70.45 - 58.29)	39.47±2.37 (45.82 - 35.81)	1.61	1.49±0.17 (1.80 - 1.20)	Type Ia	3D
<i>A. exserens</i>	72.40±3.58 (81.21 - 66.35)	43.27±2.41 (48.67 - 37.53)	1.67	1.61±0.12 (1.80 - 1.43)	Type Ib	5C
<i>A. garaventae</i> (A)	68.19±3.23 (75.94 - 60.85)	42.53±1.55 (45.83 - 40.00)	1.60	1.47±0.13 (1.75 - 1.27)	Type Ib	5D
<i>A. gayana</i>	63.01±2.93 (68.93 - 56.33)	36.53±3.02 (44.64 - 29.92)	1.72	2.42±0.17 (2.72 - 2.12)	Type Ia	3E
<i>A. haemantha</i>	64.95±3.16 (71.57 - 58.53)	40.00±2.58 (47.17 - 33.74)	1.62	2.03±0.16 (2.32 - 1.74)	Type Ia	3F
<i>A. hookeri</i>	66.43±3.58 (74.51 - 58.48)	43.85±4.01 (51.41 - 33.96)	1.51	1.54±0.10 (1.75 - 1.34)	Type Ia	3G
<i>A. hookeri</i> ssp. <i>recumbens</i>	71.28±4.03 (78.52 - 65.40)	42.26±2.76 (46.40 - 37.81)	1.69	1.47±0.08 (1.60 - 1.35)	Type Ia	3H
<i>A. kingii</i>	63.68±2.43 (67.61 - 57.09)	36.73±1.59 (39.42 - 33.51)	1.73	1.76±0.12 (2.12 - 1.60)	Type Ia	3I
<i>A. leporina</i> (A)	73.57±3.42 (80.47 - 66.41)	41.38±2.86 (51.05 - 36.64)	1.78	1.81±0.17 (2.18 - 1.50)	Type Ia	3J
<i>A. ligtu</i> (A)	67.66±2.47 (73.19 - 62.25)	38.31±2.08 (42.74 - 34.29)	1.77	1.81±0.14 (2.12 - 1.53)	Type Ia/b	5J
<i>A. magnifica</i> var. <i>magenta</i>	74.12±3.90 (81.01 - 67.69)	41.14±2.64 (46.34 - 35.92)	1.80	2.70±0.19 (3.44 - 2.01)	Type Ia/b	5K
<i>A. pallida</i> (A)	68.77±2.23 (74.73 - 65.03)	39.70±2.82 (45.96 - 30.72)	1.73	1.50±0.08 (1.70 - 1.35)	Type Ia/b	5L
<i>A. paupercula</i>	71.68±3.08 (76.28 - 66.02)	43.32±2.18 (48.00 - 39.57)	1.73	1.70±0.11 (1.90 - 1.53)	Type Ia	3K
<i>A. pelegrina</i> (A)	75.19±4.00 (82.89 - 68.50)	42.52±1.49 (50.33 - 35.76)	1.77	2.19±0.13 (2.67 - 1.84)	Type Ia	3L
<i>A. pelegrina</i> fo. <i>alba</i>	74.03±4.42 (81.4 - 65.50)	41.09±2.56 (45.2 - 36.86)	1.80	2.64±0.16 (3.29 - 2.04)	Type Ia	3M

Table 1. Cont.

Name of Taxa	Polar length (P)	Equatorial diameter (E)	P/E	Exine thickness (ET)	Exine sculpture	Fig. No.
<i>A. polyphylla</i>	61.75±2.82 (67.55 - 57.24)	41.21±2.77 (46.34 - 35.21)	1.50	1.64±0.11 (1.90 - 1.43)	Type Ia	3N
<i>A. presliana</i> (A)	72.78±3.02 (78.48 - 66.40)	41.22±2.66 (46.41 - 35.90)	1.77	1.52±0.12 (1.75 - 1.27)	Type Ib	5E
<i>A. pulchra</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	3O
<i>A. pygmaea</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4A
<i>A. revoluta</i>	64.35±2.68 (71.66 - 59.39)	39.42±2.36 (44.49 - 34.60)	1.63	1.53±0.11 (1.70 - 1.34)	Type Ia	4B
<i>A. spathulata</i>	71.21±3.41 (80.98 - 65.09)	41.97±2.67 (48.03 - 37.22)	1.70	1.74±0.10 (1.92 - 1.53)	Type II	6B
<i>A. umbellata</i>	70.23±3.29 (78.83 - 65.29)	40.92±3.18 (54.13 - 36.18)	1.72	1.67±0.10 (1.82 - 1.53)	Type Ib	5F
<i>A. versicolor</i>	62.43±2.83 (67.74 - 57.68)	37.20±2.07 (42.52 - 33.22)	1.68	1.58±0.12 (1.80 - 1.27)	Type II	6C
<i>A. violacea</i> (A)	66.17±3.21 (75.51 - 61.67)	39.63±2.73 (45.69 - 35.66)	1.67	1.62±0.11 (1.90 - 1.45)	Type Ia/b	5M
<i>A. werdermannii</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4C
<i>A. zoellneri</i>	70.78±3.05 (75.74 - 64.45)	42.77±3.23 (52.52 - 38.49)	1.65	1.72±0.12 (1.99 - 1.52)	Type Ib	5G
Brazilian species group						
<i>A. amabilis</i>	67.09±4.26 (81.25 - 60.64)	38.64±3.47 (45.16 - 33.31)	1.73	2.31±0.21 (2.60 - 1.80)	Type Ib	5H
<i>A. amazonica</i> (A)	63.15±2.79 (67.78 - 55.67)	34.91±2.49 (40.48 - 30.18)	1.81	1.91±0.16 (2.19 - 1.53)	Type Ia	4D
<i>A. apertiflora</i>	62.79±3.03 (69.17 - 55.96)	37.12±3.28 (43.71 - 29.30)	1.69	1.92±0.14 (2.18 - 1.54)	Type II	6D
<i>A. brasiliensis</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	-
<i>A. cunha</i> (A)	68.87±3.01 (74.73 - 62.47)	39.37±3.71 (47.24 - 33.35)	1.75	2.14±0.15 (2.42 - 1.80)	Type Ia	4E
<i>A. filipendula</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4F
<i>A. foliosa</i>	63.50±2.43 (68.67 - 60.21)	37.71±1.96 (41.29 - 32.96)	1.68	1.87±0.13 (2.12 - 1.54)	Type Ib	5I
<i>A. gardneri</i>	69.02±6.12 (83.80 - 61.84)	37.90±3.38 (44.58 - 31.98)	1.82	2.06±0.43 (2.80 - 1.35)	Type Ia	4G
<i>A. hygrophila</i> (A)	67.38±2.40 (73.07 - 62.32)	37.84±2.19 (43.58 - 34.93)	1.78	1.52±0.17 (1.92 - 1.10)	Type Ia	4H

Table 1. Cont.

Name of Taxa	Polar length (P)	Equatorial diameter (E)	P/E	Exine thickness (ET)	Exine sculpture	Fig. No.
<i>A. inodora</i>	68.01±3.58 (75.11 - 60.49)	38.27±1.89 (42.12 - 34.69)	1.78	1.89±0.14 (2.18 - 1.65)	Type Ia/b	5N
<i>A. insignis</i>	65.46±2.68 (71.12 - 61.48)	36.66±1.69 (40.12 - 33.12)	1.79	1.84±0.13 (2.10 - 1.62)	Type Ia	4I
<i>A. isabellana</i> Reitz & Klein Hatschbach	71.06±5.47 (78.75 - 61.77) 71.24±2.37 (75.04 - 65.66)	39.89±4.30 (44.50 - 31.04) 39.36±2.13 (43.93 - 35.01)	1.78 1.81	2.11±0.31 (2.48 - 1.53) 1.53±0.10 (1.75 - 1.40)	Type Ia/b Type Ia/b	- 5O
<i>A. longistyla</i>	n.d.	n.d.	n.d.	n.d.	Type II	6E
<i>A. monticola</i>	66.12±3.41 (72.46 - 61.75)	34.49±1.84 (37.65 - 32.34)	1.92	1.35±0.06 (1.45 - 1.27)	Type Ia	4J
<i>A. orchidioides</i>	62.00±2.69 (66.04 - 54.72)	34.74±2.97 (42.32 - 30.28)	1.78	1.41±0.10 (1.60 - 1.25)	Type II	6F
<i>A. plantaginea</i>	68.82±2.28 (74.44 - 64.48)	38.16±2.12 (41.49 - 33.14)	1.80	1.68±0.14 (1.98 - 1.50)	Type Ia	4K
<i>A. psittacina</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4L
<i>A. pulchella</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4M
<i>A. sellowiana</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4N
<i>A. stenopetala</i>	n.d.	n.d.	n.d.	n.d.	Type Ia	4O
<i>A. virdiflora</i> (A)	59.27 (60.51 - 57.97)	34.37 (36.10 – 33.00)	1.72	2.06 (2.29 - 1.77)	Type Ia	5A
Peruvian endemic species						
<i>A. lineatiflora</i>	63.84±4.07 (70.91 - 56.77)	38.56±2.44 (43.20 - 34.47)	1.66	1.71±0.16 (2.12 - 1.44)	Type Ia	5B
Interspecific hybrid						
<i>A. hybrid</i>	70.41±3.51 (77.89 - 64.09)	42.41±4.12 (48.62 - 31.06)	1.66	1.51±0.13 (1.80 - 1.25)	Type Ia/b	6A

Table 2. Cumulative variance and eigen vectors of principal component analysis (PCA) using quantitative palynological characters for 41 taxa of *Alstroemeria*.

Principal Component Axis	1	2	3
Cumulative Variance (%)	43.943	79.729	99.859
Characters	Eigen vectors		
P	0.517	0.568	-0.288
E	0.752	0.013	0.071
P/E	-0.396	0.619	-0.467
ET	-0.108	0.542	0.833

P Polar length; E Equatorial diameter; ET Exine thickness

Figure captions

Figure 1. Light micrographs of *Alstroemeria* pollen. **A, B.** Pollen grains without auriculae: **A.** *Alstroemeria cunha* (Silva & Abe 2624, MO04765898); **B.** *Alstroemeria spathulata* (Billiet & Jadin 5616, MO4222542). **C–F.** pollen grains with auriculae at each end of the sulcus: **C.** *Alstroemeria achirae* (Sarwar & Miyake 09/007, SAPS); **D.** *Alstroemeria leporina* (Muñoz et al. 2679, MO4317593); **E.** *Alstroemeria ligtu* (Zöllner 8115, MO2294521); **F.** *Alstroemeria pallida* (Teillier et al. 2339, MO4028686). Scale bars – 10 µm.

Figure 2. Scanning electron micrographs of *Alstroemeria* pollen (whole grain). **A–C.** Pollen grains without auriculae: **A.** *Alstroemeria spathulata* (Billiet & Jadin 5616, MO4222542); **B.** *Alstroemeria aurea* (Zöllner 8765, MO2392560); **C.** *Alstroemeria magnifica* var. *magenta* (Sarwar & Hoshino 09/033, SAPS). **D–F.** Pollen grains with auriculae at each end of the sulcus: **D.** *Alstroemeria pelegrina* (Sarwar & Hoshino 09/034, SAPS); **E.** *Alstroemeria presliana* (Sarwar & Hoshino 09/030, SAPS); **F.** *Alstroemeria amazonica* (Teixeira et al. 470, MO3581619). Scale bars – 10 µm.

Figure 3. Scanning electron micrographs of *Alstroemeria* pollen. **A.** *Alstroemeria achirae* (Sarwar & Miyake 09/007, SAPS). **B.** *Alstroemeria andina* ssp. *venustula* (Squeo 88006,

MO4389533). **C.** *Alstroemeria aurea* (Zöllner 8765, MO2392560). **D.** *Alstroemeria diluta* ssp. *diluta* (Sarwar & Miyake 09/008, SAPS). **E.** *Alstroemeria gayana* (Sarwar & Hoshino 09/032, SAPS). **F.** *Alstroemeria haemantha* (Meyer 9317, MO2171359). **G.** *Alstroemeria hookeri* (Sarwar & Hoshino 09/24, SAPS). **H.** *Alstroemeria hookeri* ssp. *recumbens* (Worth & Morrison 16223, MO1575405). **I.** *Alstroemeria kingii* (Muñoz et al. 2954, MO4317596). **J.** *Alstroemeria leporina* (Muñoz et al. 2679, MO4317593). **K.** *Alstroemeria paupercula* (Dillon & Teillier 5083, MO04894342). **L.** *Alstroemeria pelegrina* (Sarwar & Hoshino 09/034, SAPS). **M.** *Alstroemeria pelegrina* fo. *alba* (Sarwar & Hoshino 09/035, SAPS). **N.** *Alstroemeria polyphylla* (Worth & Morrison 16195, MO5257733). **O.** *Alstroemeria pulchra* (Zöllner 9220, MO2415686). Scale bars – 1 µm.

Figure 4. Scanning electron micrographs of *Alstroemeria* pollen. **A.** *Alstroemeria pygmaea* (Farfán et al. 550, MO6063958). **B.** *Alstroemeria revoluta* (Sarwar & Miyake 09/013, SAPS). **C.** *Alstroemeria werdermannii* (Muñoz et al. 2951, MO4317594). **D.** *Alstroemeria amazonica* (Teixeira et al. 470, MO3581619). **E.** *Alstroemeria cunha* (Brande 16375, NY00545636). **F.** *Alstroemeria filipendula* (Martínez s.n., MO2148619). **G.** *Alstroemeria gardneri* (Heringer et al. 429, K). **H.** *Alstroemeria hygrophila* (Heringer et al. 656, MO3199009). **I.** *Alstroemeria insignis* (Hoehne 1207, NY00545627). **J.** *Alstroemeria monticola* (Hind & Queiroz 50949, NY00684885). **K.** *Alstroemeria plantaginea* (Irwin et al. 30246, NY00910707). **L.**

Alstroemeria psittacina (Lourteig et al. 2721, MO2321119). **M.** *Alstroemeria pulchella* (Comte 5115, MO3180737). **N.** *Alstroemeria sellowiana* (Hatschbach 1876, MO3148447). **O.** *Alstroemeria stenopetala* (Heringer 18284, MO3012976). Scale bars – 1 µm.

Figure 5. Scanning electron micrographs of *Alstroemeria* pollen. **A.** *Alstroemeria virdiflora* (Assis et al. 368, NY00910706). **B.** *Alstroemeria lineatiflora* (Lopez et al. 7611, MO2698083). **C.** *Alstroemeria exserens* (Pederson 14272, MO4297484). **D.** *Alstroemeria garaventae* (Sarwar & Hoshino 09/028, SAPS). **E.** *Alstroemeria presliana* (Sarwar & Hoshino 09/030, SAPS). **F.** *Alstroemeria umbellata* (Teillier & González 2341, MO4028685). **G.** *Alstroemeria zoellneri* (Zöllner 18162, MO04665801). **H.** *Alstroemeria amabilis* (Ribas et al. 2252, MO04760573). **I.** *Alstroemeria foliosa* (Irwin et al. 34465, MO2279228). **J.** *Alstroemeria ligtu* (Zöllner 8115, MO2294521). **K.** *Alstroemeria magnifica* var. *magenta*, **L.** *Alstroemeria pallida* (Teillier et al. 2339, MO4028686). **M.** *Alstroemeria violacea* (Sarwar & Miyake 09/006, SAPS). **N.** *Alstroemeria inodora* (Hatschbach & Silva 68303, MO04760572). **O.** *Alstroemeria isabellana* (Hatschbach 43366, MO3584821). Scale bars – 1 µm.

Figure 6. Scanning electron micrographs of *Alstroemeria* pollen. **A.** *Alstroemeria* hybrid (Sarwar & Miyake 09/019, SAPS). **B.** *Alstroemeria spathulata* (Billiet & Jadin 5616, MO4222542). **C.** *Alstroemeria versicolor* (Zöllner 8113, MO2294516). **D.** *Alstroemeria*

apertiflora (ITAIPU Binacional 503, MO3606580). **E.** *Alstroemeria longistyla* (Irwin et al. 8137, NY00910712). **F.** *Alstroemeria orchidioides* (Irwin et al. 18125, NY00910710). Scale bars – 1 μm .

Figure 7. Two-dimensional graph representing species in the PCA. Names of species abbreviated with first three (- four) letters of epithets. Brazilian taxa are positioned mainly within the open circle. \triangle members of Cluster 1, \diamond members of Cluster 2 and \circ members of Cluster 3 in Figure 8.

Figure 8. Dendrogram made from quantitative data by Agglomerative Hierarchical Analysis. Names of species abbreviated with first three (- four) letters of epithets.

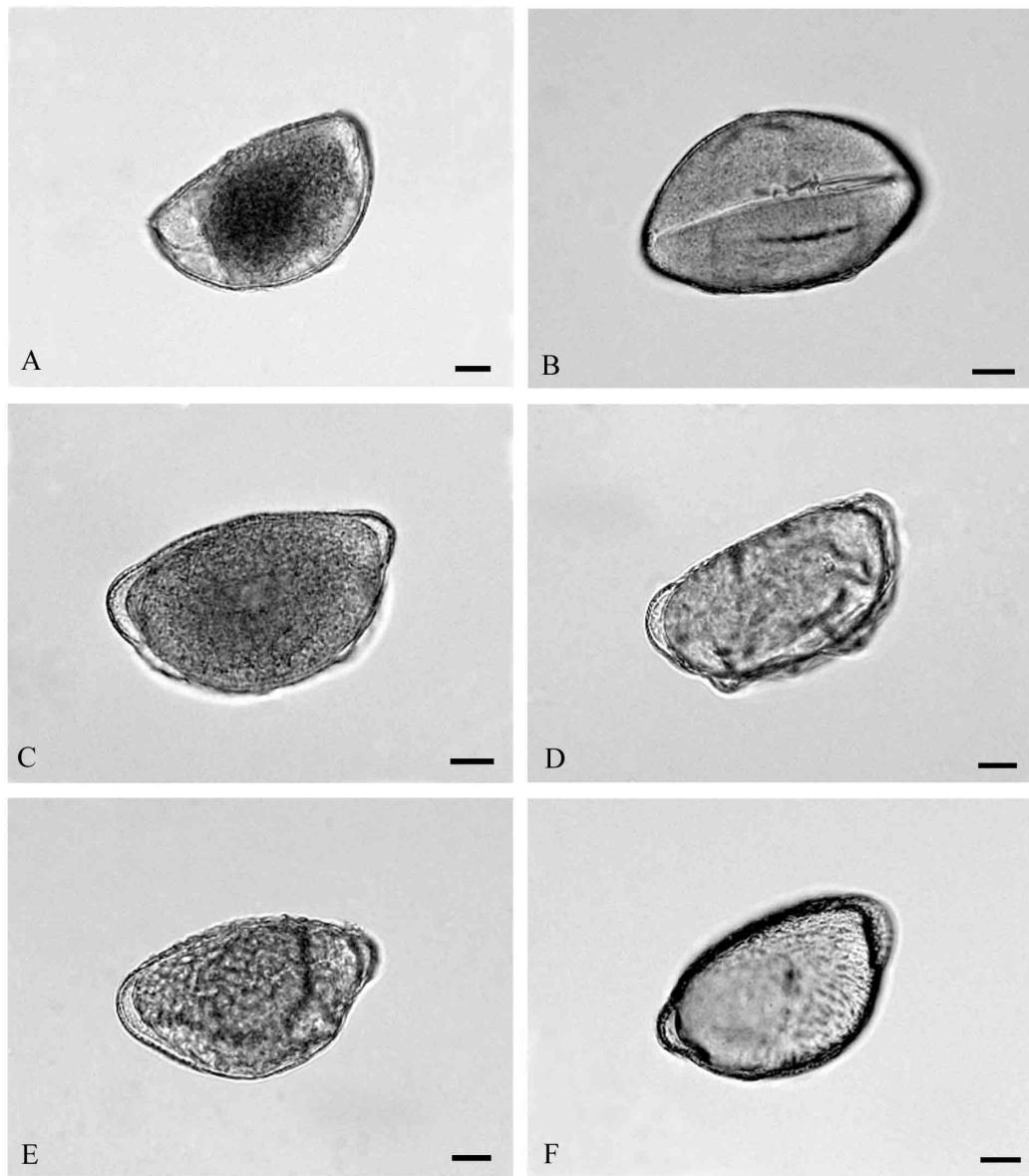


Figure 1.

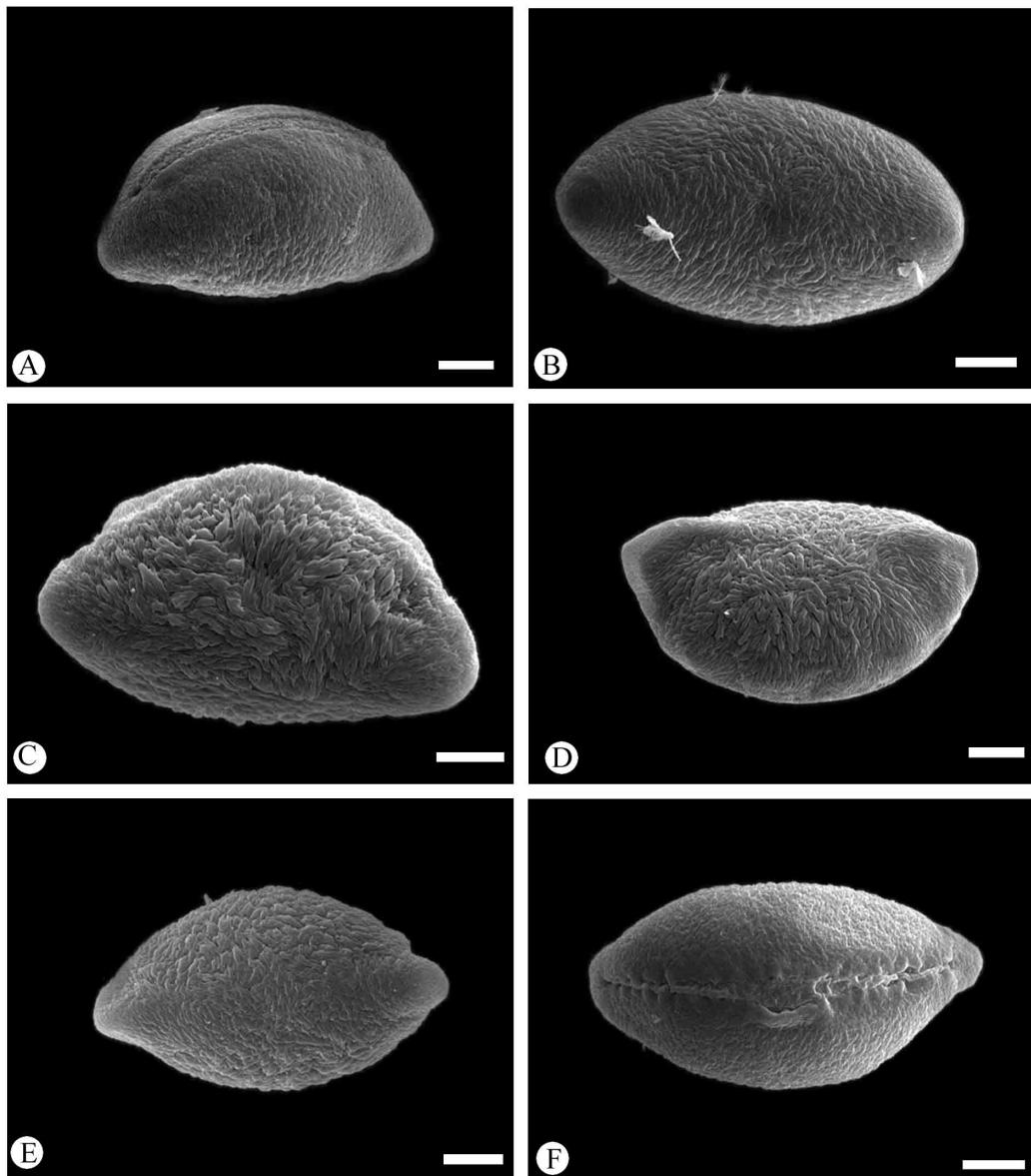


Figure 2.

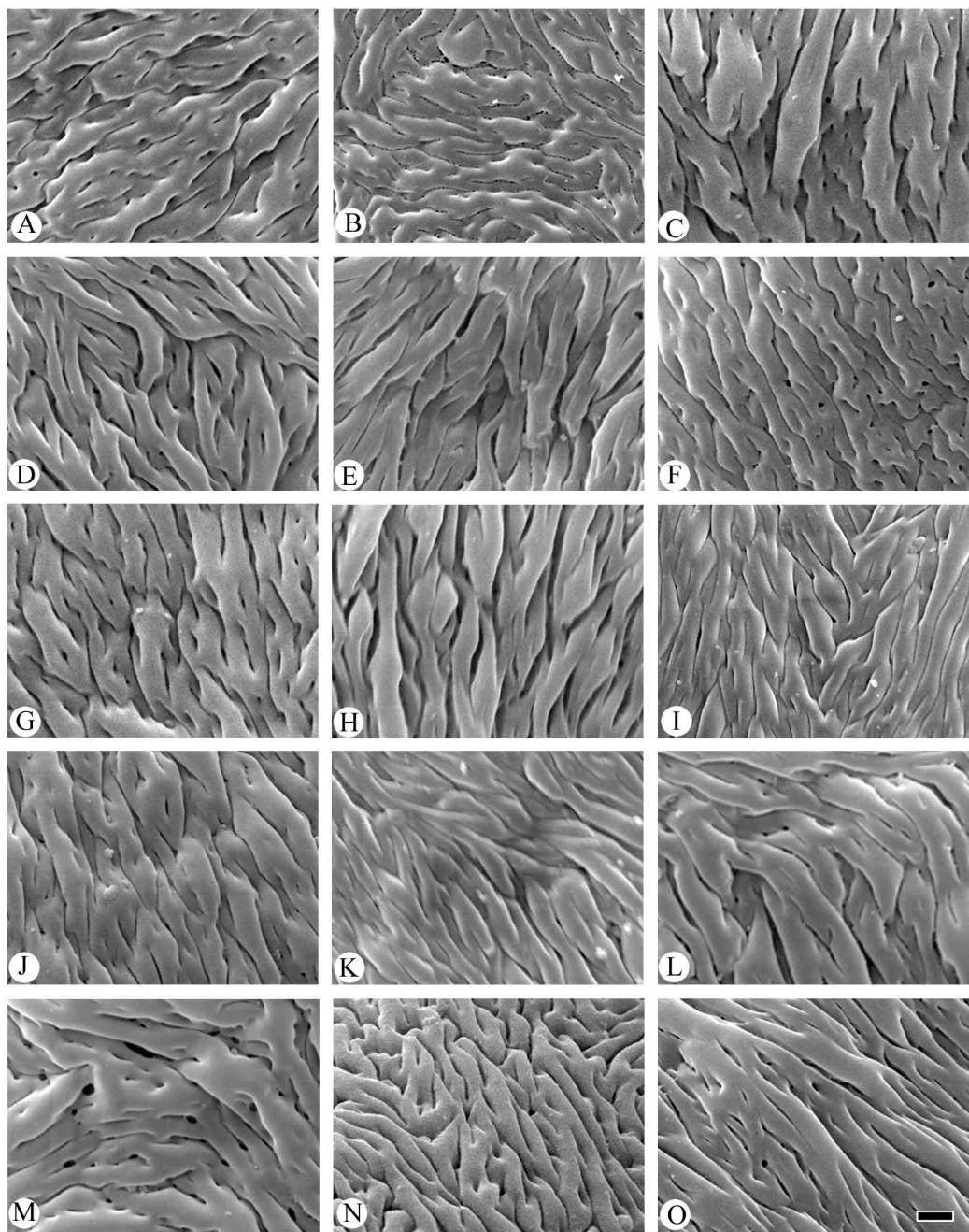


Figure 3.

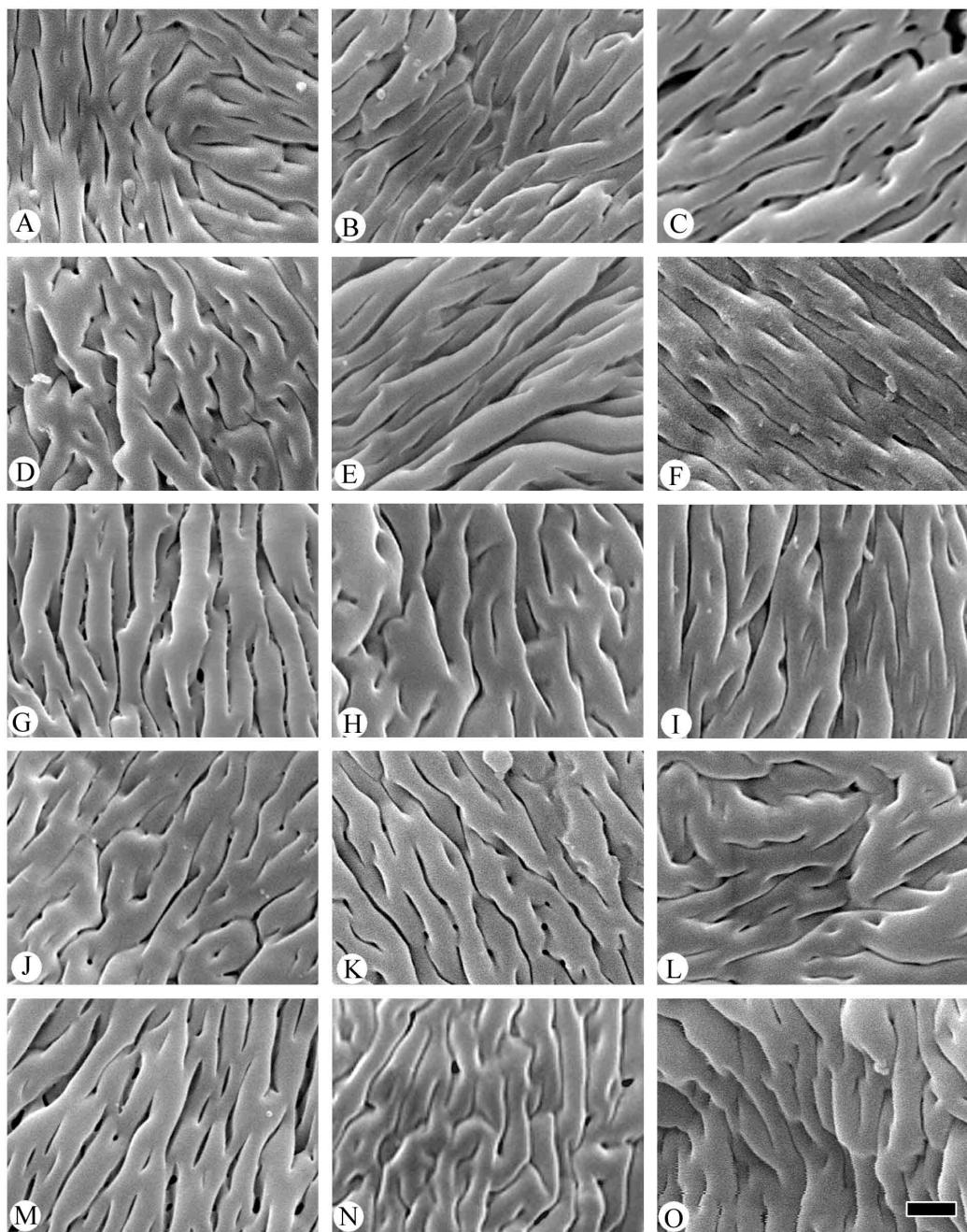


Figure 4.

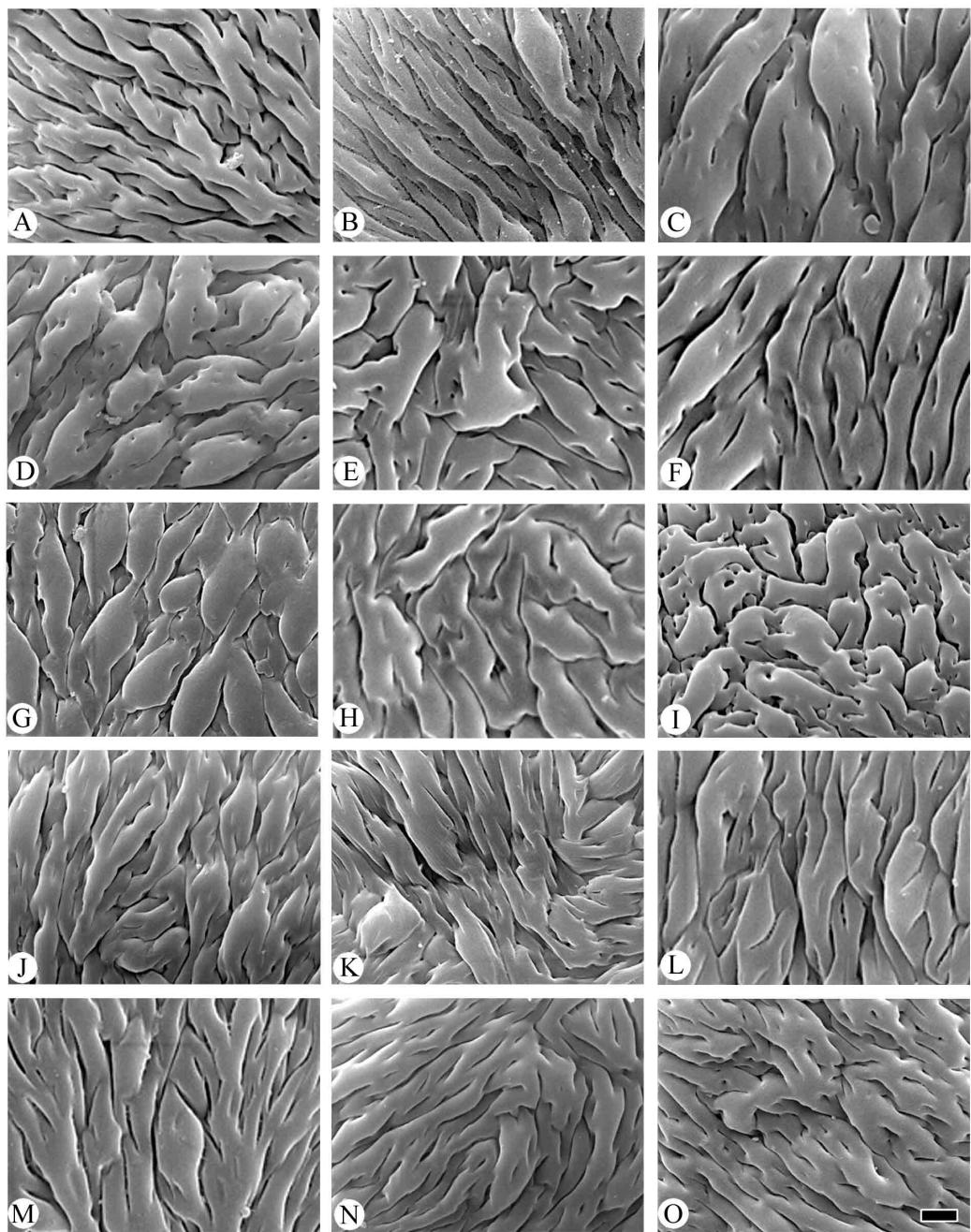


Figure 5.

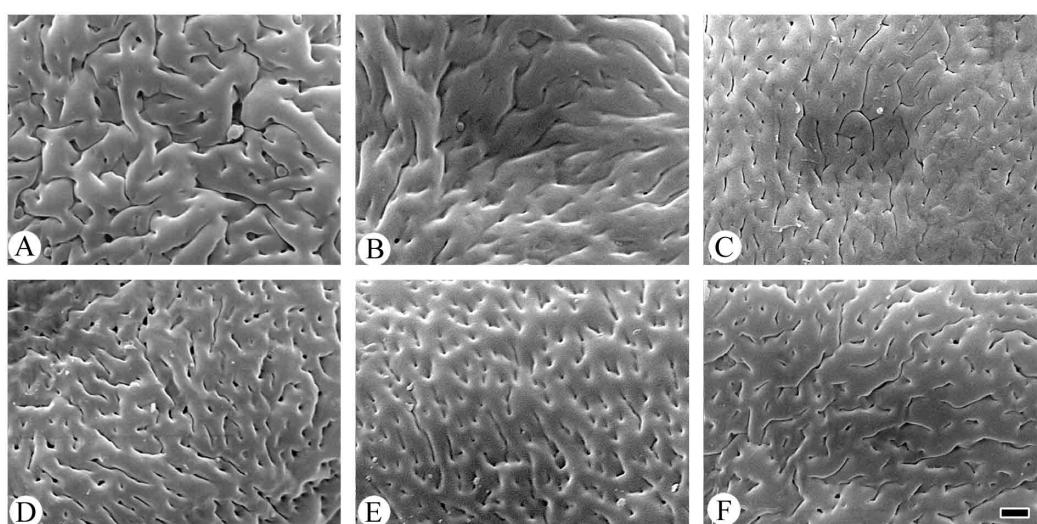


Figure 6.

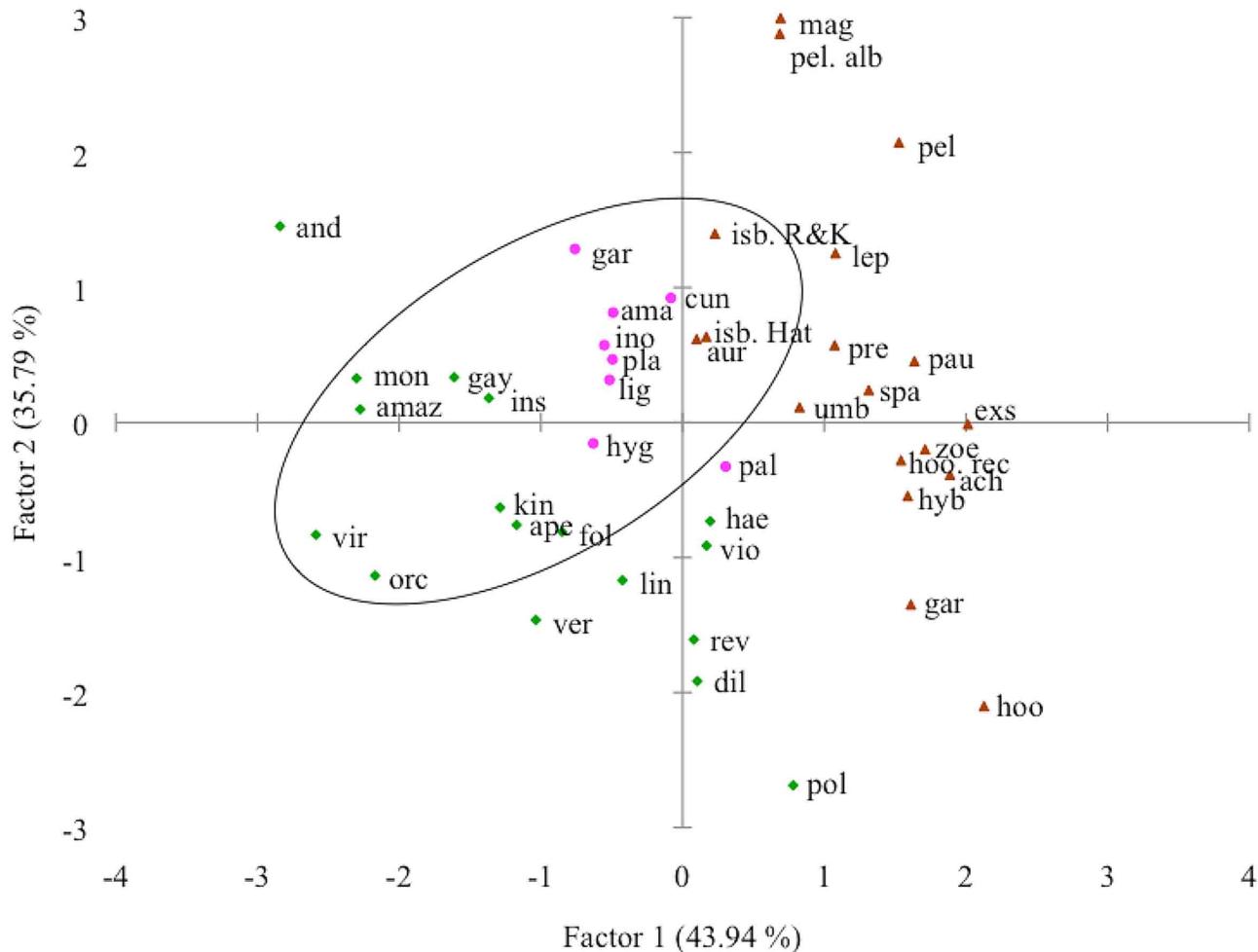


Figure 7.

Dissimilarity

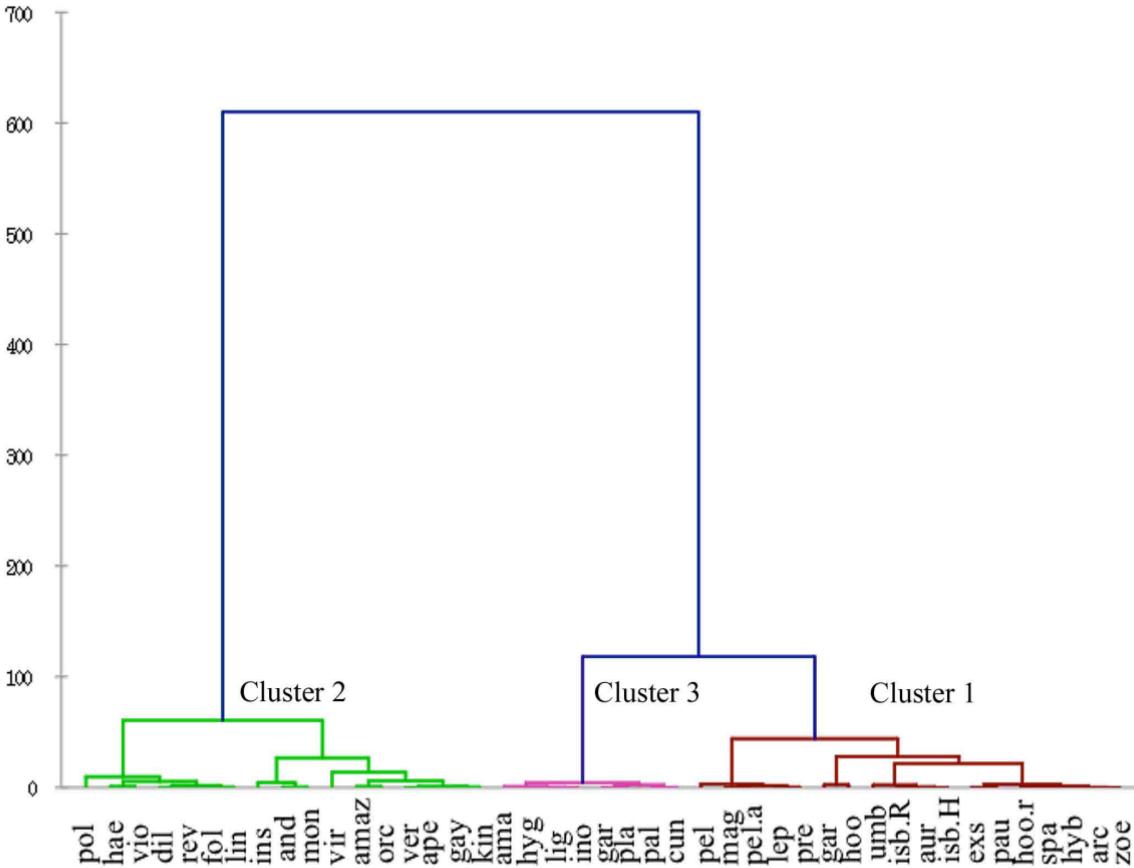


Figure 8.