Pollen Morphology of Plantaginaceae in Jordan

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Abstract: A total number of 11 Plantaginaceae species were investigated palynologically by LM and SEM. 5 different pollen types have been recognized and the keys for these observations were prepared. All specimens investigated have polypantoporate type with many pores in ecto- and endo-aperture, but with more or less or even no annuli and costae. Presence of sXine 1 (columella), sXine 2 (tectum) and sXine 3 (scabrate) as parts of pollen exine were observed. Thin and short columella, scabrate tectum were observed also. 18-40, 1-2, 3-6 μm were be pollen diameter, exine thickness and pore diameter, respectively. Verrucae of the specimens examined were be the channel with few or without punctae.

Key words: Plantaginaceae, pantoporate, exine, annulus, operculum

INTRODUCTION

Plantaginaceae in Jordan has one genus, Plantago, which has 19 species distributed through the different phytogeographical regions.[1-3]

The basic structure of pollen grains is well-known[4-3]. Many publications gave descriptions and keys for those studied as guidelines for the others[6-30]. The palynological terms and expressions used by different researchers and investigators are sometimes making confusion and misleading[4-5,31]. All previous studies treated annulus and operculum as two parts from ectoaperture[7-11,12,14,15,24,29]. As two structures were developed to a greater or lesser extent, they were either present or absent with no intermediate state[6-3].

The development of annulus and operculum in pollen grains have been discussed by many investigators[6,9,14,15,19,25].

Annulus formed by a thickening and accumulation of verrucae aggregations round the pores to form nearly partial annulus appearance, or a complete thickened ring round the pores. While in case of operculum, it is developed and appeared as lid fixed in appropriate way with the rest of sXine structures, but in other cases, granules of sXine are giving the operculum a partial structure. Pollen grains were classified mainly depending on the presence or absence of the two structures into either annulate or operculate, nonannulate or nonoperculate[4-5,31].

The spine-like projection in sXine 3 seen by LM has been classified as microechinula, while classified by other investigators using SEM as scabrate. The inner surface of the nexine is divided into discrete areas called endocracks, while at this time, this description is completely eliminated and substituted by channels as new term in pollen morphology[4-5,9,11,12,14,15,19,24,25,30].

Many investigators were trying to study the pollen grains as a major component in biological aerosol particles of the atmosphere and their application, others were trying to use pollen grains as a new indicator for pollution. But the major type of investigators were using pollen grains as palynological evidence in palaeobotany[6,9,11,14,15,25,26,30].

The purpose of this palynological study was trying to use the pollen morphology in taxonomy of plantaginaceae for delimitation the different taxonomic ranks from each other depending on concrete palynological evidences for indications since this family was not yet understood palynologically, which was helpful to give an illustrated guide to pollen grains of plantaginaceae.

MATERIALS AND METHODS

Fresh polliniferous material was collected from fresh specimens belonging to 11 plantago species by using collecting brushes and field vials introduced to the laboratory to be studied.

For LM studies, the material was prepared according to acetolysis method of Erdtman. 4 duplicates for each species were prepared and 6 slides from each were made[30].

LM micrographs and measurements were taken by using Nikon HFX-11 microscope using ocular micrometer scale by glycerin Jelly method[31].

For SEM studies, the pollen grains prepared by acetolysis method were coated by carbon layer first, then by gold to increase the conduction and electron yield, micrographs were taken by SEM[31].

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Table 1: Pollen structures and measurements of Plantago species investigated

<table>
<thead>
<tr>
<th>Species</th>
<th>Pollen class (μm)</th>
<th>Annulus</th>
<th>Operculum</th>
<th>Margins</th>
<th>Costae</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plantago lanceolata L.</td>
<td>(6)-10(15) pantoate</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Plantago coronopus L.</td>
<td>(3)-8(10) pantoate</td>
<td>++</td>
<td>0</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>Plantago major L.</td>
<td>(4)-5-10 pantoate</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Plantago maris-mortui Eig.</td>
<td>(4)-5-10 pantoate</td>
<td>0</td>
<td>0</td>
<td>++</td>
<td>0</td>
</tr>
<tr>
<td>Plantago arabica Boiss.</td>
<td>(1)-5(8) pantoate</td>
<td>0</td>
<td>0</td>
<td>++</td>
<td>0</td>
</tr>
<tr>
<td>Plantago ciliata Desf.</td>
<td>3-9 pantoate</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Plantago psyllium L.</td>
<td>(1)-5(8) pantoate</td>
<td>0</td>
<td>0</td>
<td>++</td>
<td>0</td>
</tr>
<tr>
<td>Plantago ovata L.</td>
<td>(3)-6(8) pantoate</td>
<td>++</td>
<td>0</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Plantago arenaria Waldst and Kit</td>
<td>(1)-5(8) pantoate</td>
<td>0</td>
<td>0</td>
<td>++</td>
<td>0</td>
</tr>
<tr>
<td>Plantago amplifica Cav.</td>
<td>3-9 pantoate</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Plantago crusifolia Forsk.</td>
<td>(1)-5(8) pantoate</td>
<td>0</td>
<td>0</td>
<td>++</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 2: Pollen structures and measurements of Plantago species (plantaginaceae)

<table>
<thead>
<tr>
<th>Species</th>
<th>Pollen dia. (μm)</th>
<th>Exine (μm)</th>
<th>Pore (μm)</th>
<th>Annulus (μm)</th>
<th>Shape and Ornamentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plantago lanceolata L.</td>
<td>(20)-24-30(34)</td>
<td>1-2</td>
<td>3-4</td>
<td>2</td>
<td>circular, verrucose with small punctae, secine scabrescences and nexine surface are not well-defined</td>
</tr>
<tr>
<td>Plantago coronopus L.</td>
<td>22-28(30)</td>
<td>2</td>
<td>2-4</td>
<td>2.5</td>
<td>circular, verrucose with few punctae, secine scabrescences and nexine surface are not well-defined</td>
</tr>
<tr>
<td>Plantago major L.</td>
<td>18-27</td>
<td>1-1.5</td>
<td>3-5</td>
<td>-</td>
<td>circular, verrucose without punctae, (Verrucose ++), secine and nexine surfaces are not well-defined</td>
</tr>
<tr>
<td>Plantago maris-mortui Eig.</td>
<td>20-26</td>
<td>1-1.5</td>
<td>3-4</td>
<td>-</td>
<td>circular, verrucose without punctae, (Verrucose ++), secine scabrescences and secine and nexine surfaces are not well-defined</td>
</tr>
<tr>
<td>Plantago arabica Boiss.</td>
<td>22-32</td>
<td>1-2</td>
<td>3-5</td>
<td>-</td>
<td>circular, verrucose with few or no punctae, (Verrucose +), secine scabrescences and nexine surfaces are almost not well-defined</td>
</tr>
<tr>
<td>Plantago ciliata Desf.</td>
<td>(30)-32(40)</td>
<td>1.5-2</td>
<td>4-6</td>
<td>-</td>
<td>circular, verrucose with small punctae, (Verrucose ++), secine scabrescences and nexine surfaces are almost not well-defined</td>
</tr>
<tr>
<td>Plantago psyllium L.</td>
<td>22-32</td>
<td>1-2</td>
<td>3-5</td>
<td>-</td>
<td>circular, verrucose with or without punctae, (Verrucose +), secine scabrescences, and nexine surface are almost not well-defined</td>
</tr>
<tr>
<td>Plantago ovata L.</td>
<td>22-32</td>
<td>1.5-2</td>
<td>3-5</td>
<td>2.5</td>
<td>circular, verrucose with few or no punctae, (Verrucose +), secine scabrescences, and nexine surface are almost not well-defined</td>
</tr>
<tr>
<td>Plantago arenaria Waldst and Kit</td>
<td>22-32</td>
<td>1.5-2</td>
<td>3-5</td>
<td>-</td>
<td>circular, verrucose with few or no punctae, (Verrucose +), secine scabrescences, and nexine surface are almost not well-defined</td>
</tr>
<tr>
<td>Plantago amplifica Cav.</td>
<td>(30)-32(40)</td>
<td>1.5-2</td>
<td>4-6</td>
<td>-</td>
<td>circular, verrucose with few or no punctae, (Verrucose +), secine scabrescences, and nexine surface are almost not well-defined</td>
</tr>
<tr>
<td>Plantago crusifolia Forsk.</td>
<td>22-32</td>
<td>1.5-2</td>
<td>3-5</td>
<td>-</td>
<td>circular, verrucose with few or no punctae, (Verrucose +), secine scabrescences, and nexine surface are almost not well-defined</td>
</tr>
</tbody>
</table>

++: the structure is well-defined  + : the structure is not well-defined  0 : the structure is not present

RESULTS AND DISCUSSION

It is obvious from the results obtained (Table 1 and 2) that the only possible way to delimit the species palynologically is by using the number of pores in each pollen grain because all of the examined species have the same pollen class (Pantorate).

Five Pollen types were distinguished among the species examined (Table 1 and 2 and Fig. 1-5);

- **Plantago lanceolata type**: It has well-developed opercula truly formed, similar to lid fixed in appropriate way. The annuli, margins of ectoaperture, costae of endoaperture are also highly developed and they considered as distinguishing features in pollen grains. This pollen type was represented by one species, Plantago lanceolata L.

- **Plantago coronopus type**: The annuli and ectoaperture margins in this pollen type are highly developed while the opercula and endoaperture costae were poorly developed. This pollen type was represented by 2 species; Plantago coronopus L. and Plantago ovata L.

- **Plantago crusifolia type**: It is with highly developed ectoaperture margins, while the costae were not present. Both the annuli and operculae may or may not present. This pollen type was represented by 4 species from those examined; Plantago arenaria Waldst. kit, Plantago psyllium L., Plantago crusifolia Forsk. and Plantago arabica Boiss.

- **Plantago major type**: It has no annuli and costae. The opercula and ectoaperture surface were poorly developed. This pollen type was represented by 2 species; Plantago maris-mortui Eig and Plantago major L.
Fig. 1(1-6): 1: SEM showing overall view of pollen grain of *Plantago lanceolata* L. (1770x)
2: SEM showing overall view of pollen grain of *Plantago coronopus* L. (1750x)
3: LM showing the optical cross section in pollen grain of *Plantago coronopus* L. (2000x)
4: LM showing the optical cross section in pollen grain of *Plantago major* L. (2000x)
5: SEM showing the pore in pollen grain with operculum and annulus in *Plantago lanceolata* L. (1520x)
6: LM showing the optical cross section in pollen grain of *Plantago marit-morui* L. (2000x)
Fig. 2(1-6): 1: SEM showing the overall view of pollen grain with pore and disintegrated operculum in *Plantago major* L. (1960x)
2: LM showing the pollen grain of *Plantago coronopus* L., it is showing clearly the pore with complete annulus (2000x)
3: LM showing the optical cross section in pollen grain of *Plantago lanceolata* L., pore with operculi are obvious (2000x)
4: LM showing the pore with granular operculum in pollen grain of *Plantago arabis* Boiss. (2000x)
5: SEM showing the pore and ornamentation in pollen grain of *Plantago major* L. (5480x)
6: LM showing the pore with costae in pollen grain of *Plantago citata* Desf. (2000x)
Fig. 3(1-6): 1: SEM showing the overall view of pollen grain of *Plantago pellitum* L., the pores and opercula are not highly focused (190x).
2: SEM showing pore and verrucae in pollen grain of *Plantago pellitum* L. (1730x)
3: LM showing the overall view of pollen grain of *Plantago arabica* Boiss., Verrucae annuli and operculi are seen (2000x)
4: LM showing verrucae and ornamentation in pollen grain of *Plantago pellitum* L. (2000x)
5: SEM showing the pore with annulus and operculum in pollen grain of *Plantago ovata* Forsk. (1490x)
6: LM showing the annulus and radiating nexine channels in pollen grain of *Plantago ovata* Forsk. (2000x)
Fig. 4 (1-6): 1: SEM showing the overall view pores in pollen grain of *Plantago arenaria* Waldst. and Kit. (1995x)
2: LM showing the optical cross section in pollen grain of *Plantago arenaria* Waldst. and Kit. (2000x)
3: LM showing verrucae and ornamentation in pollen grain of *Plantago arenaria* Waldst. and Kit. (2000x)
4: LM showing pore and ornamentation in pollen grain of *Plantago amplexicaulis* Cav. (2000x)
5: SEM showing the overall view with pores in pollen grain of *Plantago amplexicaulis* Cav. (1540x)
6: SEM showing the ornamentation with pores and opercula in pollen grain of *Plantago amplexicaulis* Cav. (4435x)
Fig. 5(1-6): 1: SEM showing the overall view pores in pollen grain of *Plantago crassifolia* Forsk. (2010x)
2: LM showing the optical cross section in pollen grain of *Plantago arenaria* Waldst. and Kit. (2000x)
3: LM showing verrucac and ornamentation in pollen grain of *Plantago ovata* Forsk (2000x)
4: LM showing pore and ornamentation in pollen grain of *Plantago arenaria* Waldst. and Kit. (2000x)
5: LM showing pore and verrucac in pollen grain of *Plantago arenaria* Waldst. and Kit. (2000x)
6: LM showing pore and costae in pollen grain of *Plantago crassifolia* Forsk. (2000x)
• **Plantago ciliata** type: It has no annuli and costae, with poorly developed opercula and ectoaperture margins. This pollen type was represented by 2 species from those examined, *Plantago amplexicaulis* Cav. and *Plantago ciliata* Desf.

With respect to exine structures, all the species examined were similar, they have short and thin columella (sexine 1), variable thickness of tectum (sexine 2), with scabre (sexine 3).

Pollen measurements were variable related to pollen diameter (size), exine thickness, pore diameter and annulus broad (Table 1, 2 and Fig. 1-5).

The using of this palynological study in taxonomy of plantaginaceae to delimit the different species of *Plantago* from each other is useful, because it was obvious from the results, that there were some palynological parameters can be depend upon to distinguish 5 types of pollen grains. They were; the presence or absence and the highly or poorly development of opercula, annulus, surface and margins of ectoaperture and costae of endocaperture.

Depending on the results obtained, the following taxonomic key can be concluded:

- Highly annulate:
  a) Highly operculate.......... *Plantago lanceolata* type
  b) Not highly operculate...... *Plantago coronopus* type

- Poorly annulate:
  a) Pore size up to 40 µm............. *Plantago ciliata* type
  b) Pore size up to 32 µm............. *Plantago crassifolia* type
  c) Pore size up to 27 µm............. *Plantago major* type

It is clear from this study compared to the previous studies that each study using palynological characters as taxonomic evidence in delimitation to certain taxonomic categories has special case be controlled by, accordingly, the palynological characters used in this study may not valid as evidences in taxonomy of other taxonomic ranks, because the palynological characters used by the previous studies are steadily not valid for this study.

**ACKNOWLEDGMENT**

The researcher would like to thank the kind help given by technicians in particular to Christine Gruber from SEM unit in institute of Botany-free University of Berlin, Germany.

**REFERENCES**