Palynological studies in tribe Chlorideae (Poaceae) from salt range of Pakistan

Farooq Ahmad¹*, Mir Ajab Khan², Mushtaq Ahmad², Muhammad Zafar², Ameer Khan³ and Zafar Iqbal⁴

¹Department of Botany, University of Agriculture, Faisalabad, Pakistan.
²Department of Plant Sciences, Quaid-i-Azam University, Islamabad, Pakistan.
³Department of Biological Sciences, University of Sargodha, Pakistan.
⁴Department of Botany, Hazara University, Mansehra, Pakistan

Accepted 3 June, 2011

Pollen morphology of five species belonging to three genera of tribe Chlorideae (Poaceae) was examined by light microscopy (LM) and scanning electron microscopy. The studies showed that pollen in all species were circular in polar view, however, there are variations in equatorial view of pollen and other quantitative characters that is, polar and equatorial diameter, pore diameter and exine thickness, that are valuable in the identification and differentiation of species. Average pollen fertility in the tribe is 77.37%. Verrucate type of sculpturing is found in all species except Tetrapogon villosus, which showed the rugulate type of sculpturing and can be differentiated from Tetrapogon cenchriformis on the basis of its sculpturing pattern. The studies revealed that pollen characters are important in the taxonomy of grasses at the specific and generic level and can be useful in delimiting taxa of different tribes.

Key words: Palynological studies, chlorideae, salt range.

INTRODUCTION

Poaceae is one of the largest families among the angiosperms and is represented in every phytogeographic region in the world, comprising about 10,000 species and 651 genera (Clayton and Renvoize, 1986). The tribe Chlorideae belongs to sub family Chloridoideae, having about 45 genera throughout the tropics in the world; out of these, 7 genera and 15 species are present in Pakistan. In this study, from salt range area, 5 species belonging to 3 genera of this tribe are collected. Two problematic genera Chloris and Tetrapogon have two species each, while Cynodon has one species. Tetrapogon cenchriformis is collected from the area and it is the new report from Pakistan and not previously mentioned in the flora of Pakistan.

Pollen morphology has proved to be a valuable tool in plant taxonomy. Pollen morphology was not considered in the earlier taxonomic studies. Palynology can be helpful in solving problems related to grass systematics and can provide basis for additional features for identification of plant species (Aftab and Parveen, 2006). Pollen morphology of grasses has been studied by Kohler and Lange (1979), Chaturvedi et al. (1994, 1998) and Ma et al. (2001).

In this study, both qualitative and quantitative characters of pollen in problematic species of tribe Chlorideae were studied, to identify and differentiate species at the specific and generic level, as some characters such as grain size and sculpturing pattern are of significance in taxonomy of grasses (Woodehouse, 1935).

MATERIALS AND METHODS

The research work was conducted in the Experimental Taxonomy lab and Herbarium of Quaid-i-Azam University, Islamabad. The research work is confined to palynological studies of 5 species belonging to 3 different genera of tribe Chlorideae collected from salt range of Pakistan.

Light microscopy (LM) and scanning electron microscopy (SEM) was used to study pollen morphology and the terminology used is that of Erdtman (1952) and Moore and Webb (1978). Florets were dissected and anthers were placed on the slide with the help of forceps, added a drop of 45% acetic acid and crushed with iron rod. Pollens were acetolysed according to modified method of Ahmad et al. (2008), who followed Erdtman (1952).
The pollen is circular in polar view and prolate spheroidal in equatorial view. The polar axis diameter is 22.5 µm (15 to 22.5 µm). The P/E ratio is 1.15 and pollen is monoporate and ectoporate. Pore diameter is 1.65 µm (1.25 to 2.0 µm) while the pollen fertility is 79.68%.

Sculpturing is verrucate and verrucae are narrowly spaced (Figure 1c).

Tetrapogon cenchiformis (A. Rich.)Clayton

The pollen is circular in polar view and prolate spheroidal in equatorial view. The polar axis diameter is 24.37 µm (20 to 30 µm) and equatorial axis diameter is 23.5 µm (20 to 27.5 µm). P/E ratio is 1.03. The pollen are exoporate and monoporate. Pore diameter is 2.7 µm (2.5 to 3.5 µm) and exine thickness is 0.95 µm (0.75 to 1.00 µm). Pollen fertility is 89.18%. Sculpturing is verrucate and verrucae are narrowly spaced (Figure 1d).

Tetrapogon villosus Desf.

The pollen is circular in polar view and oblate spheroidal in equatorial view. Polar axis diameter is 24.16 µm (22.5 to 27.5 µm) and equatorial axis diameter is 24.77 µm (22.5 to 30 µm). P/E ratio is 0.97. The pollen are ectoporate and monoporate. The pore diameter is 1.5 µm (1.2 to 1.7 µm) and exine thickness is 0.93 µm (0.75 to 1.0 µm). Pollen fertility is 80%. Sculpturing is rugulate and the rugulae are widely spaced (Figure 1e).

DISCUSSION

Few studies have been conducted on the pollen morphology of Chloridoideae (Huang, 1975). According to Liu et al. (2005), pollen grains in subfamily Chloridoideae are generally radically symmetrical and prolate spheroidal. In this study, all the species in tribe Chlorideae showed circular pollen in polar view. Circular nature of pollen is the structural adaptation of grasses for effective pollination by insects (Gimenis, 1991; Edeoga and Okoli, 1996). Pollen in C. barbata are larger than C. dolicostachya, having polar diameter (23.50 µm) and equatorial diameter (24.37 µm), while pore diameter (1.5 µm) and exine thickness is more in C. dolicostachya than C. barbata. Pollen in both species of Tetrapogon differs with each other in equatorial view, as it is oblate spheroidal in T. villosus and prolate spheroidal in T. cenchiformis. Pollen is large in polar view (24.16 µm) in T. cenchiformis, but small in equatorial view (23.5 µm) than T. villosus. Other quantitative characters such as P/E ratio pore diameter and exine thickness are recorded more in T. cenchiformis. These variations in size pore diameter and exine thickness serve as point of differentiation in different species (Mbagwu et al., 2008).

Siddiqui and Qaisar (1988) studied 4 species of Chlorideae for palynology and average grain size was

RESULTS

The qualitative and quantitative characters of pollen of 5 species belonging to tribe Chlorideae are given as follows:

Chloris barbata Sw.

The pollen is circular in polar view and spheroidal to oblate spheroidal in equatorial view. The polar axis diameter is 23.50 µm (22 to 25.90 µm) and equatorial axis diameter is 24.37 µm (17.5 to 27.5 µm). P/E ratio is 0.95. Pollen is monoporate and ectoporate. Pore diameter is 1.40 µm (1 to 2 µm) and exine thickness is 0.75 µm (0.65 to 0.95 µm). Pollen fertility is 53.84%. Sculpturing is verrucate and verrucae are narrowly spaced (Figure 1a).

Chloris dolicostachya Lag.

The pollen is circular in polar view and spheroidal to oblate spheroidal in equatorial view; the polar axis diameter is 21.89 µm (20 to 25 µm) and equatorial axis diameter is 23.18 µm (20 to 27.5 µm). The P/E ratio is 0.94 while the pollen is monoporate and ectoporate. The pore diameter is 1.5 µm (1.0 to 2.0 µm) and exine thickness is 0.89 µm (0.75 to 1.0 µm). Pollen fertility is 84.16%. Sculpturing is verrucate (Figure 1b).

Cynodon dactylon (Linn.)Pers

The pollen is circular in polar view and spheroidal to sub prolate in equatorial view. The polar axis diameter is 22.5 µm (15 to 22.5 µm) and equatorial axis diameter is 19.54 µm (15 to 22.5 µm). The P/E ratio is 1.15 and pollen is monoporate and ectoporate. Pore diameter is 1.65 µm (1.25 to 2.0 µm) while the pollen fertility is 79.68%.

Sculpturing is verrucate and verrucae are narrowly spaced (Figure 1c).

Tetrapogon cenchiformis (A. Rich.)Clayton

The pollen is circular in polar view and prolate spheroidal in equatorial view. The polar axis diameter is 24.37 µm (20 to 30 µm) and equatorial axis diameter is 23.5 µm (20 to 27.5 µm). P/E ratio is 1.03. The pollen are exoporate and monoporate. Pore diameter is 2.7 µm (2.5 to 3.5 µm) and exine thickness is 0.95 µm (0.75 to 1.00 µm). Pollen fertility is 89.18%. Sculpturing is verrucate and verrucae are narrowly spaced (Figure 1d).

Tetrapogon villosus Desf.

The pollen is circular in polar view and oblate spheroidal in equatorial view. Polar axis diameter is 24.16 µm (22.5 to 27.5 µm) and equatorial axis diameter is 24.77 µm (22.5 to 30 µm). P/E ratio is 0.97. The pollen are ectoporate and monoporate. The pore diameter is 1.5 µm (1.2 to 1.7 µm) and exine thickness is 0.93 µm (0.75 to 1.0 µm). Pollen fertility is 80%. Sculpturing is rugulate and the rugulae are widely spaced (Figure 1e).

DISCUSSION

Few studies have been conducted on the pollen morphology of Chloridoideae (Huang, 1975). According to Liu et al. (2005), pollen grains in subfamily Chloridoideae are generally radically symmetrical and prolate spheroidal. In this study, all the species in tribe Chlorideae showed circular pollen in polar view. Circular nature of pollen is the structural adaptation of grasses for effective pollination by insects (Gimenis, 1991; Edeoga and Okoli, 1996). Pollen in C. barbata are larger than C. dolicostachya, having polar diameter (23.50 µm) and equatorial diameter (24.37 µm), while pore diameter (1.5 µm) and exine thickness is more in C. dolicostachya than C. barbata. Pollen in both species of Tetrapogon differs with each other in equatorial view, as it is oblate spheroidal in T. villosus and prolate spheroidal in T. cenchiformis. Pollen is large in polar view (24.16 µm) in T. cenchiformis, but small in equatorial view (23.5 µm) than T. villosus. Other quantitative characters such as P/E ratio pore diameter and exine thickness are recorded more in T. cenchiformis. These variations in size pore diameter and exine thickness serve as point of differentiation in different species (Mbagwu et al., 2008).

Siddiqui and Qaisar (1988) studied 4 species of Chlorideae for palynology and average grain size was
recorded, 18.59 to 31.46 \mu m, while in this study, pollen size ranged from 15 to 30 \mu m. In \textit{C. dactylon}, pollen are spheroidal to sub prolate in equatorial view and pore diameter (1.65 \mu m) in this species more than the otherspecies studied in the tribe except \textit{T. cenchriformis} which has maximum pore diameter and grain size than other species. Siddiqui and Qaisar (1988) found the grain size in \textit{C. dactylon} (35.14 \mu m), but in this study pollen diameter in this species is 15 to 22.5 \mu m. Parveen (2006) studied \textit{Cynodon arculatus}, having pollen diameter from, 19.74 to 24.71 \mu m. According to her observations, pollen are spheroidal and monoporate. Pollen is also
Table 1. Pollen characters of different species in tribe Chlorideae.

<table>
<thead>
<tr>
<th>Species name with voucher number</th>
<th>Shape in polar view</th>
<th>Shape in equatorial view</th>
<th>Sculpturing pattern</th>
<th>Polar axis diameter (µm)</th>
<th>Equatorial axis diameter (µm)</th>
<th>P/E ratio (µm)</th>
<th>Pore diameter (µm)</th>
<th>Exine thickness (µm)</th>
<th>Pollen fertility (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Chloris barbata</em> (369)</td>
<td>Circular</td>
<td>Spheroidal to oblate spheroidal</td>
<td>Verrucate</td>
<td>23.50 (22-25.90)</td>
<td>24.37 (17.5-27.5)</td>
<td>0.95</td>
<td>1.40 (1.0-2.0)</td>
<td>0.75 (0.65-0.95)</td>
<td>53.84</td>
</tr>
<tr>
<td><em>Chloris dolicostachya</em> (330)</td>
<td>Circular</td>
<td>Spheroidal to oblate spheroidal</td>
<td>Verrucate</td>
<td>21.89 (20-25)</td>
<td>23.18 (20-27.5)</td>
<td>0.94</td>
<td>1.5 (1.0-2.0)</td>
<td>0.89 (0.75-1.0)</td>
<td>84.16</td>
</tr>
<tr>
<td><em>Cynodon dactylon</em> (17)</td>
<td>Circular</td>
<td>Spheroidal to sub prolate</td>
<td>Verrucate</td>
<td>18.5 (15-22.5)</td>
<td>19.54 (15-22.5)</td>
<td>1.15</td>
<td>1.65 (1.25-2.0)</td>
<td>0.75 (0.6-1.0)</td>
<td>79.68</td>
</tr>
<tr>
<td><em>Tetrapogon cenchriformis</em> (268)</td>
<td>Circular</td>
<td>Prolate spheroidal</td>
<td>Verrucate</td>
<td>24.37 (20-30)</td>
<td>23.5 (20-27.5)</td>
<td>1.03</td>
<td>2.7 (2.5-3.5)</td>
<td>0.95 (0.75-1.0)</td>
<td>89.18</td>
</tr>
<tr>
<td><em>Tetrapogon villosus</em> (32)</td>
<td>Circular</td>
<td>Oblate spheroidal</td>
<td>Rugulate</td>
<td>24.16 (22.5-27.5)</td>
<td>24.77 (22.5-30)</td>
<td>0.97</td>
<td>1.5 (1.2-1.7)</td>
<td>0.93 (0.75-1.0)</td>
<td>80</td>
</tr>
</tbody>
</table>

monoporate and ectoporate in this study, as the number and position of aperture is of prime significance in palynology (Ogbebor, 1996). Variations are found in equatorial view as spheroidal to oblate spheroidal pollen are found in different species, even differences are found in two species of same genus that is, *Tetrapogon*, but in genus *Chloris* both species are similar in equatorial view. So these variations observed in qualitative as well as qualitative characters may be helpful in the identification of different species and genera in the tribe. All the species in this tribe showed the verrucate type of sculpturing except *T. villosus* which has regulated type of sculpturing; hence, sculpturing pattern is an important tool to differentiate *T. villosus* from other species of the genus and tribe.

Pollen fertility ranges from 53.84 to 89.18% (Table 1). The average pollen fertility in the tribe Chlorideae is 77.37%. Maximum pollen fertility is recorded in *T. cenchriformis*. The degree of fertility of hybrids may give some indication of the degree of relationship between its parents. In general, hybrids between species of a genus that are not closely related tend to be sterile or of low fertility, whereas hybrids between taxonomically more closely related species or intra-specific taxa tend to be more fertile (Khan, 1991). Thus, there is a correlation between hybrid fertility and taxonomic relationship. Pollen fertility is a valuable tool in taxonomic studies to distinguish putative hybrids from the parent plants and is also useful to determine the degree of fertility in those plants that are grown under unfavorable condition (Lawrence, 1951). The genetic variations of a flora can be observed by studying their pollen fertility (Tellaria, 1991). It is a helpful tool to assess the stability of species in a particular area.

**Conclusion**

This study shows that all the species present in tribe Chlorideae have circular pollen. In *Chloris* and *Tetrapogon*, variations are observed in qualitative and quantitative characters of pollen. These variations are valuable in the identification of species. Verrucate type of sculpturing pattern is observed in the tribe except *T. villosus*. Maximum pollen fertility is observed in *T. cenchriformis*, it is concluded from this study that variations exist in qualitative and quantitative characters of pollen, in different species of the tribe and sculpturing pattern in genus *Chloris* that are helpful in
identification, differentiation and delimiting of different taxa.

REFERENCES