

4. TRANSMISSION ELECTRON MICROSCOPY OF PARTIALLY DEGRADED TELIOSPORES OF *USTILAGO MAYDIS* (DE CANDOLLE) CORDA

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Abstract

During our experimental studies on the ultrastructure of the teliospores of *Ustilago maydis* 2-aminoethanol, KMnO_4 1% and acetic anhydride were used as organic solvents and oxidizing agents. Important alterations were observed at the fine structure of the wall, and in the inner part of the teliospore. Similarities were established between the ultrastructural data of experiment No.: 191 (2-aminoethanol, KMnO_4 and acetic anhydride) and the X-ray irradiated teliospores over 15 minutes.

Key words: Palynology, recent, *Ustilago maydis*, partial degradation, TEM.

Introduction

In the course of our LM and TEM research programs on the partial dissolved, degraded and irradiated sporomorphs, we have established important differences in the wall biopolymer resistance of the different taxa of the spores and pollen grains, cf. KEDVES and GÁSPÁR (1994), KEDVES and UNGVÁRI (1996), KEDVES, KÁROSSY and BORBOLA (1997), KEDVES et al. (1998). Biopolymer and molecular structures were discovered in the walls of the sporomorphs after partial degradation with different experimental influences. The protective function of the melanins in the spore wall was pointed out and/or discussed in several papers, e.g.. PIROZYNSKI (1977). At the teliospores of *Ustilago maydis* the X-ray irradiation demonstrated the protective function of the melanins in the wall, moreover molecular structures sensu strictu were also observed by KEDVES, PÁRDUTZ and BORBOLA (1998).

The aim of this paper is to present the TEM results of the partially degraded teliospores of *Ustilago maydis* in comparison with the results of the irradiated spores.

Materials and Methods

The material of investigation was collected by Dr. Á. PALÁGYI on 22.08.1981. The spores were frozen at -20°C after collection. The temperature of partial degradation experiments was 30°C , the different methods were as follows:

Experiment No.: 188. - 20 mg teliospores + 1 ml 2-aminoethanol, length of time: 24 hours.

Experiment No.: 189. - 20 mg teliospores + 1 ml 2-aminoethanol, length of time: 24 hours, after washing + 10 ml KMnO_4 1%, during 24 hours.

Experiment No.: 190. - 20 mg teliospores + 1 ml 2-aminoethanol, length of time: 24 hours, after washing + 10 ml KMnO_4 1% during 48 hours.

Experiment No.: 191. - 20 mg teliospores + 1 ml 2-aminoethanol, length of time: 24 hours, after washing + 10 ml KMnO_4 1% during 24 hours, washing again + 2 ml acetic anhydride, length of time: 24 hours.

After experiment the spores were postfixed with OsO_4 aq. dil. 1% and embedded in Araldite. The ultrathin sections were made with glass knives on a Porter Blum ultramicrotome in the Electron Microscopical Laboratory of the Institute of Biophysics of the Biological Center of Hungarian Academy of Sciences. TEM pictures were taken in a Tesla BS-540, resolution 6-7 Å. All pictures are unretouched.

Results

Experiment No.: 188. (Plate 4.1., figs. 1,2). The exospore of the wall was partially degraded. Electron dense globular units were observed within the substance of the wall. Important degradations were observed in the fine structure of the exospore (Plate 4.1., fig. 2). The endospore swelled.

Experiment No.: 189. (Plate 4.1., figs. 3,4). The fine structure of the exospore was highly degraded, sometimes completely disappeared. The epispore remain in a good condition, the endospore swelled. The protoplasm was extremely contracted (Plate 4.1., fig. 3).

Experiment No.: 190. (Plate 4.1., figs. 5,6). Under the hardly degraded exospore a thin epispore layer was observed. The endospore is degraded. Electron dense degraded protoplasm is in the middle of the teliospore (Plate 4.1., fig. 5).

Experiment No.: 191. (Plate 4.2., figs. 1-3). The results of this experiment are extremely interesting. The exospore is superficially degraded. This degradation process revealed globular biopolymer units on the outer and inner surface of this layer by; the degradation of the further layers are not uniform. Under the exospore the finely granular epispore was more or less well preserved (Plate 4.2., fig. 1). Sometimes this part of the wall seemed to be two layered. Electron dense endospore may be present. Sometimes the inner part of the endospore is characteristically electron dense (Plate 4.2., fig. 2). As such after the experiment, this part of the wall appears to be three layered.

Discussion and Conclusions

1. Among the LM results it is necessary emphasize that, based on our previous investigations (KEDVES and GÁSPÁR, 1994), the teliospores of *Ustilago maydis* were extremely resistant to dissolution with diethylamine and merkapto-ethanol. The thick exospore of *Equisetum arvense* L. and the sporopollenin of the pollen grains of *Quercus robur* L. dissolute with the above mentioned agents.

Plate 4.1.

- 1-6. *Ustilago maydis* (DE CANDOLLE) CORDA. TEM pictures of partially degraded teliospores.
1,2. Experiment No.: 188, 1. Negative No.: 6955, 10.000x, 2. Negative No.: 6340, 50.000x.
3,4. Experiment No.: 189, 3. Negative No.: 6960, 10.000x, 4. Negative No.: 6961, 50.000x.
5,6. Experiment No.: 190, 5. Negative No.: 6376, 10.000x, 6. Negative No.: 6379, 50.000x.
Exs = exospore, Eps = epispore, Ens = endospore, Cp = cytoplasm.

Plate 4.2.

- 1-3. *Ustilago maydis* (DE CANDOLLE) CORDA. TEM pictures of partially degraded teliospores. Experiment No.: 191, 1. Negative No.: 6618, 25.000x, 2. Negative No.: 6616, 50.000x, 3. Negative No.: 6617, 50.000x.
Exs = exospore, Eps = epispore, Ens = endospore.

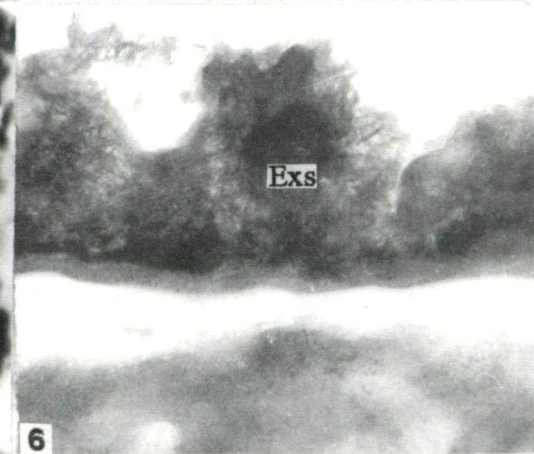
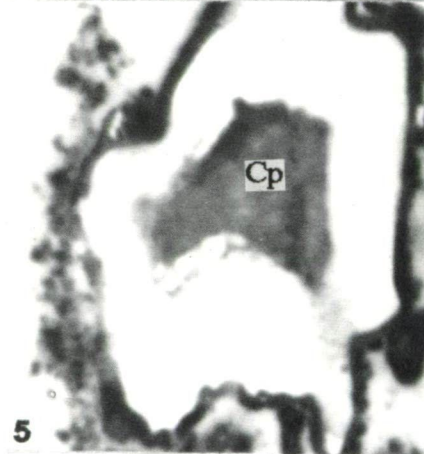
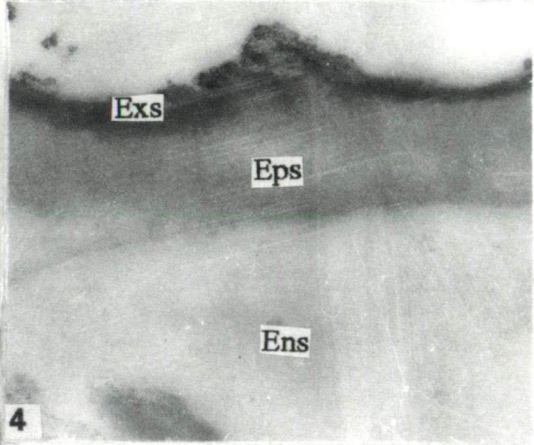
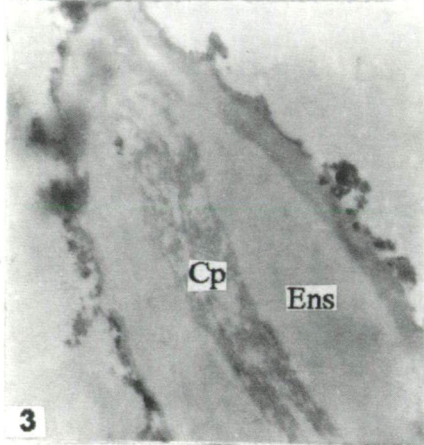
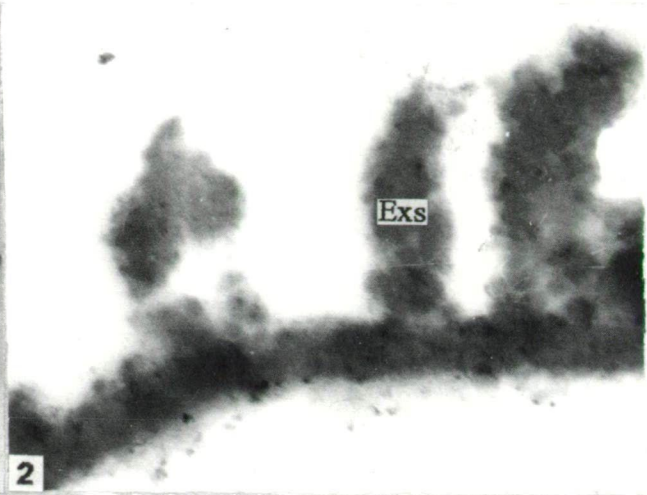
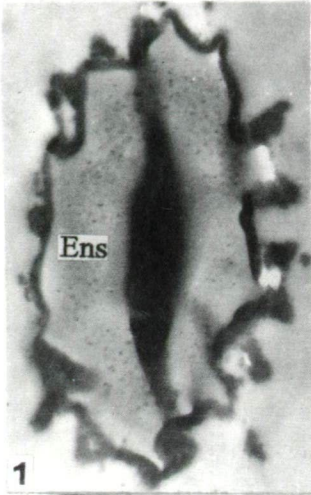


Plate 4.1.

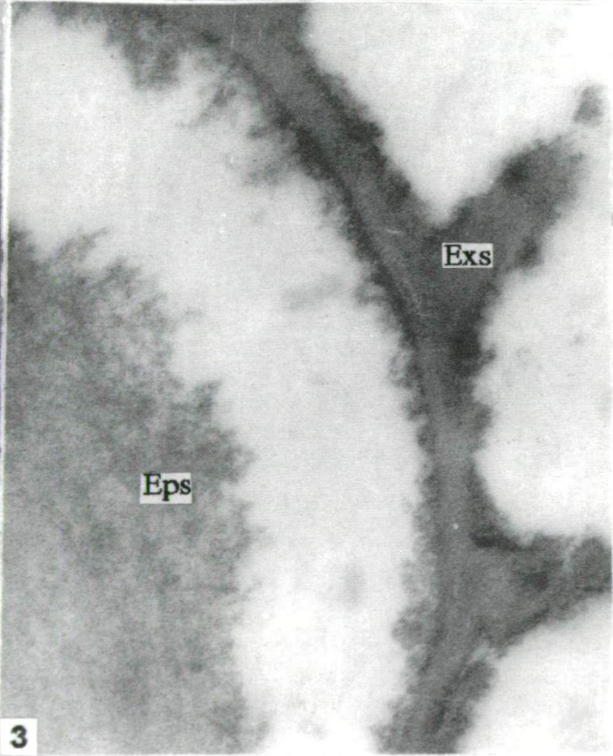
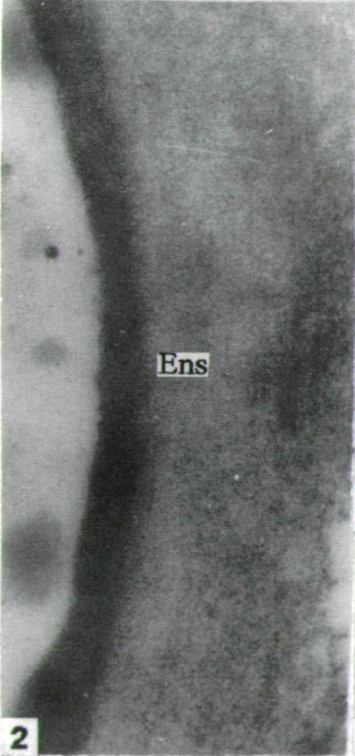
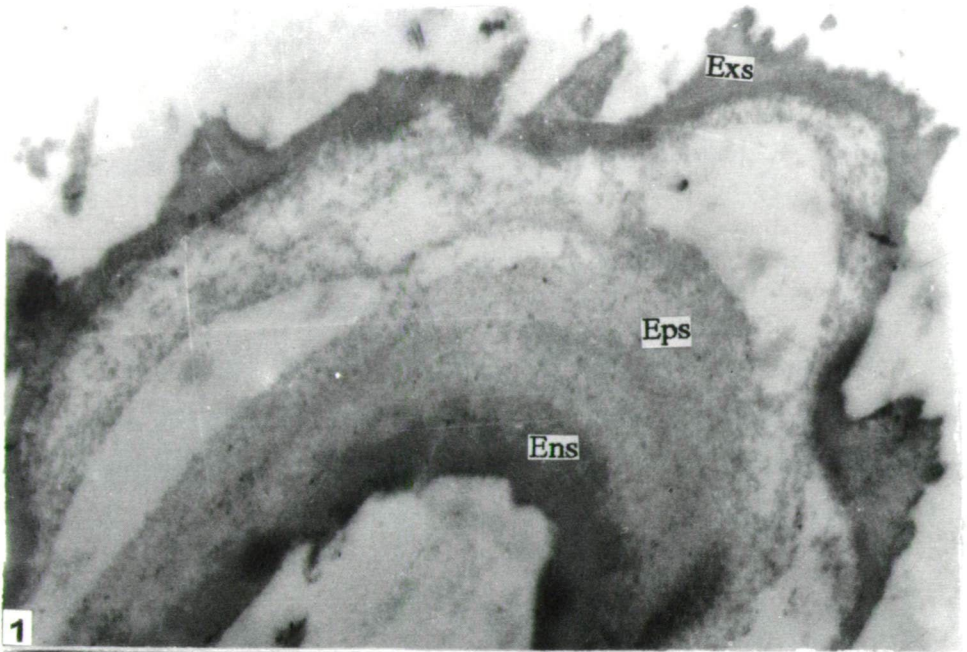


Plate 4.2.

2. By TEM method, the partial degradation experiments revealed different kinds of biopolymer structures in the wall layers of the teliospores of *Ustilago maydis*. Globular biopolymer units were observed on the surfaces. Presumably the TEM method is also a factor of the partial degradation.

3. Sometimes the TEM data within one experiment were not completely identical. This may be consequence of the state of maturity of the spores not being completely on the same level or was not in a fertile state.

4. It is interesting that the TEM results of the experiment No.: 191 are similar to or more or less identical with the X-ray irradiated teliospores over 15 minutes.

5. It may be pointed out, that the application of the acetic anhydride as degradation agents may be useful again for the further partial degradation experiments.

Acknowledgements

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