11. LM INVESTIGATIONS ON PARTIALLY DISSOLVED SPOROMORPHS IV.

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Abstract

Five species from the genus Encephalartos (E. ferox BERTOL., E. horridus LEHM., E. lehmannii ECKLON, E. longifolius (JACQ.) LEHM., E. transvenosus STAPF. et DAVY) were investigated with the partial dissolution method. We established that the molecular system of the sporopollenin of the ectexine of these pollen grains is resistant against the used organic solvents. In contrast to the previous results it is unusual, that the merkaptoethanol altered in an important measure the basic morphology of these originally monosulcate pollen grains. Some of the secondary altered pollen grains are similar to ancient Normapolles taxa (Interpollis), other ones to evolved Postnormapolles (Alnus type) of the angiosperms.

Key words: Palynology, recent Encephalartos, partial dissolution, LM method.

Introduction

During our previous investigations morphologically and taxonomically different kinds of pollen grains were the subjects of partial dissolution. As a terminal part of the research program of the OTKA 1/7 T 14692 five species of one genus were chosen from the genus *Encephalartos*. In this case the morphological characteristic feature was the same, *monosulcate* type what is important in the evolution of the *angiosperm* pollen grains (cf. DOYLE, 1977). The distribution of the recent species of the genus *Encephalartos* is Central and South Africa (SCHUSTER, 1931, GREGUSS, 1968).

ERDTMAN (1954) published the LM morphology of *E. altensteinii* and he wrote as follows; p. 130: "distal part of grain with open furrow". In 1965 ERDTMAN summarized the previous papers concerning the pollen morphology of the genus *Encephalartos*. Besides this work, the earliest paper was published by SMITH (1907).

The basic LM morphology with the intraspecific morphological variations was published by KEDVES and AILER (1990) and a peculiar polar differentiation was described. The *trichotomosulcate* form was occasionally observed at the pollen grains of *E. humilis* VERDOORN. The high temperature effect on the pollen grains of *E. transvenosus* STAPF. et DAVY was investigated in this paper. The first partial dissolution experiments were published by KEDVES and GÁSPÁR (1994) on the pollen grains of *E. ferox* BERTOL. The alterations in consequence of the merkaptoethanol were also observed at these first experiments. Recently the Duhoux effect was investigated on the pollen grains of six species of the genus *Encephalartos*, and of *Ceratozamia mexicana* (KEDVES et al., 1999).

Materials and Methods

Pollen samples of several species of the genus *Encephalartos* were disposed for our investigations by Dr. P. VORSTER (Department of Botany, The University of Stellenbosch, Rep. of South Africa). The method of investigations was our standard method, which was first published in 1997 (KEDVES, KÁROSSY and BORBOLA).

Results

As a general establishment, the resistance of the molecular system of the sporopollenin of the ectexine may be pointed out. The resistance against the diethylamine solvent is worth of mentioning.

But the LM morphological alteration in consequence of the merkaptoethanol may be pointed out. The polar circular differentiation of the exine was observed at all of the experiments. Regarding the details the following will be pointed out.

Encephalartos ferox BERTOL. (Plate 11.1., figs. 1-42)

Diethylamine and methanol have not altered the basic morphology of the pollen grains only after 330 days of dissolution the outer layers of the ectexine detached at the poles. The morphological characteristic features altered in an important measure with the dissolution with merkaptoethanol. The polar separation of the outer layers of the ectexine is characteristic after 30 and 90 days of dissolution. From 150 days the basic monosulcate morphology is not discernible. No important alterations were observed after partial dissolution with ethanol and n-propanol. Minor more or less similar alterations were observed after the dissolution with n-butanol and i-amyl alcohol. There are polar separations of the outer layer of the ectexine and the secondarily rounded ambitus.

Encephalartos horridus LEHM. (Plate 11.2., figs. 1-42)

The characteristic annular differentiation on the proximal surface is more characteristic after dissolution with diethylamine. Merkaptoethanol resulted in very important morphological alterations after 30 days of dissolution. Extremely early extinct *angiosperm* pollen like forms (*Interpollis*) are illustrated in picture 37 (Plate 11.2.). The alterations which appeared in consequence of further alcohol are not characteristics.

Encephalartos lehmannii ECKLON (Plate 11.3., figs. 1-42)

At this species also the dissolution with merkaptoethanol resulted in important alterations. The secondary forms are different from the previous one. The ambitus of the altered forms is quadrangular or pentagonal. In this way the original morphological characteristic features may not be discernible at these pollen grains. The other used solvents resulted in only minor alterations, which were discussed previously.

Encephalartos longifolius (JACQ.) LEHM. (Plate 11.4., figs. 1-42)

The morphological alterations of the partially dissolved pollen grains of this species are nearly identical to those of the previous species. Differences: After 30 days of dissolution the monosulcate forms are characteristic. "Alnus-like" morphology appeared after 150 days of dissolution (Plate 11,4., fig. 16).

I M E/D AYS

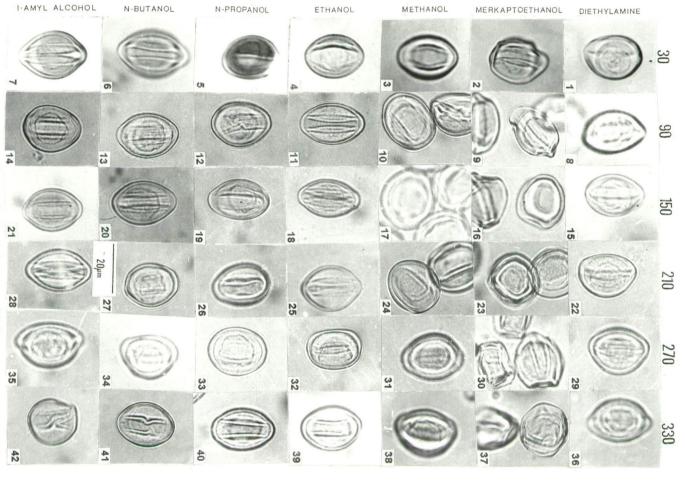


Plate 11.1., 1-42. Encephalartos ferox BERTOL

IME/DAYS

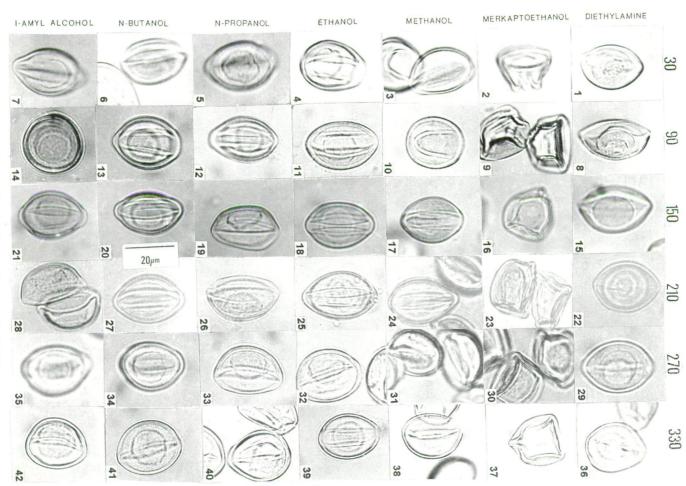


Plate 11.2., 1-42. Encephalarios horridus LEHM.

TIME/DAYS

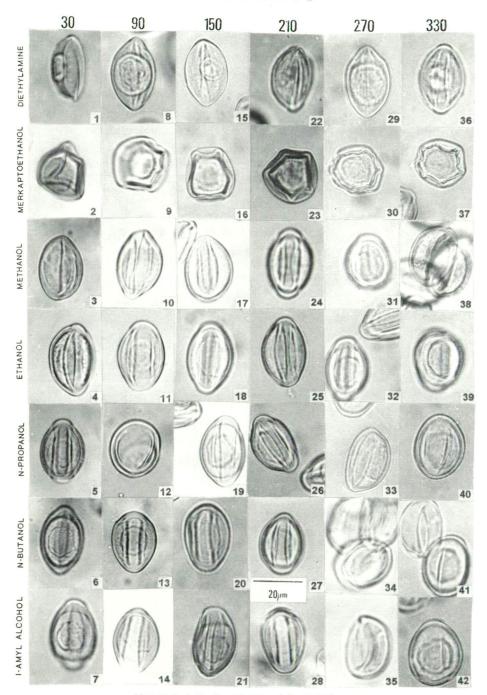


Plate 11.3., 1-42. Encephalartos lehmannii ECKLON.

Encephalartos transvenosus STAPF. et DAVY (Plate 11.5., figs. 1-42)

The dissolution's effect of the merkaptoethanol is similar to the previous species. Worth of mentioning alterations in contrast to the previous are as follows: Characteristic alterations after 330 days of dissolution with methanol. At this species the i-amyl alcohol resulted also in taxonomically important alterations after 210-330 days of dissolution.

Discussion and Conclusions

- 1. The molecular system of the sporopollenin of the investigated *Encephalartos* pollen grains is resistant to the organic solvents, which were used during our experiments. In the first place the resistance against diethylamine may be pointed out.
- 2. The secondary forms which appeared after the partial dissolution with merkaptoethanol are unusual and important. The secondary forms may be similar to extinct and recent brevaxonate *angiosperm* pollen grains. Differences in the altered forms of the different species investigated were established.
- 3. It is also interesting that further alcohol resulted in not so important alterations in contrast to the previously investigated angiosperm pollen grains, for example alterations of the *Betula verrucosa* EHR., *Carya illinoensis* (WANG) KOCH (KEDVES, KÁROSSY and BORBOLA, 1997).

Finally further transmission electronmicroscopic investigations of the partially dissolved pollen grains seem to be necessary.

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TIME/DAYS

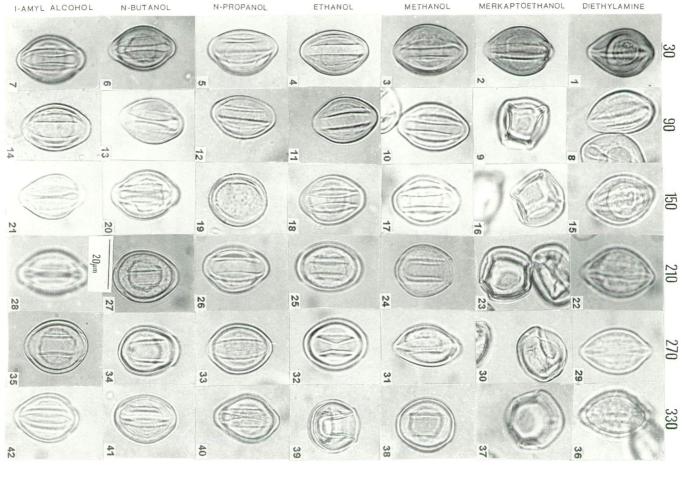
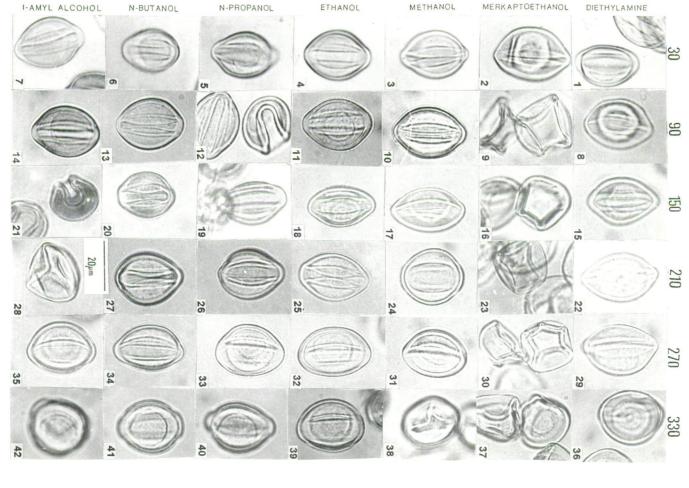


Plate 11.4., 1-42. Encephalartos longifolius (JACQ.) LEHM.

MERKAPTOETHANOL

DIETHYLAMINE



ETHANOL

METHANOL

Plate 11.5., 1-42. Encephalartos transvenosus STAPE et DAVY.

I-AMYL ALCOHOL

References

- DOYLE, J. (1977): Patterns of evolution in early angiosperms. Patterns of Evolution, 501-546.
- ERDTMAN, G. (1954): An Introduction to Pollen Analysis. Waltham Mass., U.S.A., Almqvist and Wiksell, Stockholm.
- ERDTMAN, G. (1965): Pollen and Spore Morphology/Plant Taxonomy *Gymnospermae*, *Bryophyta* (Text) (An Introduction to Palynology, III). Almqvist and Wiksell, Stockholm.
- GREGUSS, P. (1968): Xylotomy of the living Cycads with a description of their leaves and epidermis. Akadémiai Kiadó, Budapest.
- KEDVES, M. and AILER, P. (1990): Investigations on recent monosulcate *Gymnospermatophyta* pollen grains. Acta Biol. Szeged. 36, 103-105.
- KEDVES, M., BORSODI, A., DOBÓ, K., KOVÁCS, E. et SZÉCSÉNYI A. (1999): L'effet de l'hydratation sur les grains de pollen des *Cycadales*. Actes du IV^{éme} Symposium International de Palynologie Africaine 15-30 Avril 1999 Sousse/Tunisie, 41.
- KEDVES, M. and GÁSPÁR, I. (1994): Les alterations sécondaires des spores et des grains de pollen dissous partiellement. Trabajos de palinología básica y aplicada. Eds.: Isabel MATEU ANDRÉS, Michèle DUPRÈ OLLIVIER, Jaime GÜEMES HERAS y Maria Elvira BURGAZ MORENO. Universitat de Valencia, 153-161.
- KEDVES, M., KÁROSSY, Á. and BORBOLA, A. (1997): LM investigations of partially dissolved sporomorphs. Plant Cell Biology and Development (Szeged) 8, 44-55.
- SCHUSTER, J. (1931): Über das Verhältnis der systematischen Gliederung, der geographischen Verbreitung und der paläontologischen Entwicklung der Cycadaceen. Bot. Jahrb. 64, 165-260.