








# SPORE ATLAS OF ISOSPORATE FERNS OF PUNTA LARA NATURE RESERVE, ARGENTINA

## ÁTLAS DE ESPORAS DE HELECHOS ISOSPORADOS DE LA RESERVA NATURAL PUNTA LARA, ARGENTINA

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### SUMMARY

**Background and aims:** The morphological characteristics of the spores provide information for the identification of fern taxa. To deepen the analyses of the morphological characteristics of the spores of the isosporated ferns inhabiting the Punta Lara Nature Reserve using light microscopy.

**M&M:** The spores of the isosporate ferns that grow in the Punta Lara Nature Reserve (Buenos Aires Province, Argentina) were studied under a light microscope. The study was carried out with fresh materials collected during three years (2016-2018) at different sites of the Reserve. Twenty fern taxa, 16 native and 4 exotic, were analysed. The registered characteristics were: colour, shape, major and minor equatorial diameters, polar diameter, length and characteristics of the laesura and wall ornamentation.

**Results:** The colour of the spores may be yellow, light brown, dark brown and only in *Equisetum* they are green (chlorophyllous). Most spores are monolete (13 species), other trilete (6 species) and only one species has alete spores. The spore walls may be smooth, wrinkled, folded, verrucated, tuberculated, reticulated and winged.

**Conclusions:** The morphology of the spores allows the identification of the ferns of the Reserve at a generic and specific level and provides information for palaeopalynological and aeropalynological studies and of soil spore banks.

### KEY WORDS

Ferns, light microscope, morphology, Punta Lara Reserve, spores

### RESUMEN

**Introducción y objetivo:** Las características morfológicas de las esporas brindan información para la identificación de taxa de helechos. Profundizar los análisis de las características morfológicas de las esporas de los helechos isosporados que habitan en la Reserva Natural de Punta Lara utilizando microscopía óptica.

**M&M:** Se estudiaron con microscopio óptico las esporas de helechos isosporados que crecen en la Reserva Natural Punta Lara (provincia de Buenos Aires, Argentina). El estudio fue llevado a cabo con material fresco colectado durante 3 años (2016-2018) en diferentes sitios de la Reserva. Se analizaron 20 taxa de helechos, 16 nativos y 4 exóticos. Las características registradas fueron: color, forma, diámetros ecuatoriales mayor y menor, diámetro polar, longitud y características de la lesura y ornamentación de la pared.

**Resultados:** El color de las esporas puede ser amarillo, castaño claro, castaño oscuro y solo en el caso de *Equisetum* son verdes (clorofílicas). La mayoría de esporas son monoletes (13 especies), otras triletes (6 especies) y una sola especie posee esporas aletes. Las paredes de las esporas pueden ser lisas, plegadas, verrucosas, tuberculadas, reticuladas y ruguladas-crestadas.

**Conclusiones:** La morfología de las esporas permite la identificación de los helechos de la Reserva a nivel de género y especie y aporta información para estudios paleopalínológicos, aeropalínológicos y de bancos de esporas del suelo.

### PALABRAS CLAVE

Esporas, helechos, microscopía óptica, morfología, Reserva Punta Lara.

## INTRODUCTION

The Punta Lara Nature Reserve (PLNR) (34°47'33" S & 58°0'28.32" W) is a protected area located on the banks of La Plata River, 12 km north of the city of La Plata (Province of Buenos Aires, Argentina). From the phytogeographical point of view the Reserve is located in the Chaco Domain, Oriental Pampean District of the Pampean Province of the Neotropical Region (Cabrera, 1976; Arana *et al.*, 2017). It covers 6.000 hectares and extends over 8 kilometers of coastline where different environments and communities develop, among them the gallery forests, which constitute the world's southernmost subtropical riparian forests (Cabrera & Dawson, 1944). Guerrero *et al.* (2018) postulated that the marginal forest of Punta Lara is a recently installed association, established in the mid-nineteenth century when the climate of the Río de la Plata region presented an increase in rainfall and minimum temperature, a trend that continues until the present. Currently, the Reserve is the core area of the Pereyra Iraola Biosphere Reserve within the framework of the UNESCO MAB Program (Barbetti, 2008). In this zone, the marginal forests develop on the border of incoming streams formed by La Plata River and comprise arboreal species, shrubs, epiphytes and an herbaceous stratum represented by various species of ferns, among other groups of plants.

The first floristic studies in the Reserve were carried out by Cabrera & Dawson (1944), which described 8 species of isosporated ferns. Later, Moschione (1987) cited 16 species of ferns. More recent investigations conducted by this research group reported the presence of 20 native species of isosporate ferns growing in different habitats of the Reserve (Giudice *et al.*, 2011, 2014). Some exotic species such as *Adiantum capillus-veneris* L., *Christella dentata* (Forssk.) Brownsey & Jermy, *Cyclosorus interruptus* (Willd.) H. Itô and *Pteris tremula* R. Br., are also registered (Giudice *et al.*, 2011; Arana *et al.*, 2020).

Our studies in this area also included the reproductive aspects of some native ferns, as well as the analyzes of soil spore banks as contributions to biodiversity conservation in

this area (Ramos Giacosa *et al.*, 2014, 2017; Luna *et al.*, 2016; Ramos Giacosa *et al.*, 2017; Gorrer *et al.*, 2018).

The spores provide useful systematic characters to distinguish genera, and in some cases species (Tryon & Lugardon, 1991). In particular, the characteristics of the spores under light microscope are a relevant source of information for paleobotanical and paleoecological studies, and also in the aerobiological and forensic fields (Harris, 1957; Vaughn *et al.*, 1990; Farfán-Santillán *et al.*, 2016).

Previous descriptions of the spores of some ferns that grow in Buenos Aires Province are found in Michelena (1989, 1993, 1998) and Giudice (1999). Also, some characteristics of the spores of certain taxa growing in the Reserve were provided during the reproductive studies in the genera *Ctenitis* (Ramos Giacosa *et al.*, 2017), *Microgramma* C. Presl, and *Pleopeltis* Humb. & Bonpl. ex Willd. (Gorrer *et al.*, 2018), *Amauropelta* Kunze, *Christella* H. Lév. and *Goniopteris* C. Presl (= *Thelypteris* Schmidel) (Giudice *et al.*, 2014; Ramos Giacosa *et al.*, 2014).

As a complement of the mentioned works, the objective of this study was to deepen the analyses of the morphological characteristics of the spores of the isosporated ferns inhabiting the Punta Lara Nature Reserve using light microscopy. This information will be useful for systematic, palynological and paleo-environmental studies and conservation in protected areas.

## MATERIAL AND METHODS

The study was carried out with fresh materials collected during three years (2016-2018) at different sites within four cores of the Reserve (Fig. 1). The spores belong to the following 16 native taxa: *Adiantum raddianum* C. Presl, *Amauropelta decurtata* (Link) Salino & T. E. Almeida, *Asplenium claussenii* Hieron., *A. sellowianum* C. Presl ex Hieron., *A. ulbrichtii* Rosenst., *Blechnum auriculatum* Cav., *Ctenitis submarginalis* (Langsd. & Fisch.) Ching, *Doryopteris concolor* (Langsd. & Fisch.) Kuhn,

*D. pentagona* Pic. Serm., *Equisetum giganteum* L., *Gastoniella chaerophylla* (Desv.) Li Bing Zhang & Liang Zhang, *Goniopteris burkartii* C. Chr. ex Abbiatti, *Microgramma mortoniana* de la Sota, *Pleopeltis macrocarpa* (Bory ex Willd.) Kaulf., *P. minima* (Bory) J. Prado & R.Y. Hirai, *Rumohra adiantiformis* (G. Forst.) Ching and 4 exotic species very frequent in the study area: *Adiantum capillus-veneris* L., *Christella dentata* (Forssk.) Brownsey & Jermy, *Cyclosorus interruptus* (Willd.) H. Itô and *Pteris tremula* R. Br. The voucher specimens are deposited in the Herbarium LP of the Museum of Natural Sciences of La Plata (Table 1).

Likewise, the microscopic preparations are preserved in the Laboratorio de Anatomía Comparada, Propagación y Conservación de Embriófitas “Dr. Elías R. de la Sota”, Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata. For the taxa nomenclature, the update of the Catalog of Vascular Plants of the Southern Cone was followed (Zuloaga *et al.*, 2019). The spores were placed on to microscope slides with glycerin jelly without prior treatment and studied and photographed under a Nikon E200 light microscope (LM).

The analyzed characteristics were: color, shape in polar and equatorial view, laesura type and wall ornamentation. The quantitative data refers to the major and minor equatorial diameters, polar diameter and laesura length. The spore’s measures were randomly estimated on 20 spores in each sample. The averages of the 4 quantitative variables were obtained for each species and also for the respective genera and families. With these data, graphics were made to compare the quantitative variables between species, genera and families using Microsoft Excel 2013. The palynological terminology used in the descriptions refers to Tryon & Lugardon (1991). The figures were made using the Corel Draw X3 program.

## RESULTS

The morphological characteristics of the spores of each species are given in Table 2. The spore measurements are given in Table 3. The spores are illustrated in Figs. 2-6.



**Fig. 1.** Study area. The circle include the base station, “El burrito” trail, “Fern ditch”, “El Mirador” Stall, Yabotí Bridge, “Las Cañas” Stream and “La Medialuna” Stall. The triangle symbolizes “La Araucaria” Stall. The rhombus symbolizes “El Ché” trail. The quadrate symbolizes Baldovinos Channel.

The spores showed coloration from yellow, light brown to dark brown, and only in the case of *Equisetum* they were green (chlorophyllous). Most spores are monolete (thirteen), other trilete (six) and only one alete.

The largest spores belong to the family Polypodiaceae (*Microgramma mortoniana* 61.4  $\mu\text{m}$  largest equatorial diameter) and the smallest ones to Pteridaceae (*Doryopteris pentagona*, 28.1  $\mu\text{m}$  and *Pteris tremula* 30.5  $\mu\text{m}$ ).

According to the type of wall ornamentation, we find granular, folded, verrucous, tuberculated, psilate, reticulated and regulated-cristated spores.

Taking into account the morphological characteristics of the spores under LM, a dichotomous key is presented below.

Comparisons regarding the quantitative characteristics (measurements) of the spores are presented in Figs. 7-9.

**Table 1.** Material studied (\*exotics).

<b>Specie</b>	<b>Collector and collection number</b>	<b>Location (PLNR)</b>
<i>Adiantum capillus-veneris*</i>	Berrueta <i>et al.</i> 119, 237	Base station
<i>Adiantum raddianum</i>	Berrueta 176, Berrueta & Gorrer w/n, Berrueta <i>et al.</i> w/n	Base station
<i>Amauropelta decurtata</i>	Berrueta 109, 204	“El Burrito” trail
<i>Asplenium clausenii</i>	Berrueta <i>et al.</i> w/n	“Fern ditch”
<i>Asplenium sellowianum</i>	Berrueta <i>et al.</i> 235, 308	“La Araucaria” Stall
	Berrueta <i>et al.</i> 245	“Fern ditch”
<i>Asplenium ulbrichtii</i>	Berrueta & Gorrer 120, 127	“Fern ditch”
	Berrueta & Gorrer 160	“La Araucaria” Stall
<i>Blechnum auriculatum</i>	Berrueta <i>et al.</i> 508, 509, 510	“Las Cañas” Stream
<i>Christella dentata*</i>	Berrueta 105, 154, 205	“El Burrito” trail
<i>Ctenitis submarginalis</i>	Ramos Giacosa 24	“La Medialuna” Stall
<i>Cyclosorus interruptus*</i>	Berrueta & Gorrer w/n	“El Burrito” trail
<i>Doryopteris concolor</i>	Berrueta <i>et al.</i> 288, 289, 290	“La Araucaria” Stall
<i>Doryopteris pentagona</i>	Ramos Giacosa 30	“Las Cañas” Stream
<i>Equisetum giganteum</i>	Berrueta <i>et al.</i> w/n, Bejar w/n	“El Burrito” trail
<i>Gastoniella chaerophylla</i>	Berrueta 265, Berrueta <i>et al.</i> 337, Berrueta <i>et al.</i> w/n	“El Ché” trail
<i>Goniopteris burkartii</i>	Berrueta 157	“Las Cañas” Stream
	Berrueta 230, 530	Yabotí Bridge
<i>Microgramma mortoniana</i>	Berrueta 215	“El Mirador” Stall
	Berrueta 216	“Fern ditch”
	Bejar w/n	“Las Cañas” Stream
<i>Pleopeltis macrocarpa</i>	Berrueta 217	“Fern ditch”
	Berrueta 218	“Las Cañas” Stream
	Berrueta & Gorrer w/n	“Fern ditch”
<i>Pleopeltis minima</i>	Ramos Giacosa w/n	Baldovinos Channel
<i>Pteris tremula*</i>	Berrueta <i>et al.</i> 139, Berrueta <i>et al.</i> 366, Berrueta w/n	“El Mirador” Stall
<i>Rumohra adiantiformis</i>	Berrueta <i>et al.</i> 625, 626	Baldovinos Channel
	Ramos Giacosa 23	“Las Cañas” Stream

**Key to the identification of the ferns of the PLNR using the characteristics of the spores**

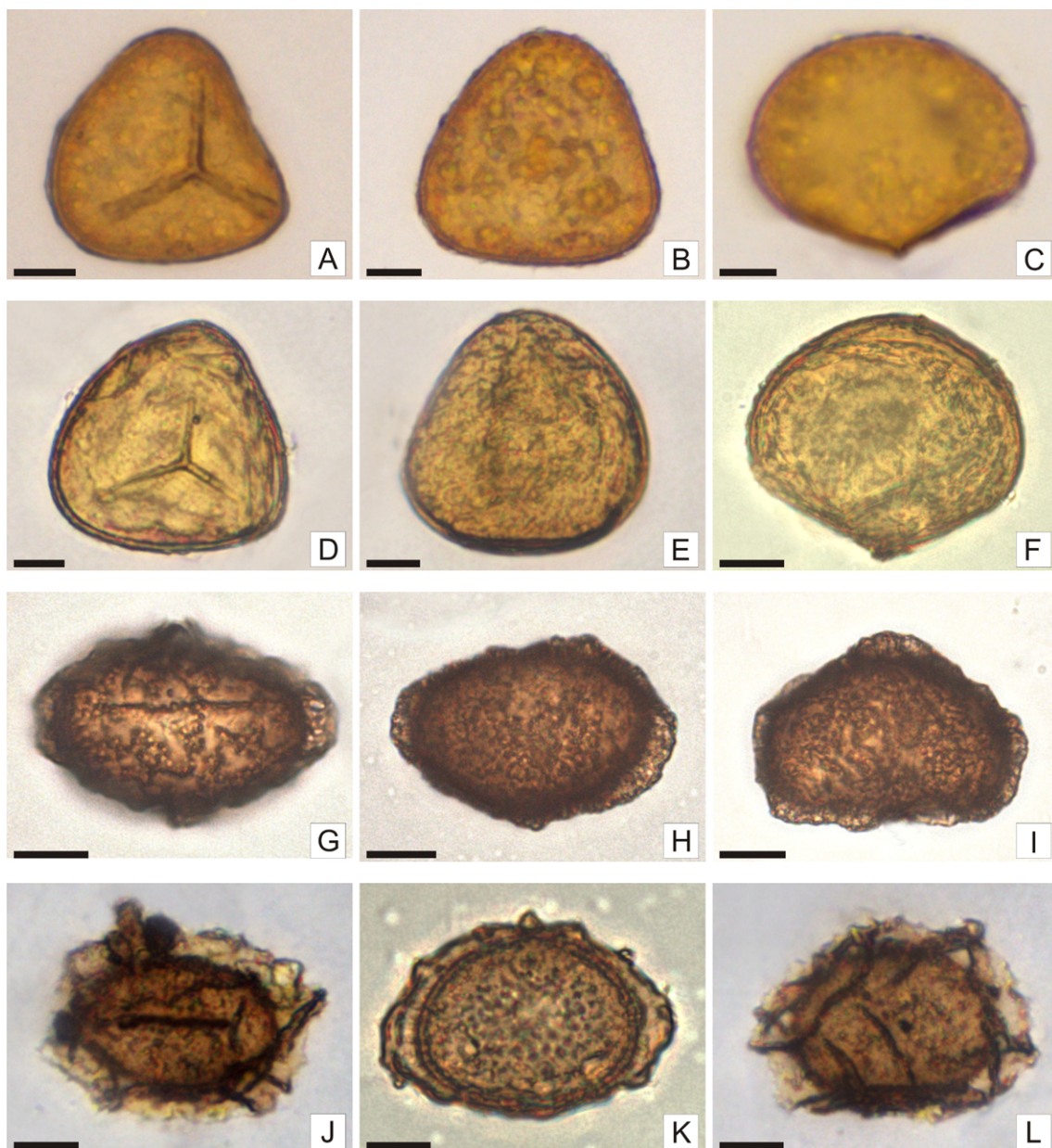
1. Alete spores, with elaters. .... *Equisetum giganteum*
- 1'. Monolete or trilete spores, without elaters.
  2. Monolete spores.
    3. Psilate ornamentation. .... *Blechnum auriculatum*
    - 3'. Verrucate or folded ornamentation.
      4. Verrucate ornamentation.
        5. Evident verrucae. .... *Microgramma mortoniana*
        - 5'. Small verrucae, not clearly evident. .... *Pleopeltis* sp.
      - 4'. Folded ornamentation.
        6. Short and subglobose folds.
          7. Abundant folds, densely distributed. .... *Ctenitis submarginalis*
          - 7'. Scarce folds. .... *Rumohra adiantiformis*
        - 6'. Alate-cristate, reticulate, alate or echinulate-alate folds.
          8. Alate-cristate folds. .... *Asplenium* sp.
          - 8'. Other types of folds.
            9. Reticulate folds. .... *Amauropelta decurtata*
            - 9'. Equinulate-alate or alate folds.
              10. Equinulate-alate folds. .... *Cyclosorus interruptus*
              - 10'. Alate folds.
                11. Folds partially fused. .... *Chrystella dentata*
                - 11'. Folds totally fused. .... *Goniopteris burkartii*
    - 2'. Trilete spores.
      12. With equatorial ridge.
        13. Tuberculate ornamentation. .... *Pteris tremula*
        - 13'. Verrucate ridged ornamentation. .... *Gastoniella chaerophylla*
      - 12'. Without equatorial ridge.
        14. Granular ornamentation. .... *Adiantum* sp.
        - 14'. Rugulate-cristate or psilate ornamentation.
          15. Rugulate-cristate. .... *Doryopteris concolor*
          - 15'. Psilate. .... *Doryopteris pentagona*

Table 2. Morphological characteristics of fern spores of the Punta Lara Nature Reserve (\*exotics).

Taxa	Shape in equatorial view	Shape in polar view	Color	Aperture	Ornamentation
<i>Adiantum capillus-veneris</i> (Fig. 1A-C) *	Trilete (convex/convex) - Monolete (plane-concave/convex)	Trilete: triangular, straight sides and round corners - Monolete: ellipsoidal	Yellow	Trilete/ Monolete	Granulate
<i>Adiantum raddianum</i> (Fig. 1D-F)	Convex-subconical/convex	Triangular, convex sides to subglobose	Yellow	Trilete	Granulate
<i>Amauropelta decurtata</i> (Fig. 1G-I)	Plane/convex	Ellipsoidal	Dark brown	Monolete	Reticulate folds
<i>Asplenium clausenii</i> (Fig. 1J-L)	Plane/convex	Ellipsoidal	Brown	Monolete	Alate-cristate folds
<i>Asplenium sellowianum</i> (Fig. 2A-C)	Convex/hemispheric	Subcircular	Brown	Monolete	Alate-cristate folds
<i>Asplenium ulbrichtii</i> (Fig. 2D-F)	Plane/convex	Ellipsoidal	Brown	Monolete	Alate-cristate folds
<i>Blechnum auriculatum</i> (Fig. 2G-I)	Plane/convex	Ellipsoidal	Yellow	Monolete	Psilate
<i>Christella dentata</i> (Fig. 2J-L) *	Plane/convex	Ellipsoidal	Dark brown	Monolete	Alate folds partially fused
<i>Ctenitis submarginalis</i> (Fig. 3A-C)	Plane/convex	Ellipsoidal	Light brown	Monolete	Short and subglobose folds
<i>Cyclosorus interruptus</i> (Fig. 3D-F) *	Plane/convex	Ellipsoidal	Dark brown	Monolete	Echinulate-alate folds
<i>Doryopteris concolor</i> (Fig. 3G-I)	Convex/convex	Triangular, convex sides to subglobose	Yellow	Trilete	Rugulate-cristate
<i>Doryopteris pentagona</i> (Fig. 3J-L)	Convex/convex	Triangular, convex sides to subglobose	Yellow	Trilete	Psilate
<i>Equisetum giganteum</i> (Fig. 4A-C)	Globose	Globose	Green	Alete	Psilate with elaters
<i>Gastoniella chaerophylla</i> (Fig. 4D-F)	Convex/convex subhemispheric	Triangular, straight to convex sides and round corners	Light brown	Trilete	Verrucate-ridged with equatorial ridge
<i>Goniopteris burkartii</i> (Fig. 4G-I)	Plane/convex	Ellipsoidal	Light brown	Monolete	Alate folds, totally fused
<i>Microgramma mortoniana</i> (Fig. 4J-L)	Plane/convex	Ellipsoidal	Yellow	Monolete	Verrucate
<i>Pleopeltis macrocarpa</i> (Fig. 5A-C)	Plane/convex	Ellipsoidal	Dark yellow	Monolete	Verrucate
<i>Pleopeltis minima</i> (Fig. 5D-F)	Plane/convex	Ellipsoidal	Yellow	Monolete	Verrucate
<i>Pteris tremula</i> (Fig. 5G-I) *	Convex/convex	Triangular, convex sides	Dark brown	Trilete	Tuberculate with equatorial ridge
<i>Rumohra adiantiformis</i> (Fig. 5J-L)	Plane/convex	Ellipsoidal	Brown	Monolete	Folded, short and subglobose folds

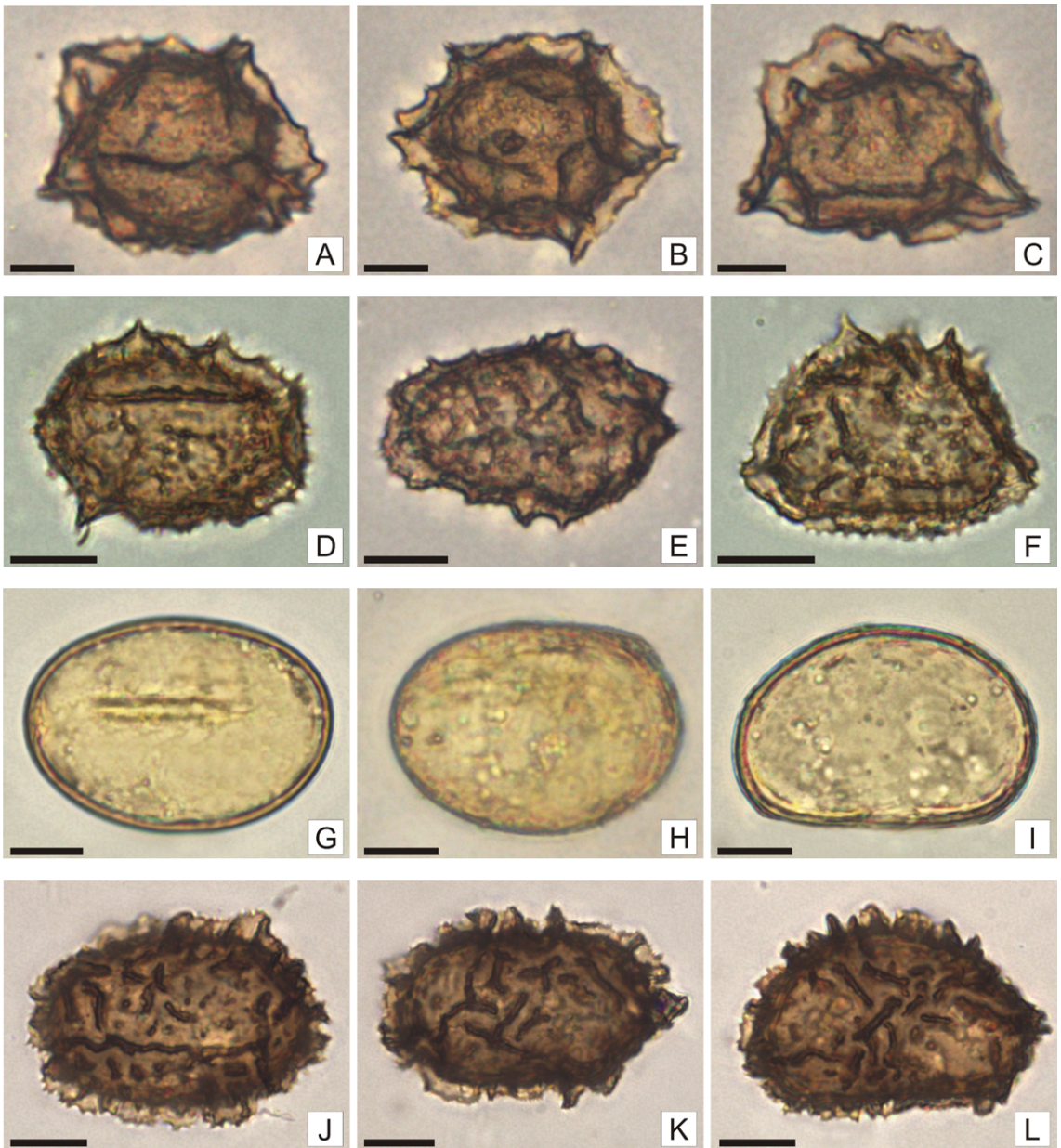
Table 3. Spore measurements. Dimensions in  $\mu\text{m}$ .

Taxa	Major equatorial diameter (DEMa)	Minor equatorial diameter (DEMe)	Polar diameter (DP)	Laesura length (L)
<i>Adiantum capillus-veneris</i>	35 (46.1) 52.5	38.5 (45.8) 52.5	35 (39.2) 45.5	17.5 (20) 24.5
<i>A. raddianum</i>	35 (42.7) 48	38.5 (44.3) 49	31.5 (40.3) 45.5	14 (15.8) 22.8
<i>Asplenium claussenii</i>	28 (37.9) 45.5	21 (26.3) 31.5	24.5 (28.4) 35	14 (15.3) 17.5
<i>A. sellowianum</i>	35 (44.9) 49	27.1 (32.4) 35.4	28 (31.8) 35	14 (24) 31.5
<i>A. ulbrichtii</i>	24 (31.8) 35	20.3 (24.1) 28	21.7 (23.7) 28	16.5 (20.1) 24.5
<i>Blechnum auriculatum</i>	35 (42.1) 49	24.5 (29) 31.5	24.5 (28) 31.5	17.5 (27.6) 35
<i>Christella dentata</i>	35 (39.4) 45.5	22.8 (27.6) 31.5	22.8 (28.4) 31.5	15.8 (24.3) 32.4
<i>Ctenitis submarginalis</i>	35 (41) 49	23.6 (29.9) 42	24.5 (29.7) 42	14 (17.5) 21
<i>Cyclosorus interruptus</i>	45.5 (53.8) 61.3	28 (31.7) 35	28 (32.4) 38.5	28 (35) 42
<i>Doryopteris concolor</i>	24.5 (29.6) 34.1	24.5 (29.6) 35	22.8 (27) 31.5	10.5 (13.7) 15.8
<i>D. pentagona</i>	24.5 (28.1) 31.5	24.5 (27.9) 31.5	24.5 (26) 28	10.5 (12.5) 17.5
<i>Equisetum giganteum</i>	47.3 (51) 56			
<i>Gastoniella chaerophylla</i>	35 (36.6) 40.3	35 (39.3) 42	24.5 (28.6) 33.3	10.5 (14.3) 17.5
<i>Microgramma mortoniana</i>	54.3 (61.4) 73.5	38.5 (42.9) 49	33.3 (39.1) 45.5	28 (36.5) 45.5
<i>Pleopeltis macrocarpa</i>	52.5 (60.6) 68.3	38.5 (44.6) 52.5	33.3 (42.1) 49	28 (35.7) 45.5
<i>P. minima</i>	45.5 (49.7) 59.5	28 (35) 42	29.8 (33.1) 42	24.5 (28.4) 35
<i>Pteris tremula</i>	24.5 (30.5) 35	27.1 (31.2) 35	22.8 (25.7) 28	10.5 (12.8) 17.5
<i>Rumohra adiantiformis</i>	31.5 (33.1) 35	17.5 (23.2) 29.8	17.5 (21) 21.5	15.8 (16.9) 17.5
<i>Goniopteris burkartii</i>	40.3 (47) 53.4	24.5 (32) 42	28 (31) 34.1	28 (35.6) 42
<i>Amauropelta decurtata</i>	33.3 (40.9) 45.5	24.5 (29) 35	24.5 (29.9) 35	24.5 (30) 35

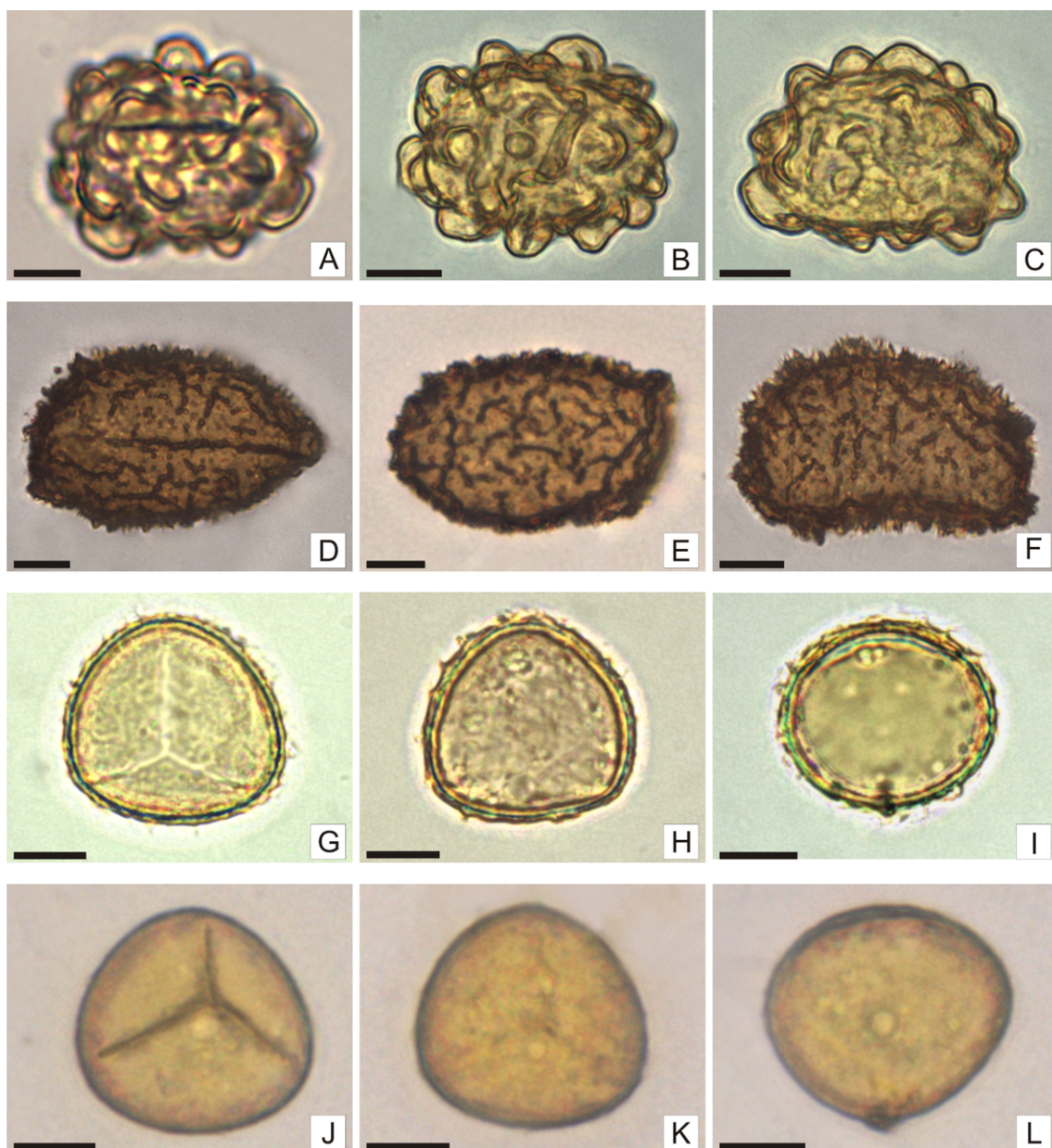


**Fig. 2.** Spores of *Adiantum capillus-veneris*, *Adiantum raddianum*, *Amauropelta decurtata* and *Asplenium clausenii*. **A-C:** *Adiantum capillus-veneris*: **A:** Trilete spore in proximal view, granulate ornamentation. **B:** Spore in distal view with straight sides. **C:** Spore in equatorial view. **D-F:** *A. raddianum*: **D:** Trilete spore in proximal view, granulate ornamentation. **E:** Spore in distal view with convex sides. **F:** Spore in equatorial view with granulate ornamentation. **G-I:** *Amauropelta decurtata*: **G:** Monolete spore in proximal view with reticulate folds. **H:** Spore in distal view with prominent wing-like folds at the edges. **I:** Spore in equatorial view with reticulate folds and wing like-folds at the edges. **J-L:** *Asplenium clausenii*: **J:** Monolete spore in proximal view, alate-cristate folds continuous at the edges. **K:** Spore in distal view with continuous alate-cristate folds along the edges. **L:** Spore in equatorial view with alate-cristate folds on the edges and on the body of the spore. Scales= A-L: 10  $\mu$ m.

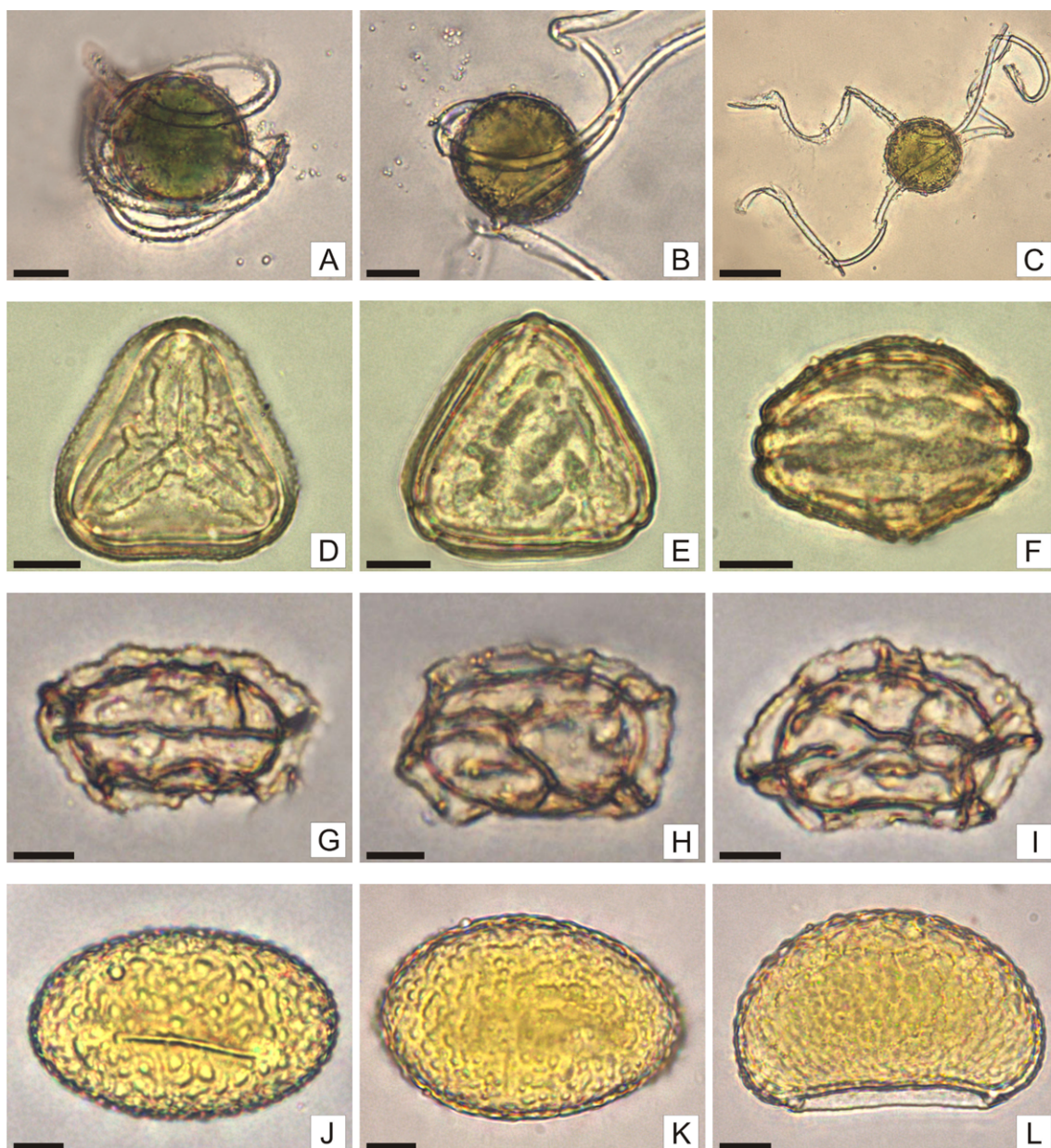




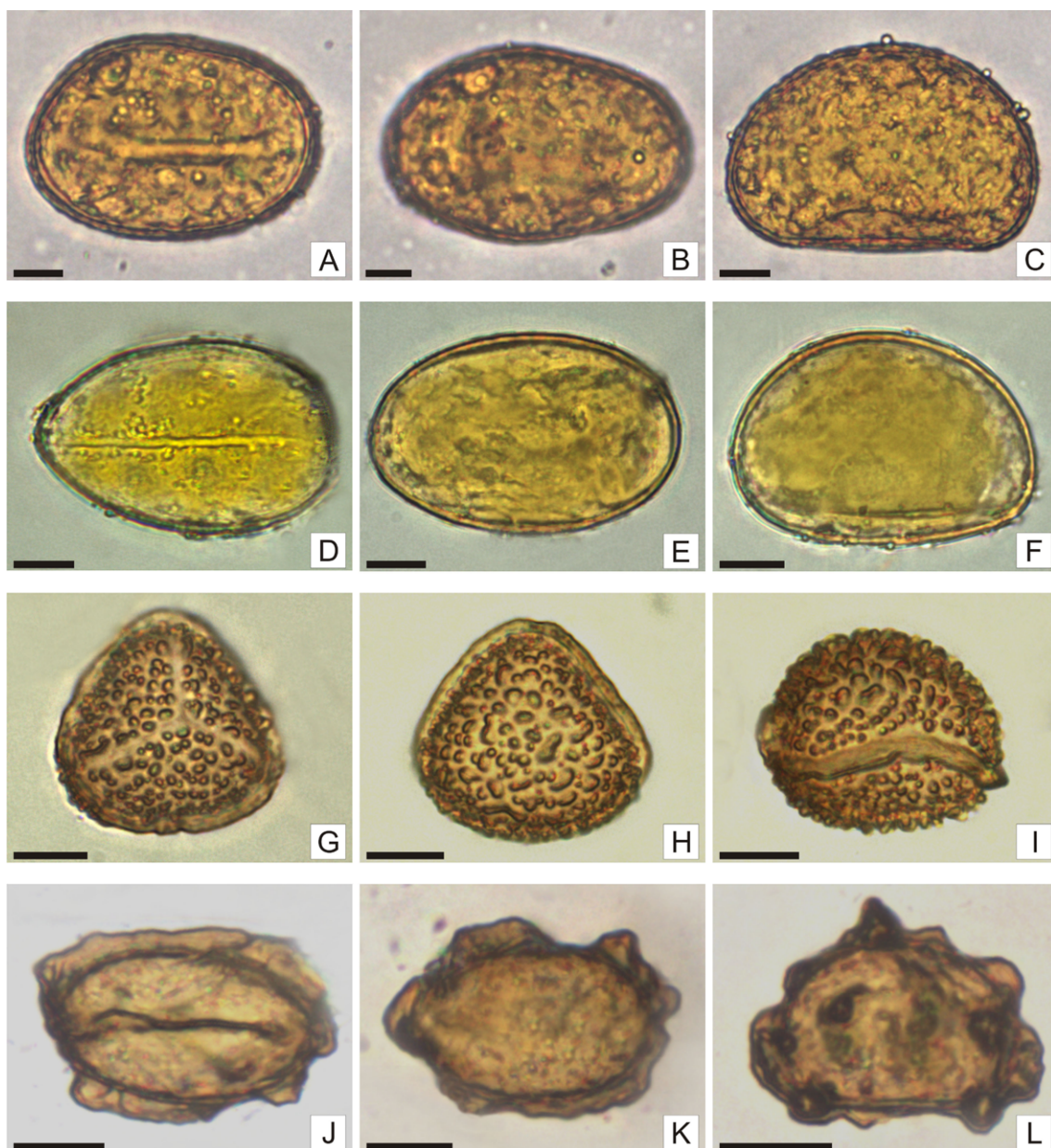
**Fig. 3.** Spores of *Asplenium sellowianum*, *Asplenium ulbrichtii*, *Blechnum auriculatum* and *Christella dentata*. **A-C:** *Asplenium sellowianum*: **A:** Monolete spore in proximal view, coarse alate-cristate folds are observed. **B:** Spore in distal view, coarse alate-cristate folds are observed both, on the edges and on the body of the spore. **C:** spore in equatorial view, coarse alate-cristate folds are observed. **D-F:** *A. ulbrichtii*: **D:** Monolete spore in proximal view, low alate-cristate folds with erose margin are observed. **E:** Spore in distal view, alate-cristate folds with erose margin are observed at the edges and on the body of the spore. **F:** Spore in equatorial view, alate-cristate folds with erose margin are observed. **G-I:** *Blechnum auriculatum*: **G:** monolete spore in proximal view. **H:** Spore in distal view with psilate ornamentation. **I:** Spore in equatorial view. **J-L:** *Christella dentata*: **J:** Monolete spore in proximal view, continuous alate folds are observed at the edges. **K:** Spore in distal view, short alate folds, partially fused in the body of the spore. **L:** Spore in equatorial view, short alate folds and erose margin are observed. Scales= A-L: 10  $\mu$ m.



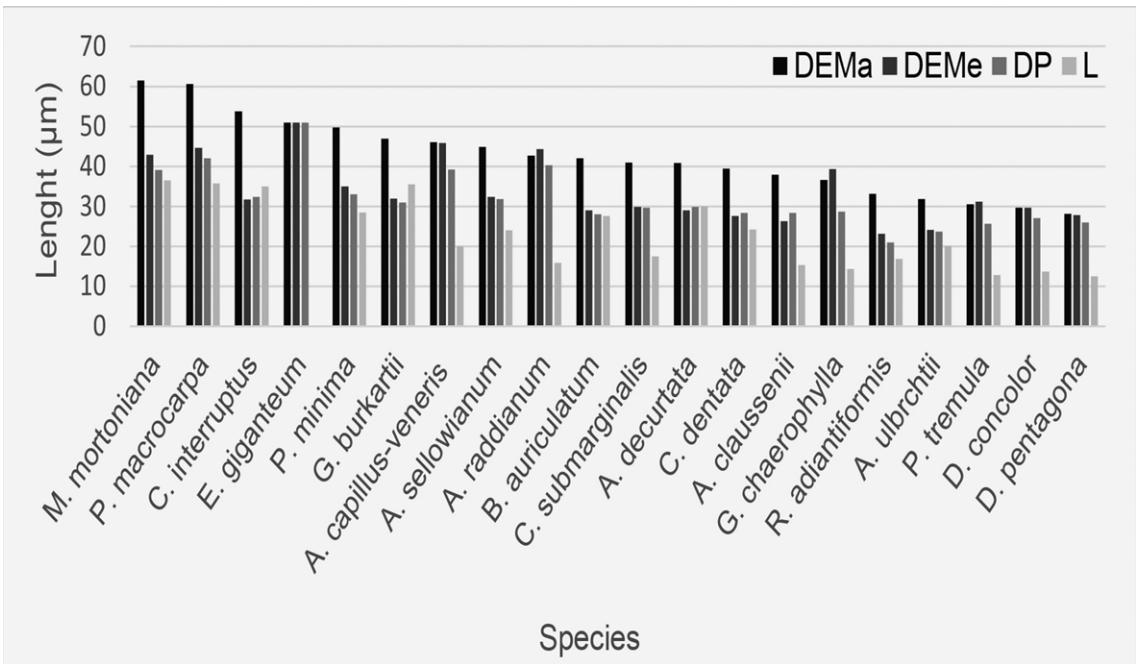
**Fig. 4.** Spores of *Ctenitis submarginalis*, *Cyclosorus interruptus*, *Doryopteris concolor* and *Doryopteris pentagona*. **A-C:** *Ctenitis submarginalis*: **A:** Monolete spore in proximal view, short and subglobose folds are observed. **B:** Spore in distal view, short and subglobose folds are observed over the entire surface. **C:** Spore in equatorial view, short and subglobose folds are observed. **D-F:** *Cyclosorus interruptus*: **D:** Monolete spore in proximal view, echinulate-alate folds are observed. **E:** Spore in distal view, short echinulate-alate folds are observed mainly in the body of the spore. **F:** Spore in equatorial view with short echinulate-alate folds. **G-I:** *Doryopteris concolor*: **G:** Trilete spore in proximal view, a surface with low rugulate-cristate are seen at the edges. **H:** Spore in distal view, low cristae or rugulae form the ornamentation. **I:** Spore in equatorial view, low cristae or rugulae are observed at the edges. **J-L:** *D. pentagona*: **J:** Trilete spore in proximal view, convex sides. **K:** spore in distal view, psilated. **L:** Spore in equatorial view. Scales= A-L: 10  $\mu$ m.



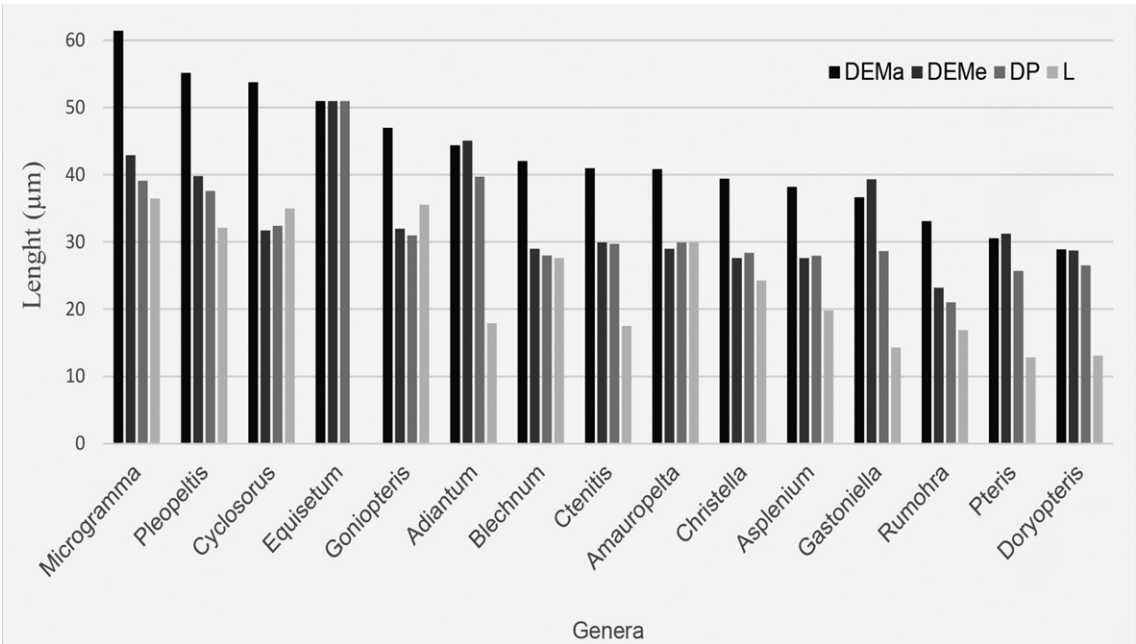
**Fig. 5.** Spores of *Equisetum giganteum*, *Gastoniella chaerophylla*, *Goniopteris burkartii* and *Microgramma mortoniana*. **A-C:** *Equisetum giganteum*: **A:** Alete spore, globose, chlorophyllous, with translucent elaters wound on the spore. **B:** Spore with elaters partially unrolled, psilate. **C:** Spore with elaters totally unrolled. **D-F:** *Gastoniella chaerophylla*. **D:** Trilete spore in proximal view, irregular ridges-verrucae surrounding the laesura. **E:** Spore in distal view, irregular ridges-verrucae are observed. **F:** Spore in equatorial view, an equatorial ridge is observed at the equator. **G-I:** *Goniopteris burkartii*: **G:** Monolete spore in proximal view, alate folds are observed at the edges. **H:** Spore in distal view, alate folds both on the edges and on the body of the spore are observed. **I:** Spore in equatorial view, alate folds totally fused. **J-L:** *Microgramma mortoniana*: **J:** Monolete spore in proximal view, the ornamentation is made up of verrucae. **K:** Spore in distal view, uniformly distributed verrucae are observed. **L:** Spore in equatorial view, verrucae tend to be small towards laesura. Scales= A-B: 20  $\mu$ m; C: 40  $\mu$ m; D-L: 10  $\mu$ m.



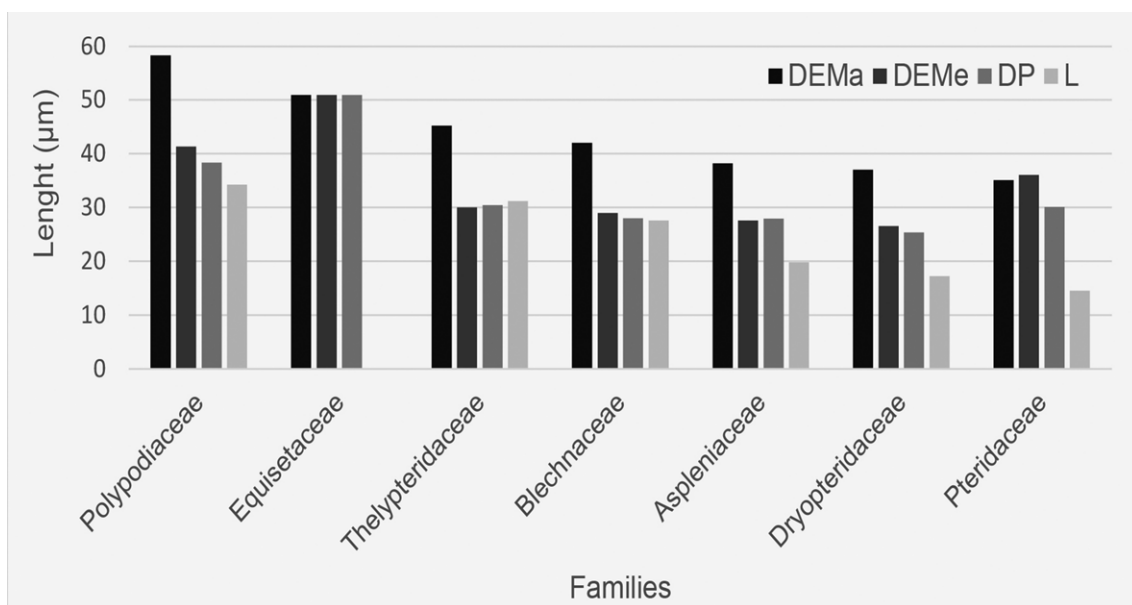
**Fig. 6.** Spores of *Pleopeltis macrocarpa*, *Pleopeltis minima*, *Pteris tremula* and *Rumohra adiantiformis*. **A-C:** *Pleopeltis macrocarpa*: **A:** Monolete spore in proximal view, very low verrucae are observed at the edges. **B:** Spore in distal view, small verrucae, not clearly evident are observed at the edges of the spore. **C:** spore in equatorial view. **D-F:** *P. minima*: **D:** Monolete spore in proximal view, small verrucae not clearly evident are observed at the edges of the spore. **E:** Spore in distal view. **F:** Spore in equatorial view. **G-I:** *Pteris tremula*: **G:** Trilete spore in proximal view, tuberculated. **H:** Spore in distal view, smaller tubercles towards the equator are observed. **I:** Spore in equatorial view, an equatorial ridge is observed at the equator, the tubercles are seen larger towards the poles. **J-L:** *Rumohra adiantiformis*: **J:** Monolete spore in proximal view, continuous folds are observed at the edges. **K:** Spore in distal view, short and long folds are observed at the edges. **L:** Spore in equatorial view, few short, subglobose folds are observed. Scales= A-L: 10  $\mu$ m.



**Fig. 7.** Average spore size by species in the Punta Lara Natural Reserve. Abbreviations= DEMa: major equatorial diameter; DEMe: minor equatorial diameter; DP: polar diameter; L: laesura.



**Fig. 8.** Average size of the spores by genera in the Punta Lara Natural Reserve. Abbreviations= DEMa: major equatorial diameter; DEMe: minor equatorial diameter; DP: polar diameter; L: laesura.



**Fig. 9.** Average spore size by family in the Punta Lara Nature Reserve. Abbreviations= DEMa: major equatorial diameter; DEMe: minor equatorial diameter; DP: polar diameter; L: laesura.

## DISCUSSION AND CONCLUSIONS

The morphological characteristics of the spores allowed the identification of most fern taxa at a specific level, except for the genera *Adiantum*, *Asplenium* and *Pleopeltis*. According to Simabukuro *et al.* (1998, 2000), the spores of related species within these genera have very similar morphological characteristics.

The largest spores belonged to the genera *Microgramma* and *Pleopeltis*, both epiphytic taxa. They also showed verrucate ornamentation. According to Tryon *et al.* (1990), epiphytic fern spores have walls with complex ornamentation.

In the exotic species *A. capillus-veneris* both, trilete and monolete spores, were found. According to Michelena (1989) and Ramos Giacosa (2014) observations in Pteridaceae and *Anemia* respectively, it is common to find different types of spores (trilete, monolete and intermediates) in the same specimen, and also at different stages of development (normal, aborted and immature). These variations were mentioned as characteristic of species where meiosis does not develop

normally, thus achieving apogamy, hybridization and polyploidy (Wagner, 1974; Devi, 1977).

Detailed knowledge of spore morphology is an important tool for paleo-environmental reconstructions (Sánchez-Dzib *et al.*, 2009; Poliakova & Behling, 2016). In this way, the knowledge of current palynological floras allows comparing with fossil ones and to establish relationships between them (Solé de Porta & Murillo-Pulido, 2005).

The spores of *A. clausenii*, *C. dentata*, *C. interruptus*, *D. pentagona*, *G. burkartii* and *P. tremula* are illustrated for the first time under the light microscope.

The information provided here constitutes a contribution to soil spore bank analyses, as well as fossil plant identification and aeropalynological studies, all of which employ spore characteristics for taxonomic identification.

## CONTRIBUTIONS OF AUTHORS

DAG, GEG and JPRG analyzed the data. DAG and JPRG edited the figures and tables. JPRG and MLL revised the language. DAG, PCB and MLL

collected the material. All authors participated in the writing of the manuscript.

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## REFERENCES

- ARANA, M. D., P. C. BERRUETA, D. A. GORRER, G. E. GIUDICE & M. L. LUNA. 2020. *Pteris tremula* (Polypodiopsida: Pteridaceae): A Naturalized Species in Argentina. *Amer. Fern J.* 110: 66-69. <https://doi.org/10.1640/0002-8444-110.2.66>
- ARANA, M. D., G. A. MARTÍNEZ, A. J. OGGERO, E. S. NATALE, & J. J. MORRONE. 2017. Map and shapefile of the biogeographic provinces of Argentina. *Zootaxa* 4341: 420-422. <https://doi.org/10.11646/zootaxa.4341.3.6>
- BARBETTI, C. 2008. Memoria Académica – FaHCE. La reserva natural Punta Lara: Área núcleo de la reserva de la biosfera Pereyra Iraola. Bases para una representación integrada en el espacio costero del Río de la Plata. Available in: [http://www.fuentesmemoria.fahce.unlp.edu.ar/trab\\_eventos/ev.806/ev.806.pdf](http://www.fuentesmemoria.fahce.unlp.edu.ar/trab_eventos/ev.806/ev.806.pdf). [Access: August, 7, 2020].
- CABRERA, A. L. 1976. *Regiones fitogeográficas argentinas. Enciclopedia Argentina de Agricultura y Jardinería*. Acme, Buenos Aires.
- CABRERA, A. L. & G. DAWSON. 1944. La Selva Marginal de Punta Lara en la ribera argentina del Río de la Plata. *Revista Mus. La Plata, Secc. Bot.* 5: 267-382.
- DEVI, S. 1977. Investigations on the surface ultrastructure of some spores of *Asplenium* (Aspleniaceae). *Brenesia* 9: 1-7.
- FARFÁN-SANTILLÁN, N., A. MENDOZA-RUIZ, B. PÉREZ-GARCÍA & E. VELÁZQUEZ-MONTES. 2016. Palinología de las especies mexicanas de Gleicheniaceae. *Bot. Sci.* 94: 281-289. <https://doi.org/10.17129/botsoci.509>
- GIUDICE, G. E. 1999. Sinopsis de las especies argentinas del género *Adiantum* L. (Pteridaceae- Pteridophyta). *Darwiniana* 37: 279-300. <https://doi.org/10.14522/darwiniana.2014.373-4.361>
- GIUDICE, G. E., J. P. RAMOS GIACOSA, M. L. LUNA, A. YÁÑEZ & E. R. DE LA SOTA. 2011. Diversidad de helechos y licofitas de la Reserva Natural Punta Lara, Buenos Aires, Argentina. *Rev. Biol. Trop.* 59: 1037-1046. <https://doi.org/10.15517/RBT.V0I0.3376>
- GIUDICE, G. E., J. P. RAMOS GIACOSA & M. L. LUNA. 2014. Registro de *Thelypteris decurtata* (Link) de la Sota ssp. *platensis* (Weath.) de la Sota en la Reserva Natural Punta Lara, Provincia de Buenos Aires. Un aporte para la conservación de helechos nativos de distribución restringida. *Historia Natural (3era. ser.)* 4: 105-110.
- GORRER, D. A., P. C. BERRUETA, J. P. RAMOS GIACOSA, G. E. GIUDICE & M. L. LUNA. 2018. Morfogénesis de la fase sexual de los helechos epífitos *Microgramma mortoniana* y *Pleopeltis macrocarpa* (Polypodiaceae) de la Reserva Natural Punta Lara, Buenos Aires, Argentina. *Rev. Biol. Trop.* 66: 1078-1089. <https://doi.org/10.15517/RBT.V66I3.31775>
- GUERRERO, E., J. DESCHAMPS & E. TONNI. 2018. La Selva Marginal de Punta Lara ¿relict o colonización reciente? *Revista Mus. La Plata* 3: 348-367. <https://doi.org/10.24215/25456377e064>
- HARRIS, W. F. 1957. A manual of the spores of New Zealand Pteridophyta. *Amer. Fern J.* 49: 114-116. <https://doi.org/10.2307/1545820>
- LUNA, M. L., A. YÁÑEZ, J. P. RAMOS GIACOSA, D. A. GORRER, P. C. BERRUETA & G. E. GIUDICE. 2016. In vitro spore culture and reproductive aspects of the annual fern *Annogramma chaerophylla* (Pteridaceae). *Bol. Soc. Argent. Bot.* 51: 675-682. <https://doi.org/10.31055/1851.2372.v51.n4.16356>
- MICHELENA, I. G. 1989. Esporas de Adiantaceae (Pteridophyta) de la Provincia de Buenos Aires, Argentina. *Bol. de la ALPP* 12: 25-31.
- MICHELENA, I. G. 1993. Esporas de las Aspleniaceae (Pteridophyta) de la provincia de Buenos Aires, Argentina. *Darwiniana* 32: 131-137. <https://www.jstor.org/stable/23222962>
- MICHELENA, I. G. 1998. Esporas de la familia Thelypteridaceae (Pteridophyta) de la Provincia de Buenos Aires, Argentina. *Polen* 9: 25-34.
- MOSCHIONE, F. N. 1987. Ampliación de la distribución de seis especies de Helechos (Pteridophyta, Filicopsida), en la ribera argentina del Río de La Plata. *Garganchillo* 1: 7-13.
- POLIAKOVA, A. & H. BEHLING. 2016. Pollen and fern spores recorded in recent and late Holocene marine sediments from the Indian Ocean and Java

- Sea in Indonesia. *Quatern. Int.* 392: 251-314.  
<https://doi.org/10.1016/j.quaint.2015.06.046>
- RAMOS GIACOSA, J. P. 2014. Abnormal spore morphology and wall ultrastructure in *Anemia tomentosa* var. *anthriscifolia* and *A. tomentosa* var. *tomentosa* (Anemiaceae). *Pl. Syst. Evol.* 300: 1571-1578. <https://doi.org/10.1007/s00606-014-0983-2>
- RAMOS GIACOSA, J. P., G. E. GIUDICE, L. PIPO & M. L. LUNA. 2014. Morfología de las esporas, desarrollo gametofítico y conservación de *Thelypteris abbiattii* y *T. hispidula* (Thelypteridaceae) de la Reserva Natural Punta Lara, Buenos Aires. *Bol. Soc. Argent. Bot.* 49: 217-226.  
<https://doi.org/10.31055/1851.2372.v49.n2.7848>
- RAMOS GIACOSA, J. P., D. A. GORRER, G. E. GIUDICE & M. L. LUNA. 2017. Gametophyte development and conservation of *Ctenitis submarginalis* (Dryopteridaceae) in Buenos Aires Province, Argentina. *Bol. Soc. Argent. Bot.* 52: 33-40.  
<https://doi.org/10.31055/1851.2372.v52.n4.18857>
- SÁNCHEZ-DZIB, Y. A., S. SOSA-NÁJERA & M. S. LOZANO-GARCÍA. 2009. Morfología polínica de especies de la selva mediana subperennifolia en la Cuenca del Río Candelaria, Campeche. *Bol. Soc. Bot. México* 84: 83-104. <https://doi.org/10.17129/botsoci.2296>
- SIMABUKURO, E. A., L. M. ESTEVES & G. M. FELIPPE. 1998. Analysis of a fern spore bank in Southeast Brazil. *Hoehnea*, 25: 45-57.
- SIMABUKURO, E. A., L. M. ESTEVES & G. M. FELIPPE. 2000. Fern spore rain collected at two different heights at Moju Guaçu (São Paulo, Brazil). *Fern Gaz.* 16: 147-166.
- SOLÉ DE PORTA, N. & M. T. MURILLO-PULIDO. 2005. Estudio palino-botánico de algunas especies de Pteridophyta de Colombia. *Rev. Acad. Colomb. Cienc.* 29: 183-2018.
- TRYON, R. M., A. F. TRYON & K. U. KRAMER. 1990. Pteridaceae. In: KRAMER, K. U. & P. S. GREEN (eds.), *The Families and Genera of Vascular Plants: Pteridophytes and Gymnosperms, Vol. 1*, pp. 230-255. Springer-Verlag, Berlin.  
[https://doi.org/10.1007/978-3-662-02604-5\\_42](https://doi.org/10.1007/978-3-662-02604-5_42)
- TRYON, A. F. & B. LUGARDON. 1991. *Spores of the Pteridophyta*. Springer-Verlag, New York.  
<https://doi.org/10.1007/978-1-4613-8991-0>
- VAUGHN, M. B. J., J. G. JONES & D. C. MILDENHALL. 1990. Forensic Palynology in the United States of America. *Palynology* 14: 193-208.  
<https://doi.org/10.1080/01916122.1990.9989380>
- WAGNER, W. H. 1974. Structure of spores in relation to fern phylogeny. *Ann. Missouri Bot. Gard.* 61: 332-353. <https://doi.org/10.2307/2395061>
- ZULOAGA, F. O., M. J. BELGRANO & C. A. ZANOTTI. 2019. Actualización del catálogo de las plantas vasculares del Cono Sur. *Darwiniana* 7: 208-278.  
<https://doi.org/10.14522/darwiniana.2019.72.861>