J. Agric. Sci. Mansoura Univ., 27(5): 2987 - 3004, 2002

COMPARATIVE ULTRASTRUCTURE STUDIES OF LEAF SURFACE FEATURES OF SOME POACEAE GENERA.

El-Sahhar, K.F.; A.M. Khattab and H.A. Abd Al-Mageed
Agricultural Botany Department, Fac. of Agric., Cairo Univ., Giza, Egypt.

ABSTRACT

Phenetic relationships of some Poaceaus genera were studied. Plants representing these genera were; Quack grass, Wild oat, Barley, Rice, Millet or wild proso, Canary grass, Sorghum, Johnson grass, Wheat and Corn. Comparative anatomy of surface features of the leaves was used as a taxonomic evidence to establish their phenetic relationships.

Scanning electron microscopy technique (SEM) was used to compare the leaf suture features of the above plants. The studies concentrated on some of these features; *i.e.* short cells, long cells, interstomatal cells, silica-bodies, stomata and hair types. Through the observations, it was possible to detect the taxonomical relationships among the studied taxa, which could be summarized as follows; some taxa showed greater similarity to each other due to sharing most of the examined features; *i.e.* Rice, Millet, Corn, Johnson grass, Wheat and Canary grass. Some other taxa exhibit similarity but to a lesser degree; *i.e*. Barley and Wild oat, Rice and Quick grass, Barley and Canary grass, Millet and Wild oat and finally, Rice and Sorghum. On the contrary, The third pattern of taxa which were far away from each other due to sharing only in one to three characters; *i.e.* Sorghum either with Canary grass or with Johnson grass.

INTRODUCTION

Poaceae (Gramineae) are well-defined family of 6 sub-families, 25 tribes, 500 genera and 8000 species. The family distributed through tropical, north-temperate and semi-arid regions (Cronquist, 1981 and Jones and Luchsinger, 1987).

The family has greater economic importance than any other family of the flowering plants. The reasons for that are; the plants used, as food crops for human consumption (Rice, Wheat, Corn and Barley), forage and grain for animals, industrial uses (starch and ethyl alcohol), shelter-bamboo, soil conservation, ornamentals and wild-life food.

Relationships among Gramineous plants have faced many problems to the taxonomists using traditional taxonomic methods based on gross morphology (Metcalfe, 1960 and Radford *et al.*, 1974). For over a century, taxonomy has used comparative anatomy of surface features of various organs, *i.e.* leaves to aid in classification. Several principles on the use of anatomical data have been published (Metcalfe, 1963; Radford *et al.*, 1974 and Jones and Luhsinger, 1987). It has become important to carry out this study, which aimed to conduct taxonomic investigations on some genera of Poaceae using the comparative anatomy of surface features of the leaves of plants representing these genera.

Scanning Electron Microscopy (SEM) technique was applied to study trichomes and distribution patterns, stomatal types and epidermal cells. This study was conducted in an attempt to disclose any relationship exists among

the investigated taxa. Through the investigations, the similarity and dissimilarity among studied grass taxa were taken into consideration, because the most important diagnostic characters are found in the leaf, *i.e.* long and short cells, interstomatal cells, silica-bodies, stomata and hair types. The reasons for that as Metcalfe (1960) reported are as follows: 1) There are minor anatomical variations within a single leaf-blade, 2) Leaves from different levels on an individual plant exhibit structural variations, 3) Leaves from plants belonging to a single species grown in different habitats also vary within limits and 4) The interspecific differences are very rare and many of them quantitative rather than qualitative.

MATERIALS AND METHODS

In this study, comparative anatomy of leaf surface features by means of Scanning Electron Microscopy (SEM) was used. These features included short cells, long cells, silica-bodies, interstomatal cells, stomata and hair types. Authentic grain samples representing 10 grass taxa were purchased from Scientific Commercial Sources, University of Illinois, Illinois, U.S.A.. These taxa are; *Agropyron repens* (L.) Beauv. (Quack grass), *Avena fatua* L. (Wild oat), *Hordeum vulgare* L. (Barley), *Oryza sativa* L. (Rice), *Panicum mallianceum* L. (Millet or wild proso), *Phalaris canariensis* L. (Canary grass), *Sorghum bicolor* (L.) Moench. (Sorghum), *Sorghum halepense* (L.) Pers. (Johnson grass). *Triticum aestivium* L. (Wheat) and *Zea mays* L. (Corn).

Germination was carried out under green house conditions. Grains were sown in plastic trays 50 x 30 cm containing vermiculate irrigated with distilled water and Hoagland solution (0.5 m of stock solution of macro-and-micro nutrients) (Hoagland and Arnon, 1950). Three weeks later, the lamina of the second leaf was cut into small pieces, each of 0.5 cm^2 by a razor blade, and then placed on the specimen holders stub with double sticky carbon adhesive discs. The specimens were mounted on heated aluminum stub to remove excess water (Berlyn and Miksche, 1976). Then scanned by Scanning Electron Microscopy model ISI-40 voltage. Both leaf surfaces were scanned at various magnifications. A photograph for 100 µ of specimens was made by using polariod positive / negative 4x5 instant sheet film according to manufacturer recommendations procedures. Prints were coated immediately after processing and left separated for few minutes to dry, then stored till surface features of the leaves were recorded.

RESULTS AND DISCUSSION

Details of diagnostic anatomical description of each taxon are summarized in Table (1) and illustrated in Figures 1 to 10.

Table 1

1c

It is worthy to notice from Table (1) that, *Agropyron repens, Avena fatua, Oryza sativa, Panicum mallianceum, Phalaris canariensis* and *Zea mays* (Figs. 1, 2, 4, 5, 6 &10) are sharing in the rectangular shape of short cells on both upper and lower surfaces of the leaf. In contrast, *Hordeum vulgare* and *Triticum aestivium* (Figs. 3 & 9) short cells on both leaf surfaces are hexagonal in shape. It was difficult to observe a distinct shape for these cells in both species of *Sorghum; bicolor* and *halepense* (Figs. 7 & 8). Solitary short cells are inapplicable in all the studied taxa. The presence of these cells in pairs was found on both leaf surfaces of *Agropyron repens, Avena fatua, Hordeum vulgare, Triticum aestivium, Zea mays* and the lower surface of leaf of *Panicum mallianceum* (Figs.1, 2, 3, 5, 9 & 10), while it was inapplicable in the other taxa. Short cells occurred in rows, either on the upper or lower surfaces of leaf of *Panicum mallianceum, Phalaris canariensis, Oryza sativa* and *Zea mays.* (Figs. 4, 5, 6 & 10).

Long cells was rectangular in shape in case of *Agropyron repens, Avena fatua, Hordeum vulgare, Oryza sativa, Panicum mallianceum, Zea mays* (Figs. 1, 2, 3, 4, 5 & 10) and the lower surface of both *Sorghum* species (Figs 7 & 8), while this shape was hexagonal on both upper and lower surfaces of leaf in *Phalaris canariensis, Triticum aestivium* (Figs. 6 & 9) and on the upper leaf surface only in both species of *Sorghum*. The side wall of the long cells was thick in *Agropyron repens, Hordeum vulgare* and *Panicum mallianceum* (Figs. 1, 3 & 5), while it was thin in *Avena fatua, Oryza sativa, Phalaris canariensis, Sorghum bicolor, Sorghum halepense* (Figs. 2, 4, 6, 7 & 8). In *Triticum aestivium* (Fig. 9), the side wall of long cell was inapplicable, while it was sinuous pitted in *Zea mays* on both leaf surfaces. The end wall of long cells, either on upper or lower surfaces of *Oryza sativa* (Fig. 4) was concave, while this wall was rounded in all other taxa.

Interstomatal cells shape was rectangular in *Agropyron repens, Oryza sativa,* both *Sorghum* species and *Zea mays* (Figs. 1, 4, 7, 8 & 9), while these cells were hexagonal in shape in *Hordeum vulgare, Phalaris canariensis, Triticum aestivium* (Figs. 3, 6 & 9) and the upper surface of *Panicum mallianceum* (Fig. 5). The tall and narrow shape of interstomatal cells was observed only in *Avena fatua* (Fig. 2). The thickest wall of interstomatal cells was recorded in *Agropyron repens* and *Hordeum vulgare* (Figs. 1 & 3), while the wall of these cells was thin in all the other taxa. The side wall of interstomatal cells was sinuous in shape on both leaf surfaces of *Oryza sativa, Panicum mallianceum,* both species of *Sorghum* and *Zea mays* (Figs.4, 5, 7, 8 & 10), In the other taxa, it was difficult to detect any shape for the side wall of interstomatal cells. Concave shape of the end wall of the interstomatal cells was the obvious shape on both leaf surfaces of all the studied taxa, except on both leaf surfaces of *Triticum aestivium* (Fig. 9), where the rounded-shape was recorded.

Silica-bodies shape on upper and lower surfaces of leaf was elongated in *Agropyron repens, Hordeum vulgare* and *Panicum mallianceum* (Figs. 1, 3 & 5). This shape was rectangular in *Zea mays* (Fig. 10), while the tall and narrow shape of silica-bodies on both leaf surfaces was recorded in both species of *Sorghum.* It was difficult to observe any shape of silica-bodies in *Avena fatua, Phalaris canariensis* and *Triticum aestivium* (Figs. 2, 6 & 9)

and the variation in shape of silica-bodies on both leaf surfaces was great in *Oryza sativa* (Fig. 4).

The greatest number of stomata among all the studied taxa was recorded on the upper leaf surface of *Oryza sativa* (21), followed by *Sorghum bicolor* and *Zea mays* (10) on the same surface. The later also shows great number of stomata (9) on the lower surface, which was the same number on the same surface of *Sorghum halepene*. The smallest number of stomata on the upper leaf surface was noticed in *Avena fatua*. Triangular shape of subsidiary wall was observed on the upper and lower surfaces of *Panicum mallianceum, Oryza sativa,* both *Sorghum* species and *Zea mays.* While the parallel-sided shape was in *Avena fatua, Hordeum vulgare, Phalaris canariensis* and *Triticum aestivium*. In *Agropyron repens* the low-dome shape of subsidiary wall on both leaf surfaces was recorded.

Macro-hairs were inapplicable on both leaf surfaces of most species studied, except in *Avena fatua, Panicum mallianceum* and *Triticum aestivium* (Figs 2, 5 & 9), where these hairs are present. The last two taxa have also micro-hairs on both leaf surfaces, in addition to *Agropyron repens* and both species of Sorghum (on lower surface only) (Figs. 1, 7 & 8). Micro-hairs were inapplicable in the other taxa. Again, *Triticum aestivium* (Fig. 9) has the third type of hair; the prickle-hairs, on both leaf surfaces. The same in *Agropyron repens, Avena fatua* and *Phalaris canariensis* (Figs. 1, 2 & 6). In *Oryza sativa* the papillae shape of prickle hairs on both surfaces was noticed. There were no prickle hairs in the other taxa.

From the abovmentioned table and figures, the results indicate that:

A) Although, *Agropyron repens*, *Avena fatua* and *Zea mays* belong to different genera, they are sharing many leaf features, *i.e.* shape of short and long cells, shape of end wall of long cells and shape of end wall of interstomatal cells. On the contrary, it could be differentiated among these taxa by using the silica-bodies shape, stomata number and the presence or absence of all hair types.

B) Both species; *Hordeum vulgare* and *Triticum aestivium* are simillar to each other in many studied features, *i.e.* short cell shape, interstomatal cell shape, end wall shape of long cell and subsidiary wall shape. While, these species vary in shape of silica-bodies and the presence or absence of hair types.

C) *Sorghum bicolor* and *Sorghum halepense* are mostly similar to each other sharing many leaf features, except number of stomata on upper and lower surfaces of leaf, where they vary.

D) It could be distinguished among *Panicum mallianceum, Phalaris canariensis* and *Oryza sativa* by using some leaf features, *i.e.* shape of silica-bodies, number of stomata and presence or absence of hair types on both leaf surfaces.

Metcalfe (1960) almost agreed with the present results concerning the stomata characters as well as the features of silica-bodies and shape of micro-hairs. In contrast, he disagreed with the results of few characters in some taxa, *i.e.* short cells in Rice, Canary grass, Sorghum and Wheat; silica-bodies in Sorghum and micro-hairs in Rice and Corn. However, the rest of the studied features were not reported.

1

3

٣٠٠٠

Results of Watson *et al.* (1985) are in harmony with the description of the epidermal characters in this study; short cells, long cells, interstomatal cells, silica-bodies and stomata of most of the studied taxa. However, the recorded characters of hairs of Quack grass, Sorghum, Johnson grass and Wheat contradicted their findings.

Tzelev (1989) studied the epidermal hairs of some grass plants. His description of micro hairs of Quack grass and Wheat are in agreement with that given in this study. On the other hand, his results on Wild oat, Barley, Rice and canary grass disagreed with the present findings.

Dahlgren *et al.* (1984) studied the epidermal features of subfamily Pooideae, which include 6 of the taxa considered here in this study. Their observations were in harmony with those given in the current study. It is worthy to mention that, according to their results; Millet has cross-shaped silica-bodies, while they showed elongated-shape in the present findings.

In essence, the following relationships are concluded; some of the studied taxa showed greater similarity to each other as they shared most of the recorded characters; *i.e.*, Rice, millet, Corn, Johnson grass, Wheat and Canary grass. Some other taxa showed similarity but to a lesser extent; *i.e.*, Barley and Wild oat, Rice and Quack grass, Barley and Canary grass, Millet and wild oat and Rice and Sorghum. In contrast, some other taxa were far away from each other since they only shared one to three of studied characters; *i.e.*, Sorghum and Canary grass and Sorghum and Johnson grass.

REFERENCES

Berlyn, G.P. and J.P. Miksche (1976). Botanical micro-technique and cytochemistry. The Iowa State University Press/Ames, Iowa p. 116-117.

Cronquist, A. (1981). An integrated system of classification of flowering plants. Columbia University Press, N.Y., p. 1142-1147.

Dahlgren, R.M.; H.T.Clifford and P.F.Yeo (1984). The families of the monocotyledons. Springer-Verlag, Berlin Heidelberg N.Y., p. 425-460.

Hoagland,D.R. and D.I., Arnon (1950). The water culture method for growing plants without soil. Calif. Agr. Expt. Sta. Bul. 347 pp.

Jones, S.B. and A.E. Luchsinger (1987). Plant Systematics. Mc. Graw- Hill, Inc., p. 436-438.

Metcalfe, C.R. (1960). Anatomy of the Monocotyledons. Vol. 1 Gramineae. Clarendon Oxford. 731 pp.

Metcalfe, C.R. (1963). Comparative anatomy as a modern botanical discipline. Adv. Bot. Res., p. 101-147.

Radford, A.E.; W.C. Dickison; J.E. Massey and C.R. Bell (1974). Vascular Plant Systematics. Harber & Row Publishers.

Tzelev, N.N. (1989). The system of grasses (Poaceae). and their evolution. The Bot. Rev. 55: 141-203.

Watson, L.; H.T. Clifford and M.J. Dallawitz (1985). The classification of Poaceae subfamilies and subtribes. Aust. J. Bot. 33:433-484.

دراسات فوق مجهرية مقارنة لخصائص سطح أوراق بعض أجناس الفصيلة النجيلية.

قاسم فؤاد السحار – عادل محمود خطاب – حجاج عوض عبد المجيد
قسم النبات الزراعى – كلية الزراعة – جامعة القاهرة – جيزة – مصر

أجريت دراسة عن العلاقات التقسيمية المظهرية بين بعض اجناس من الفصيلة النجيلية وكانت النباتات الممثلة لتلك الاجناس هى: الثيل والزمير والشعير والارز والدخن وحب العصافير والذرة الصفراء وحشيشة الفرس والقمح والذرة الشامية. وقد أشتملت الدراسة على التشريح المقارن لصفات سطح الاوراق (الخلايا القصيرة – الخلايا الطويلة – أجسام السليكا – الخلايا بين الثغور – الثغور – الشعيرات) كدليل تقسيمى لمعرفة العلاقات بين الاجناس تحت الدراسة باستخدام المجهر الالكترونى المساح SEM.

وقد أمكن من خلال هذة الدراسة أيجاد بعض العلاقات التقسيمية للنباتات المدروسة يمكن تلخيصها كما يلى:

وجود تشابة كبير بين بعض الاجناس نتيجة لاشتراكها في معظم الصفات. مثال الارز والذرة - الدخن وحشيشة الفرس - القمح وحب العصافير. كما وجد تشابها بدرجة أقل بين بعض الاجناس مثال الشعير مع الزمير وحب العصافير - الارز مع الثيل والذرة الصفراء - الدخن مع الزمير. ووجد أن بعض الاجناس تتشابة مع بعضها فى صفة الى ثلاث صفات فقط مثال الذرة الصفراء مع حب العصافير وحشيشة الفرس.