Available online at www.sciencedirect.com
ScienceDirect
Review of Palaeobotany and Palynology 141 (2006) 245–257
Review of Palaeobotany & Palynology
www.elsevier.com/locate/revpalbo

Morphology and ultrastructure of the spores of Dennstaedtiaceae from North West Argentina

G.E. Giudice [a,*], M.A. Morbelli [b], C.C. Macluf [b], M. Hernández [b], A. Ruiz [b]

[a] *Cátedra de Morfología Vegetal, Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata, Paseo del Bosque s/no, 1900 La Plata, Argentina*

[b] *Cátedra de Palinología, Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata, Paseo del Bosque s/no, 1900 La Plata, Argentina*

Received 22 July 2005; received in revised form 12 April 2006; accepted 27 April 2006
Available online 9 August 2006

Abstract

The spores of Dennstaedtiaceae species that grow in North West Argentina were analysed. These are: *Dennstaedtia cicutaria* (Swartz) Moore, *Dennstaedtia glauca* (Cavanilles) C. Christensen ex Looser, *Dennstaedtia globulifera* (Poiret) Hieronymus, *Hypolepis repens* (L.) C. Presl and *Pteridium psittacinum* (C. Presl) Maxon. LM, SEM and TEM were used in this study. Two types of spores were recognised: monolete spores in *Hypolepis* and trilete spores in *Dennstaedtia* and *Pteridium*. The exospore is homogeneous and apparently double-layered in all the studied spores. According to the plane of section, radial channels, simple or ramified, and others associated to the laesurae are visible in the exospore. The exospore surface is smooth in *Dennstaedtia* species, irregular in *Pteridium* and dentate-papillate in *Hypolepis*. The perispore is single or double-layered with a microlacunose ultrastructure. The perispore layers can be distinguished by their different contrast, structure and thickness. The perispore sculpture varies from echinulate-echinate to verrucate-ridged. General characteristics like spore type, ornamentation and laesurae features as well as ultrastructural ones like the exospore surface and the number of perispore layers could be useful for systematic purposes.

Keywords: Dennstaedtiaceae; Argentina; spores; morphology; sculpture; ultrastructure

1. Introduction

This contribution is part of the study of Pteridophyta spores that grow in North West Argentina. The project is related to studies about Biodiversity in South America and aims to provide palynological data, as a complement to the general knowledge of the species. The study includes the spore analysis of twenty-three families with forty-nine genera quoted for the region. Sixteen families have been published up to this moment. The Cyatheaceae, Dryopteridaceae pp., Pteridaceae pp., Isoetaceae and Heterosporous Filicales, are aspects of PhD Theses currently under development.

The Dennstaedtiaceae taxa that grow in this region, according to Sota (1977) and Ponce (1996), are: *Dennstaedtia cicutaria* (Swartz) Moore, *Dennstaedtia glauca* (Cavanilles) C. Christensen ex Looser, *Dennstaedtia globulifera* (Poiret) Hieronymus, *Hypolepis repens* (L.) C. Presl and *Pteridium psittacinum* (C. Presl) Masón.

Studies in this family were carried out at regional level by Morbelli (1978) who analysed the spores of species that grow in the Patagonian region and

* Corresponding author.
E-mail address: gegiudice@hotmail.com (G.E. Giudice).

0034-6667/$ - see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.revpalbo.2006.04.005

included *Hypolepis* in a key for determination at generic level. Later, Morbelli (1980) analysed the spores of the only species that grows in Patagonia, *Hypolepis rugosula* (Labillardiere) J. Smith var. *poeppigii* (Kunze) C. Christensen et Skottsberg. This author described the spores with the aid of LM, paying special attention to perispore features. Morbelli (1985) presented a brief report of the results of the study of the Dennstaedtiaceae that grow in North West Argentina which was based on observations made with the aid of LM and SEM.

Lugardon (1974) described the sporoderm ultrastructure of the spores of *Dennstaedtia bipinnata* and *Pteridium aquilinum* with the aid of TEM.

Tryon and Lugardon (1991) described with SEM and TEM the spores of *Dennstaedtia cicutaria* based on material from Panamá and Ecuador, *Pteridium aquilinum* var. *arachnoideum* from Brazil and Argentina and *Hypolepis* from New Zealand, New Guinea, Ecuador and Colombia.

In his study on the phylogeny and classification of Dennstaedtiaceae, Mickel (1973) mentioned that the spore characteristics were useful to differentiate some genera. The author commented about the wide range of variation in spore morphology, quoting shapes from tetrahedral to bi-lateral. This author also points out the need of doing careful comparative studies of spores with electron microscopes.

In their study of spores of Pteridophyta of Rio Grande do Sul, Lorscheitter et al. (2002), by using LM and SEM, described spores of *Dennstaedtia globulifera*, *Hypolepis repens* and *Pteridium aquilinum.*

The aim of this study is to analyse the spore morphology and wall ultrastructure of the Dennstaedtiaceae growing in the North West of Argentina. The results will be used for the general morphology, and the ultrastructure will be compared with those in other spore types in other taxa within the Dennstaedtiaceae as well as in other Filicales. Finally, an analysis will be carried out to see if the spore characteristics could be used successfully for systematic purposes.

2. Materials and methods

Spores were obtained from herbarium specimens of Museo de Ciencias Naturales de La Plata (LP). However, difficulties were found in obtaining material of *Dennstaedtia cicutaria* from North West Argentina, since the only specimen quoted by Sota (1977) collected in Salta would be at The New York Botanical Garden, Bronx, New York Herbarium (NY), and according to a personal communication of Dr. Morán it was not found in that institution. In order to solve partly this problem, material of the same species from Misiones Province was processed.

Spores of the different specimens were studied by using light microscope (LM), scanning electron microscope (SEM) and transmission electron microscope (TEM). For LM the spores were treated with 3% hot sodium carbonate for 2 min and acetolyzed according to Erdtman (1960). For SEM the material was treated with 3% hot (3%) sodium carbonate, washed, dehydrated, suspended in 96% ethanol and then transferred to acetate plates. After drying they were coated with gold. All the observations were performed with Olympus BH2 and BHB light microscopes and a JEOL JSMT-100 scanning electron microscope at the Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata.

For studies with TEM, dry material from herbarium specimens was hydrated following the technique proposed by Rowley and Nilsson (1972) that consists of the use of a buffer plus Alcian Blue (AB), then the material was fixed with 1% Glutaraldehyde + 1% Alcian Blue in phosphate buffer for 12 h and post-fixed with 1% OsO_4 in water plus 1% Alcian Blue. The spores were dehydrated in an acetone series and then embedded in Spurr soft mixture. Sections 3 µm thick were stained with toluidine blue and observed with LM. Ultra-thin sections were stained with 1% uranyl acetate for 15 min followed by lead citrate for 3 min. The observations were made with a Zeiss T-

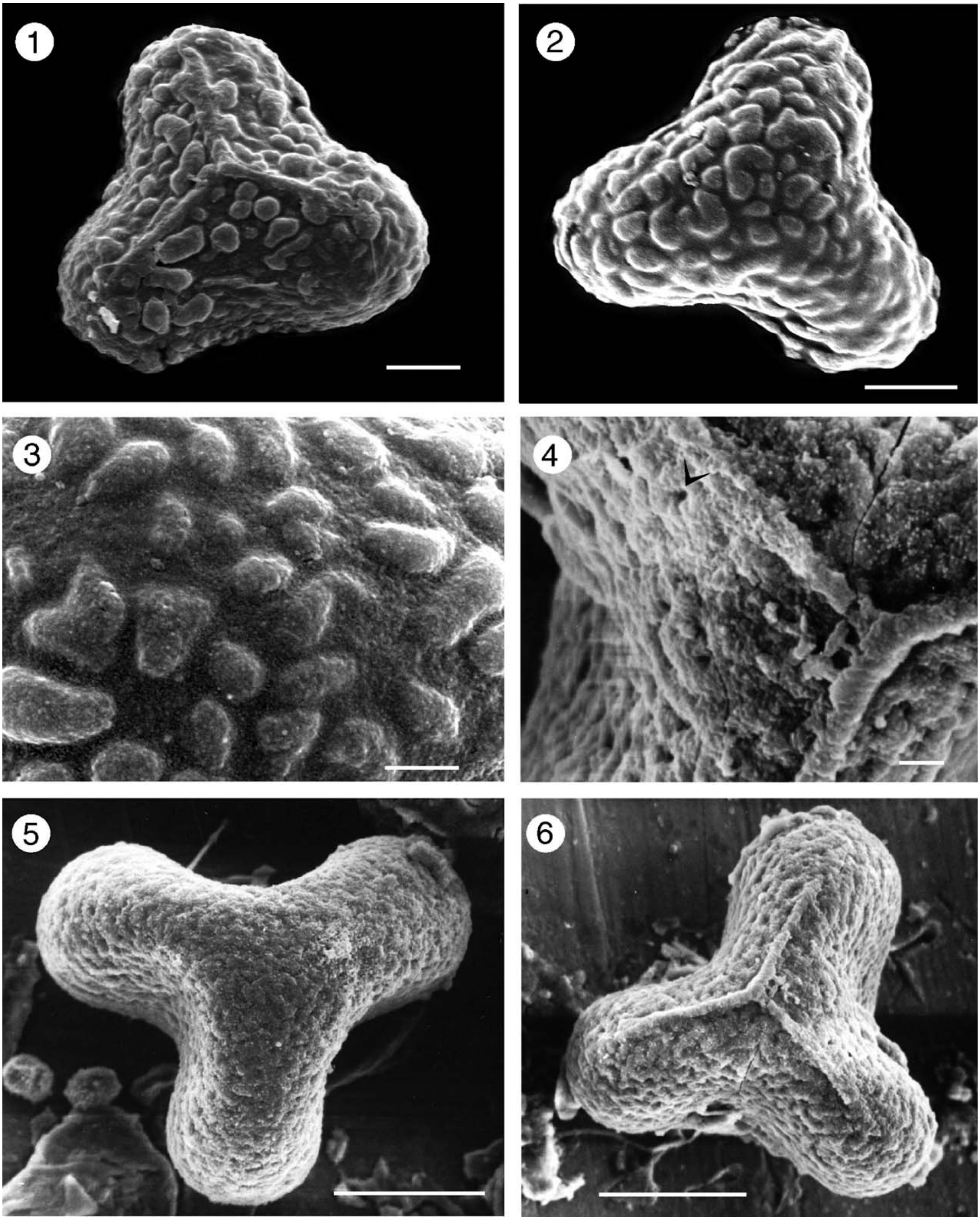
Plate I. Spores of *Dennstaedtia cicutaria* and *D. glauca* as seen with SEM.

1–3. *Dennstaedtia cicutaria*.
1. Proximal view of a spore that shows a verrucate-ridged surface. Bar: 10 µm.
2. Distal view of a spore. The ornamentation is similar to that of the proximal face. Some verrucae are fused forming ridges. Bar: 10 µm.
3. Detail of the surface that shows elements of the sculpture single or fused. Bar: 1 µm.

4–6. *Dennstaedtia glauca*.
4. Detail of the surface at the proximal area. The elements of the sculpture are tangentially elongated and partially fused. Perforations are evident (arrowhead). Bar: 1 µm.
5. Distal view of a spore with triquete outline. Bar: 10 µm.
6. Proximal view of a spore with concave sides. The elements of the sculpture are partially fused. Bar: 10 µm.

109 transmission electron microscope at the Instituto de Biología Celular, Facultad de Medicina, Universidad Nacional de Buenos Aires.

The letters MP, on the list of specimens investigated indicate the reference number of each palynological sample filed in the Laboratorio de



Palinología, Facultad de Ciencias Naturales y Museo de La Plata.

Studied material

Dennstaedtia cicutaria (Swartz) Moore

ARGENTINA: MISIONES, P. N. Iguazú, de Ferrari y Rolleri 6153 (LP).

COSTA RICA: Cartago, Tapantí, ca. 15 km S of Paraiso. Mickel 2303, (LP), MP 1461; Puntarenas, vicinity of biological fuel station of Finca Wilson 5 km S. of San Vito de Java, Mickel 3118 (LP), MP 1462.

COLOMBIA: Chocó, Río Miniquía, E. of Puerto Mitis (Bahía Solano), Lellinger and Sota 51 (LP), MP 1460; Idem, 1.5–2.5 km W of Istmina the road to Pie de Pep, Lellinger and Sota 434 (LP), MP 1459.

Dennstaedtia glauca (Cavanilles) C. Christensen ex Looser

ARGENTINA: TUCUMAN, Trancas, pie de la Cuesta Grande, Venturi 4204 (LP), MP 1465; Cumbres de Tafí, SE de Tafí, Gautier 4398, (LP), MP 1466; Tafí del Valle, Castillón 123 (LP), MP 1463a; Tafí, entre la Ciénaga y Anfama, Burkart 5152 (LP), MP 1463b; Cumbres Calchaquíes, El Huasancha, Rodriguez 1448 (LP), MP1464.

Dennstaedtia globulifera (Poiret) Hieronymus

ARGENTINA: JUJUY, Yala, Sotelo 10.057 (LP), MP 1472; Idem, Sotelo 10.077 (LP), MP 1471; El Cucho, Co. Labrado, de la Sota 4325 (LP), MP 1473.

Hypolepis repens (L.) C. Presl

ARGENTINA: JUJUY, Capital, Lagunas de Yala, Cabrera et al. 21.239 (LP), MP 1469; Idem, Krapovickas and Cristóbal 17.459 (LP), MP1468. SALTA: San Lorenzo, a orillas del arroyo, Jautier s/n (LP), MP1470. TUCUMAN: Clavillo de Aconquija, Job 1394 (LP), MP1467.

Pteridium psittacinum (C. Presl) Maxon

ARGENTINA: CORRIENTES, Tres Cerros, Co. de Susini entre grietas, Schinini, Tressens y Vanni 18552 (LP), MP1474. JUJUY: El Cucho, Co. Labrado, Sota 4360 (LP), MP1475.

3. Results

The descriptions of the spore characteristics are given under each specific name. Illustrations are SEM pictures (Plates I, II and III) and TEM micrographs (Plates IV, V, VI and VII).

Dennstaedtia cicutaria (Swartz) Moore
(Plate I, 1–3; Plate IV, 21–24)

Trilete spores, with triangular to triquete outline in polar view (Plate I, 1–2). Equatorial diameter 21.0 (23.6) 25.9 μm, polar diameter 16.1 (18.2) 20.3 μm. In equatorial view the proximal face is plane and the distal face is convex. The laesurae are straight, 7.0 (9.3) 14 μm long, and of moderate thickness (Plate I, 1).

With LM the exospore is light brown, 1.2–1.6 μm thick. With TEM it has an almost uniform thickness with uneven outer surface (Plate IV, 22–24). In a number of ultra-thin sections, it shows two clearly distinct layers: a very thin, compact inner layer, and an outer layer that forms the main part of the wall and shows minute cavities with dark contents in its innermost part (Plate IV, 21–22). However, this two-layered fine structure is poorly visible or quite indiscernible in many sections of the exospore, especially outside the apertural area (Plate IV, 23–24). According to the plane of section radial channels with contrasted contents are evident within the outer layer, these channels are particularly frequent in the aperture basal part (Plate IV, 21–23).

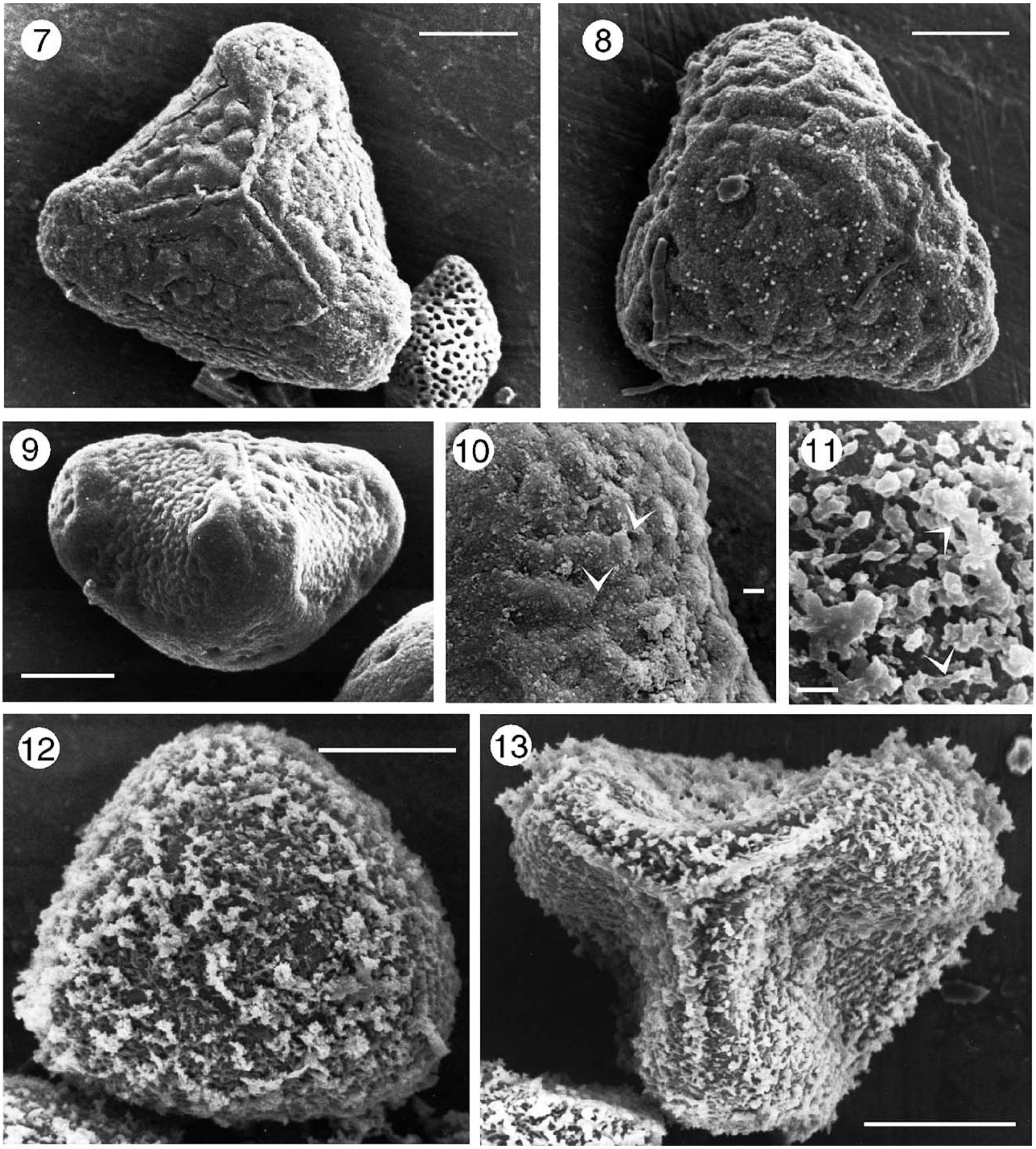
With LM the perispore is yellow, up to 1.4 μm thick. The sculpture is verrucate-ridged and similar on both polar faces. The ridges appear to result from fusion of two or several verrucae (Plate I, 1–3).

Plate II. Spores of *Dennstaedtia globulifera* and *Pteridium psittacinum* as seen with SEM.

7–10. *Dennstaedtia globulifera*.
7. Polar view of a spore with a triangular outline that shows a verrucate-ridged surface. The laesurae are crassimarginate. The elements of the sculpture decrease in size towards the equatorial area. Bar: 10 μm.
8. Distal view of a spore with a ridged surface. Some ridges are partially fused forming an incomplete reticule. Bar: 10 μm.
9. Equatorial view of a spore that shows the laevigate corners. Note that the sculpture is low at the equator. Bar: 10 μm.
10. Detail of the surface. Verrucae, ridges and perforations (arrowheads) are seen. Bar: 1 μm.
11–13. *Pteridium psittacinum*.
11. Detail of the surface that shows irregular-shaped elements of the sculpture. They have a thickened tip and are partially fused forming large strands (arrowheads). Bar: 1 μm.
12. Distal view of a spore. Bar: 10 μm.
13. Proximal view of a spore with triquete outline. The laesurae have the same sculpture (than) as the rest of the surface. Bar: 10 μm.

With TEM the perispore is more contrasted than the exospore and double-layered. The inner layer (P1) constitutes the bulk of this wall and has a microlacunose structure. It shows great variations in thickness, thus forming the verrucae and ridges of the sculpture. It is unbroken but usually thin at the laesurae apex (Plate IV, 21). The outer layer (P2) is very thin and apparently discontinuous (Plate IV, 22–24).

Comments: The spore characteristics of the material from Misiones (Argentina) are similar in surface and in section to those given by Tryon and Lugardon (1991, p. 271, 272) based on material from Panamá and Ecuador.



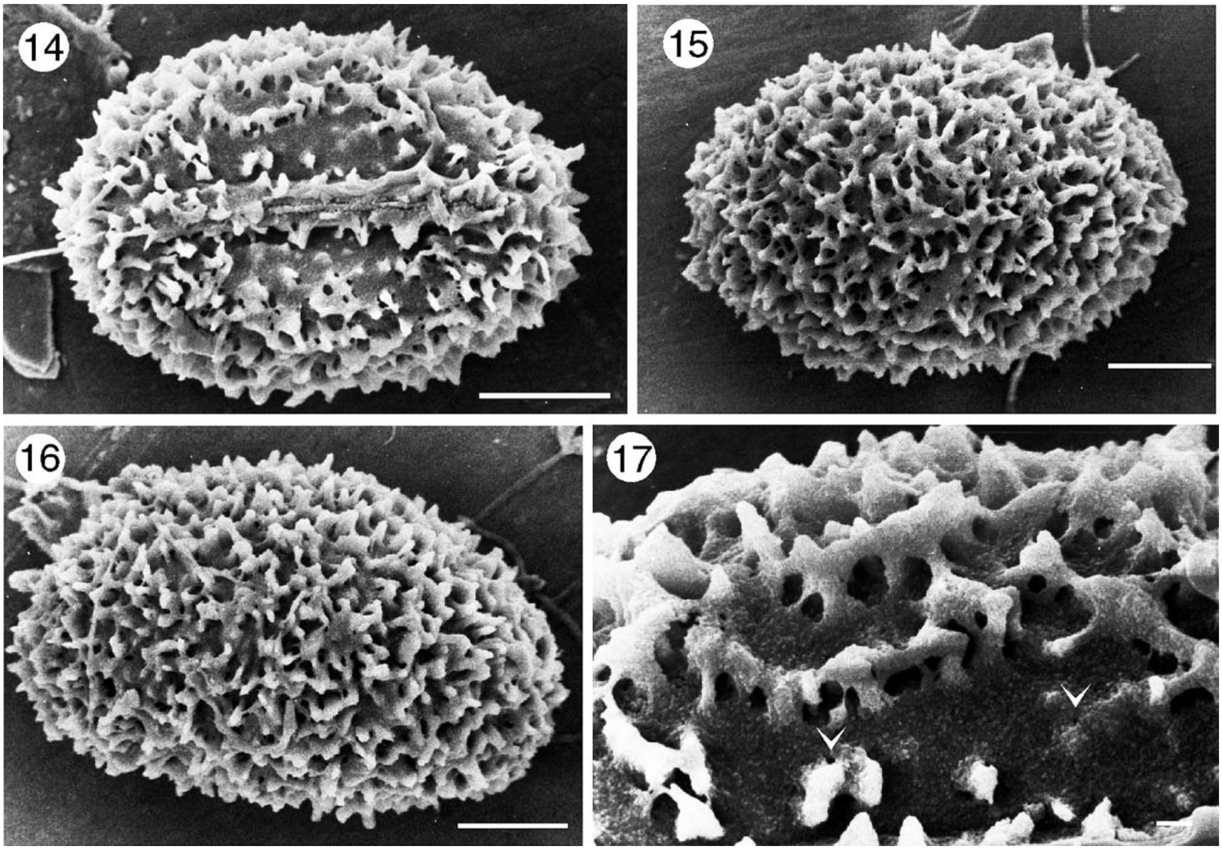


Plate III. Spores of *Hypolepis repens* as seen with SEM. 14–17:

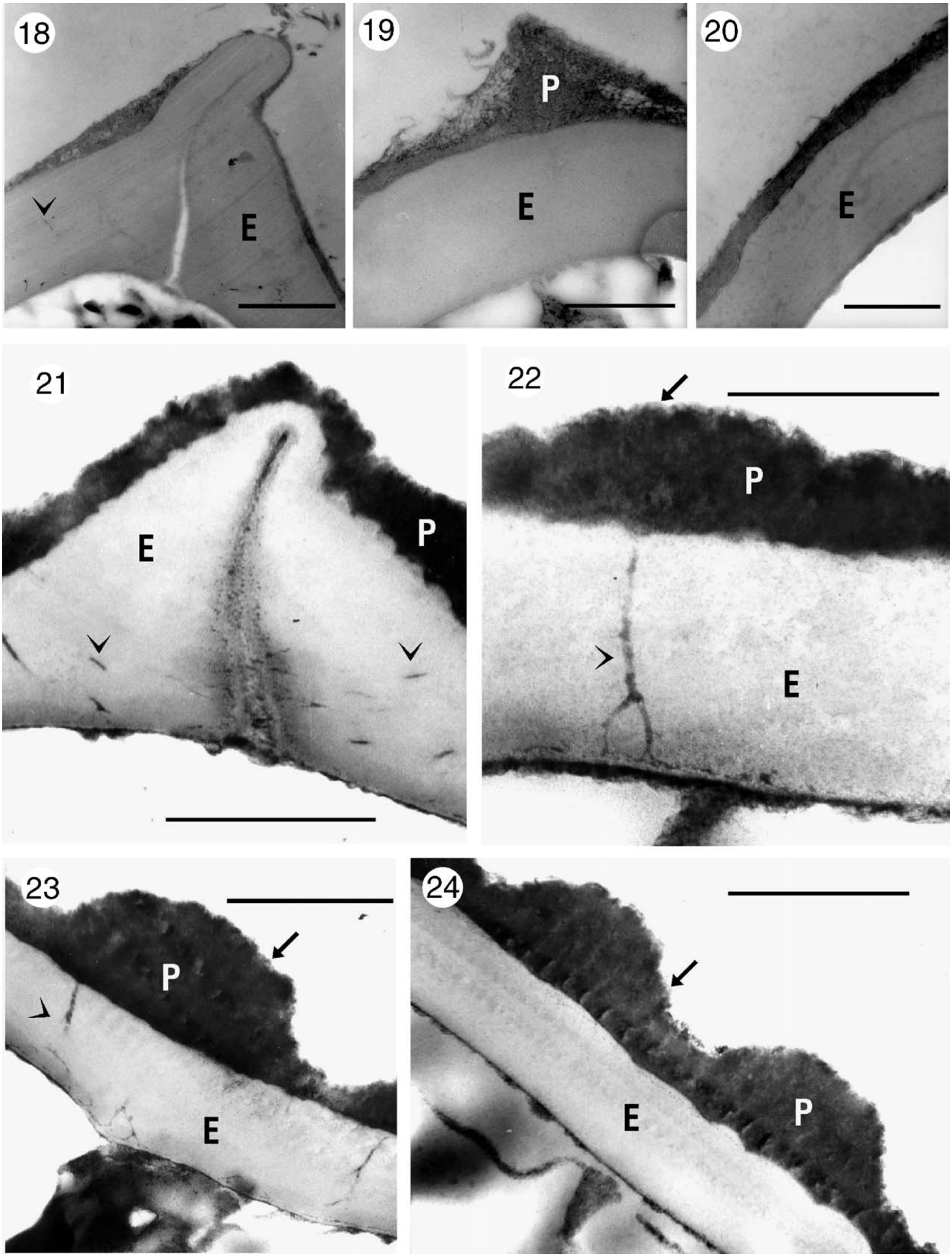
14. Monolete spore in proximal view. There are few elements of the sculpture near the laesura. Bar: 10 μm.
15. Distal view of a spore that shows elements of the sculpture in high density. Bar: 10 μm.
16. Equatorial view of a spore. Bar: 10 μm.
17. Detail of the proximal surface. The elements of the sculpture have variable shape, they fuse at the apex forming muri. The muri are also partially fused. Few perforations (arrowheads) are evident on the surface. Bar: 1 μm.

Dennstaedtia glauca (Cavanilles) C. Christensen ex Looser
(Plate I, 4–6; Plate IV, 18–20)

Trilete spores with triquete outline in polar view (Plate I, 5–6). Equatorial diameter 23.1 (25.2) 28.7 μm, polar diameter 15.4 (19.3) 27.3 μm. In equatorial view

Plate IV. Spore wall sections of *Dennstaedtia glauca* and *Dennstaedtia cicutaria* as seen with TEM. :

18–20. *Dennstaedtia glauca*.
18. Section of the sporoderm through the laesura. The exospore (E) is thick at the laesura. The perispore on the laesura is thinner than in the rest of the surface. According to the plane of sectioning some channels in the exospore are evident (arrowhead). Bar: 1 μm.
19. Section through the sporoderm that shows a perispore (P) process. It has a wide base. The perispore ultrastructure is microlacunouse, in some areas the network is more lax. The exospore (E) is homogeneous and compact. Bar: 0.5 μm.
20. Section of the sporoderm between processes of the sculpture. The perispore is apparently single-layered and uniform in thickness. Bar: 0.5 μm.
21–24. *Dennstaedtia cicutaria*.
21. Section through the sporoderm at the laesura region. It shows a thick exospore (E) with an irregular margin. According to the plane of sectioning part of channels with contrasted content (arrowhead) are evident. Bar: 1 μm.
22, 23 and 24. Some channels are branched inwards and fuse to the inner system of cavities within the inner part of exospore (arrowheads). The perispore is darkly contrasted and double-layered. The inner layer is the thickest one and forms the verrucae. The outer layer is thin, adhered to the inner one and apparently discontinuous (arrows). Bars: 1 μm.


18
E
19
P
E
20
E
21
E
P
22
P
E
23
P
E
24
E
P

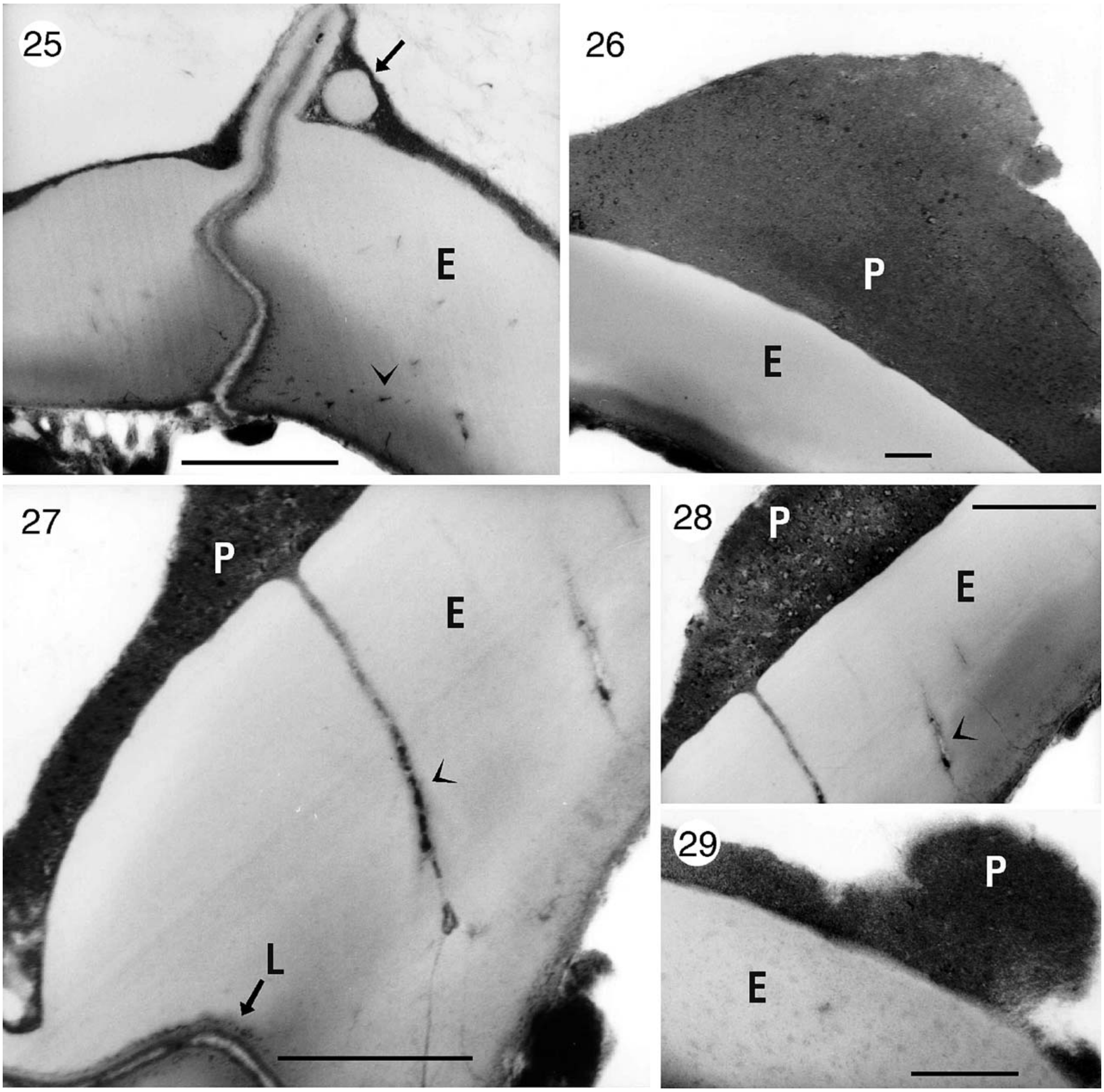


Plate V. Spore wall sections of *Dennstaedtia globulifera* as seen with TEM.

25. Section of the sporoderm through the laesura. The laesura margin is high. The exospore (E) is apparently double-layered. Cavities (arrowhead) fields with a contrasted content are evident at the inner part of the exospore. The perispore (P) is darkly contrasted and thin at the laesura. A globule is evident within the perispore (arrow) at one side of the laesura with the same structure and contrast as the exospore. Bar: 0.5 μm.

26. Section through the sporoderm. Within the exospore two levels of different contrast are evident. The perispore (P) in this area is thick. Its structure is microlacunar. Some of the microlacuna are filled with a dark contrasted material. Bar: 0.5 μm.

27–28. Section through the sporoderm in a region next to the laesura (L). According to the plane of sectioning channels filled with a contrasted content are evident (arrowheads). The system of cavities in the inner part of the exospore is evident. The perispore (P) is irregular in thickness and has a microlacunar structure. Bars: 0.5 μm.

29. Section through a perispore process. Bars: 0.5 μm.

the proximal face is plane and the distal face is hemispheric. The laesurae are straight and 1,5 μm high in section, 8,4 (10,4) 12,6 μm long, with moderately thickened margins (Plate I, 4, 6). Generally one of the laesurae is longer than the other two.

With LM the exospore is yellow, 2 μm thick, with smooth margin. With TEM it has an invariable thickness and a slightly irregular surface (Plate I, 19–20). It appears compact and homogeneous. Few unambiguous indications of the double-layered ultrastructure have been observed in investigated sections. According to the plane of sectioning, channels are visible. These channels seem to be more abundant at the base of the apertural area (Plate IV, 18).

With LM the perispore is brown, 0.2–1.2 μm thick. With TEM this wall is more contrasted than the exospore and single-layered in section. It forms the processes of the sculpture, some of them have a wide base and a sharp tip (Plate IV, 19). The perispore ultrastructure is microlacunose; in some areas its network is more lax and has cavities of larger size (Plate IV, 18–20).

The perispore sculpture consists of tangentially elongated processes. On the surface the processes are straight or vermicular, fused at random forming a dense, irregular, partially open reticule (Plate I, 4–6).

Comments: Apart from the trilete spores that characterise this species, intermediate spores between monolete and trilete as well as dilete (cfr. Sladkov, 1971: 40) were observed in the specimen *Venturi 4204* (LP).

Dennstaedtia globulifera (Poiret) Hieronymus
(Plate II, 7–10; Plate V, 25–29)

Trilete spores with a triangular to triquete outline (cfr. Kawasaki, 1973: 156) in polar view (Plate II, 7, 8). Equatorial diameter 28.0 (31.2) 35.0 μm, polar diameter 18.9 (21.0) 23.8 μm. In equatorial view the proximal face is plane and the distal face is hemispheric (Plate II, 9). The laesurae are straight with moderate thickened margins, 1.2 μm high in section and 11.2 (14.2) 18.9 μm long.

The exospore as seen with LM is yellow, smooth, 1.1–1.4 μm thick. As seen with TEM it is compact, apparently double-layered (the layers are more evident in the apertural area) (Plate V, 25). The laesura margins have an exospore thickening (Plate V, 25). According to the plane of sectioning channels are evident following different directions. Some of them seem to be ramified several times (Plate V, 27, 28).

The perispore as seen with LM is brown, 1.4 μm thick. It is thin on the sides and thick at the corners and the distal face. As seen with TEM it is darkly contrasted, single-layered with a microlacunose structure (Plate V, 26–29). Elements of variable size and rather spheric with a similar structure to that of the exospore outer layer are present (Plate V, 25).

The perispore sculpture is different on both faces. It is verrucate-ridged with elements which are partially fused proximally and ridged, and with elements partially fused distally (Plate II, 7–10). The proximal verrucae are irregular in shape and size. They are large and fused next to the laesurae, while the ones located at the centre and the equatorial zone of each facet are small. Few perforations are present (Plate II, 10).

Comments:

It is difficult to generalise the ornamentation characteristics, since it varies from one spore to another in the same sample. In general the corners and the distal face are laevigate. The perispore is continuous with a uniform thickness. Apart from the typical trilete, monolete, intermediate and tetralete spores were also observed in samples *Sotelo 10.057* (LP) and *de la Sota 4325* (LP). After the acetolysis treatment many spores lost their persipore.

Lorscheitter et al. (2002) described the spores of this species as verrucate based on material from Southern Brazil.

Hypolepis repens (L.) Presl
(Plate III, 14–17; Plate VI, 30–35)

Monolete spores, elliptic in polar view, plane-convex in equatorial view (Plate III, 14–16). Equatorial diameter 25.2 (27.9) 38.5 μm, polar diameter 25.2 (28.1) 35.7 μm. The laesura is crassimarginate, 19.6 (28.4) 35 μm long, sinuous, inclined with respect to the major symmetry axis of the spore. It is often bifurcate at 3/4 of its extension or at the end.

With LM the exospore is brown, 2.7 μm thick. With TEM it is double-layered with an echinulate-papillate surface (Plate VI, 32–34). According to the plane of sectioning many channels are evident. They have a contrasted content and are ramified and interconnected according to different directions (Plate VI, 34, 35).

With LM the perispore is yellow and 1.9 (2.8) 3.5 μm thick. It is thin at the apertural zone. With TEM it is darkly contrasted. It has two layers P1 and P2. The inner layer P1 is in contact with the exospore, it is the thickest one and shows a homogeneous microlacunose structure. Within P1 three strata are evident, inner (P1i) adhered to the exospore is ca. 0.9 μm thick and continuous. The middle stratum (P1m) is 1.5–3 μm thick and outer stratum (P1o) is discontinuous and shows a variable thickness (Plate VI, 30–33). The outer layer P2 is very thin and covers P1 (Plate VI, 30, 34).

The perispore sculpture is composed of processes 1.9 (2.8) 3.5 μm high of varied shape. They can be compact or hollow and straight or curved and frequently fused by their bases or tip. The processes are uniformly distributed while sometimes they are arranged according to the muri of a wide reticule. There were a few or no processes at the proximal face next to the laesura area (Plate III, 14–17). The perispore covers the laesura with the same type of processes, but they are slender, lower and spaced. Few perforations are also present (Plate III, 17).

Comments: The structure of the sporoderm as well as the junction between exsopore and perispore "as a zip" observed in some spores of *Hypolepis repens* studied here are similar to that of *H. crassa* according to Tryon and Lugardon (1991: 287, fig. 98.20 and fig. 98.21).

Pteridium psittacinum (C. Presl) Maxon (Plate II, 11–13; Plate VII, 36–38).

Trilete spores with triquete outline in polar view (Plate II, 12–13). In equatorial view the proximal face is plane to slightly concave. The distal face is hemispheric. Equatorial diameter 20.3 (25.2) 30.1 μm, polar diameter 16.8 (19.0) 24.5 μm. The laesurae are straight, 7.0 (9.9) 16.1 μm long and 1.5 μm. high in section, with slightly thickened margins.

With LM the exospore is brown-yellowish, 1.3 μm thick. With TEM it has an irregular margin and is double-layered with a compact structure (Plate VII, 36–38).

With LM the perispore is brown, 1.7 μm thick and it becomes thinner at the laesura. With TEM it is strongly contrasted and shows a microlacunose structure. A continuous layer adhered to the exospore (15–20 nm thick) is seen in section. It bears elements of varied size and shape, partially fused. A membrane of low contrast covers the elements of the outer layer (Plate VII, 36, 38).

The perispore surface is irregular with a variable thickness even in a single spore. It is composed of thin, radially elongated processes with thick tips curved fused at different heights forming strands which are arranged in varied directions (Plate II, 11–13). The perispore is continuous over the laesurae where it has the same sculpture.

Comments:

After chemical treatment the spores show a perispore of even thickness. Thus, thin areas with free low processes with thick tips are seen.

Lugardon (1974) described the perispore as having expansions with granulose texture without a defined shape. According to this author in acetolyzed spores followed by treatment with MnO_4K the different parts of the perispore seem to be surrounded by a thin continuous membrane.

Tryon and Lugardon (1991) described the perispore as composed of fused granules forming strands.

Lorscheitter et al. (2002), described the spores of this species as having a granulate perispore based on material from Southern-Brazil.

4. Discussion and conclusions

As it was possible to observe the junction between exospore and perispore "as a zip" in some spores of *Hypolepis repens* is only evident in immature material while in ripe spores this surface is more or less plain.

The exospore and perispore ultrastructure of *Hypolepis repens* here analysed were found similar to those described by Tryon and Lugardon (1991) for *Hypolepis crasa* (287, fig. 98.20), as well as in other genera of the Dennstaedtiaceae like *Paesia* (284, fig. 97.11 and 97.12) and *Lindsaea* (fig. 105.18 and 19).

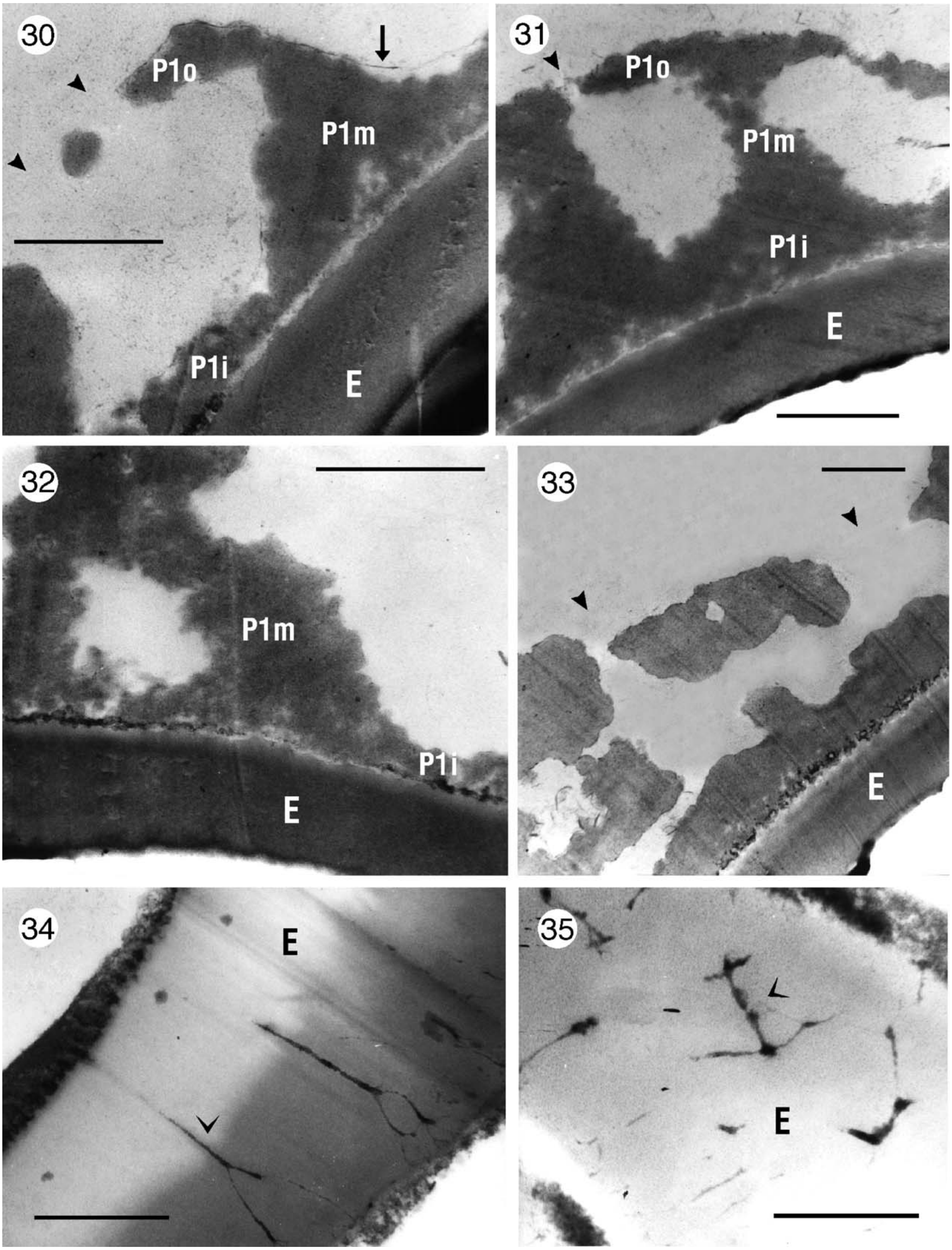
Plate VI. Spore wall sections of *Hypolepis repens* as seen with TEM.

30–33. Sections through the sporoderm that show a double-layered perispore (P). The layer P2 is evident in fig. 30 (arrow). It covers the P1 layer (P1) and is very thin. Within the P1 layer (P1) three strata are distinguishable in figs. 30–33. The inner stratum (P1i) is continuous and adhered to the exospore. The middle stratum (P1m) is camerate and has high irregular processes. The outer stratum (P1o) is discontinuous. These discontinuities are seen in figs. 30, 31 and 33 (arrowheads). Bars: 1 μm.

34–35. The exospore (E) margin is echinate-papillate. Channels (arrowheads), are evident in the exospore, some of them are ramified. 34, Bar: 0.5 μm. 35, Bar: 250 nm.

Plate VII. Spore wall section of *Pteridium psittacinum* as seen with TEM (see page 256).

36–38. The exospore (E) margin is irregular and has an homogeneous structure. The perispore (P) is darkly contrasted and formed consists by two levels with different structure. The inner level consists of a continuous thin layer that lies on the exospore. The outer level consists of irregular elements (asterisks). A thin membrane covers these elements. It can be better seen in figs. 36 and 38 (arrowheads). Bars: 0.5 μm.


30
P1o
P1m
P1i
E
31
P1o
P1m
P1i
E
32
P1m
P1i
E
33
E
34
E
35
E

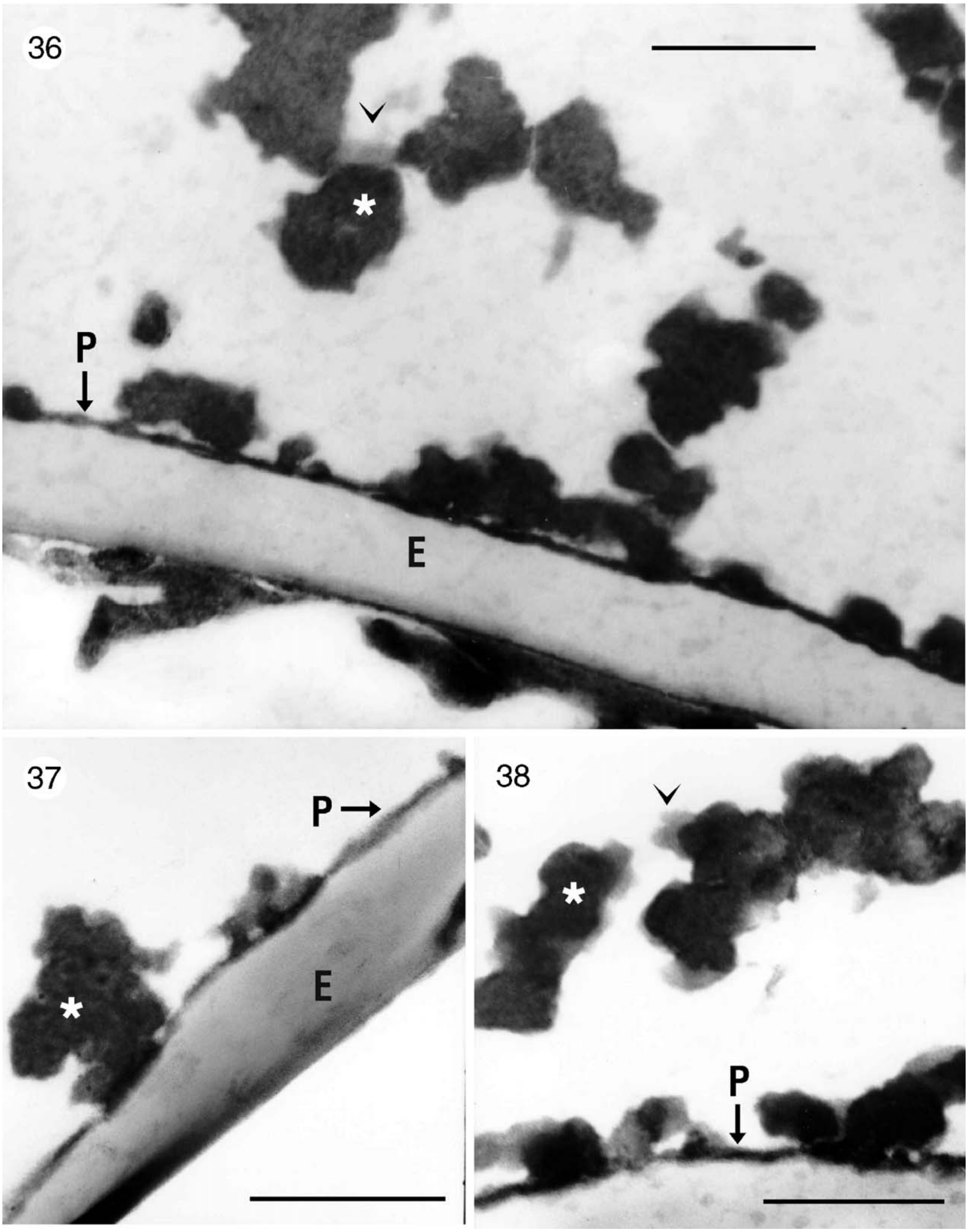


Plate VII (*caption on page 254*).

Gathering all the morphological and structural data obtained from the study of Dennstaedtiaceae that grow in North-west Argentina, we can conclude that:

The exospore is generally two-layered. These layers are more evident in young spores and in the area next to the laesurae. The outer exospore is smooth in *Dennstaedtia*, it is slightly ornamented (or irregular) in *Pteridium* and dentate-papillate in immature spores of *Hypolepis*. According to the plane of sectioning channels filled with a contrasted content are observed within the exospore in its whole extent. They are located in different directions and some of them are ramified. Cavities are also evident in the inner part of the outer exospore, and separated from the spore cavity by a thin but quite compact inner layer.

The perispore is single to double-layered. These layers are distinguishable by their thickness and contrast. The thickest wall always shows a microlacunar structure Thus the perispore of *Dennstaedtia glauca* and *D. globulifera* are single-layered while it is double-layered in *D. cicutaria.* The perispore in *Pteridium* is single-layered while in *Hypolepis* it is single-layered with three strata.

Spheroids were seen only in spores of *Dennstaedtia globulífera*. Those spheroids were found as common on spores of Filicales. Lugardon (1981) quoted "captive" globules in *D. bipinnata* (p. 97, Plate I, fig. 2). This author compared these elements with pollen grains in angiosperm anthers, which are known as Ubisch bodies. He found these globules as homologous to spermatophytes Ubisch bodies.

The sculpture in all the cases analysed is determined by the perispore. The ornamentation varies from echinate to verrucate to partially reticulate. It can be similarly uniform on the whole surface like in *Pteridium*, *Dennstaedtia glauca* and *D. cicutaria*, or it can have variations according to the area of the spore taken into consideration. For instance, in spores of *Dennstaedtia globulífera* the corners and part of the distal face are smooth while in *Hypolepis repens* the area next to the laesura is slightly ornamented with few processes.

According to palynologic data found in the present study the characteristics could be significant for systematic purposes. These characteristics might be the laesura type, the exospore margin and the perispore ornamentation, stratification and complexity. As pointed out previously, similarities in morphology and structure were found with other genera within Dennstaedtiaceae that grow in other regions of America. Thus, in our opinion this analysis would also provide data that could be useful for future studies on phylogeny within this group.

Acknowledgements

The authors thank Rafael Urrejola of the Servicio de Microscopía Electrónica de Barrido del Museo de Ciencias Naturales de La Plata, to Ing. Luis Zimmermann of the Servicio de Miscroscopía Electrónica de Transmisión del Instituto de Biología Celular, Facultad de Medicina de la Universidad de Buenos Aires and to Isabel Farías for her kindness and help in technical aspects of sample processing. This work was supported by grants from the National Council of Scientific and Technological Research (CONICET) Buenos Aires for PIP 5044, Universidad Nacional de La Plata for project 363 and PICT 12758 from ANPCyT.

References

Erdtman, G., 1960. Pollen and spore morphology and plant taxonomy. Gimnospermae, Pteridophyta, Briophyta. Pteridophyta. An Introduction to Palynology II. Almqvist & Wiksell, Stockholm.

Kawasaki, T., 1973. A morphographic key for pteridophytic spores geared for computerization. Bull Tokyo Gakugei Univ. Ser. IV 25, 154–173.

Lorscheitter, M.L., Ashraf, A., Windisch, P., Mosbrugger, V., 2002. Pteridophyte spores of Río Grande do Sul, Brazil: Part IV. Paleontogr. Abt B 263, 1–159.

Lugardon, B., 1974. La structure fine de l'exospore et de la perispore des Filicinées isosporées: II. Filicales. Commentaires. Pollen Spores 16 (2), 161–226.

Lugardon, B., 1981. Les globules des Filicinées, homologues des corps d'Ubisch des Spermatophytes. Pollen Spores 23 (1), 93–124.

Mickel, J.T., 1973. The classification and phylogenetic position of the Dennstaedtiaceae. In: Jermy, A., Crabbe, J., Thomas, B. (Eds.), The Phylogeny and Classification of the Ferns. Bot. J. Linnean Society, vol. 67 (1), pp. 135–144.

Morbelli, M.A., 1978. Spore morphologic key and photomicrographs of the Fuego-Patagonian Pteridophyta. In: Markgraf, D'Antoni (Eds.), En: Pollen flora of Argentina. Univ. Arizona Press, Tucson. 208 pp.

Morbelli, M.A., 1980. Morfolog¡a de las esporas de Pteridophyta presentes en la región fuego-patagónica, República Argentina. Opera Lilloana 28. 95 pp.

Morbelli, M.A. 1985. Estudio de las esporas de las Pteridofitas del Noroeste de Argentina. Dennstaedtiaceae. XX Jornadas Argentinas de Botánica, Salta. Resúmenes, p. 76.

Ponce, M.M., 1996. Pteridophyta. In: En Zuloaga, F.O., Morrone, O. (Eds.), Catálogo de plantas vasculares de la República Argentina: I. Monography in Systematic Botany from the Missouri Botanical Garden, vol. 60.

Rowley, J.R., Nilsson, S., 1972. Structural stabilization for electron microscopy of pollen from herbarium specimens. Grana 12, 23–30.

Sladkov, A., 1971. On pseudo-rhombic and rhombic tetrads of Pteridophyta. J. Palynol. 7, 39–42.

de la Sota, E.R., 1977. Pteridophyta. In: Cabrera, A.L. (Ed.), Flora de la Provincia de Jujuy. Col. Cient. INTA, vol. 13 (2), pp. 1–275.

Tryon, R., Lugardon, B., 1991. Spores of Pteridophyta. Springer-Verlag, New York.