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Studies on the Rosaceae I- Seed and/or Achene Macro and Micromorphology

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Abstract: The ultrastructure of seed coats and/or achenes using the SEM and certain selected macromorphological characters were performed on 47 taxa of the Rosaceae senu lato. The taxa were selected to represent the accepted four subfamilies in the family (Maloideae, Prunoideae, Rosoideae and Spiraeoideae) and most of the tribes included in them. The data were numerically analyzed by the UPGMA cluster analysis using the NTsys - pc (Rohlf, 1993). The results revealed that although the family as a whole represents a clearly monophyletic lineage; yet, the study did not support the traditional suprageneric classification of the family that was based on fruit type alone. The studied taxa were distributed across the constructed phenograms independent of the previous suprageneric classification, particularly in members of the subfamilies Rosoideae and Spiraeoideae. These results were in consistence with current studies on the family utilizing molecular criteria. These studies showed clearly that the molecular data did not support the traditional suprageneric classification of the family and that a revision was needed particularly in the Spiraeoideae and Rosoideae.

Key words: Rosaceae - seeds or achene - SEM - macro and micromorphology

INTRODUCTION

The *Rosaceae* is a large sub-cosmopolitan family of about 95 genera and 2825 species. It is located mainly in the temperate and warm areas of the Northern Hemisphere (Mabberley, 1997). However, Heywood, 1993 stated that the family consists of 122 genera and 3370 species.

Among the large genera in the family are *Potentilla* L. (500 species), *Prunus* L. (450 species) and *Crataegus* L. (200 species) (Jones and Luchsinger, 1987). The *Rosaceae* is represented in the wild Egyptian flora by about seven taxa belonging to six genera *Crataegus azarolus* L., *Crataegus sinaica* L., *Cotoneaster orbicularis* Schltall., *Potentilla supinia* L., *Rosa arabica* Crep., *Rubus sanetus* Schreb., *Sanguisorba minor* Scop. Of these taxa, only *Potentilla supinia* L. is very common, while the other are either of rare or very rare occurrence (Tackholm, 1974 and El-Hadidi and Fayed, 1995). This poor representation of the family in the wild flora of Egypt is almost compensated by the large number of cultivated taxa.

In all the systems of classification, the *Rosaceae* belong to the order *Rosales*, yet the families included in this order differ greatly either in number or in identity or both depending on the author opinion on the reliability of the used characters.

The classification of the *Rosaceae* itself raises many problems. Opinions differ as to the relation of the family with other families, the delimitation of its subfamilies, tribes, genera and even species (especially in genera

where sub sexual or asexual reproduction is the normal). Limits cannot be drawn sharply between many of the apparent genera or species. Some have splitted the family to more than 27 separate families. Many of the controversies regarding the family are of long standing and are not near solution today more than they were at the time of Linnaeus in the seventeenths century (Lawrence, 1951; Hutchinson, 1973; Heywood, 1993; Mabberley, 1997 and Judd *et al.*, 1999).

The significance of seed structure in taxonomic and phylogenetic studies has been emphasized by many authors (Netolitzky 1926, Martin 1946, Duke 1961, Corner 1976 and 1992, Rezk 1980 and 1987). SEM of seed coat surface is useful in the identification and classification of various taxa belonging to different families (Stant, 1973; Brisson and Peterson, 1976; Barthlott, 1981 and Boesewinkel and Bouman, 1984). A comparison of surface scan patterns of the seed coat has efficiently been used in studying species of some genera including *Vigna* (Kumar *et al.*, 1984), the *Abutileae* (Khushk and Vaughan, 1986), the *Vicieae* (Chernoff *et al.*, 1992) and *Ranunculus* (Xuhan and Van-Lammeren, 1994). However, as for as the authors are aware, no SEM studies on Rosaceae seed and/or achene coat were made.

The present study was carried out on 47 samples of seeds collected from different parts of the world. This study aims at using seed or achene characters (macro and micromorphological) including SEM of seed or achene coat surface together with characters from vegetative

morphology and numerical taxonomic methods to throw some light on the suprageneric classification of the *Rosaceae*.

MATERIALS AND METHODS

Seeds of the examined species and their sources are listed in Table 1. The macromorphological characters and general aspects were either described from live and herbarium specimens or compiled from Bailey (1949), Valentine and Chater, (1972), Bailey and Bailey (1976), Beckett (1983), Brickell (1998) and Watson and Dallwitz, (2002). Voucher specimens are kept at the Herbarium of Biological Sciences and Geology Department, Faculty of Education, Ain Shams University.

Table 1: Names of the studied species of the Rosaceae and its sources

| Table 1: Names of the studied species of the Rosaceae and its sources | | |
|---|--------------|------------------|
| Species | Subfamily | Source |
| Aruncus dioicus (Walt.) Fern. | Spiraeoideae | Japan |
| Gillenia trifoliata (L.) Moench. | " | Germany |
| Spiraea albiflora (Miq.) Zabel. | " | Italy |
| Spiraea betulifolia Pall. | " | Italy |
| Spiraea chamaedryfolia L. | " | Italy |
| Spiraea nipponica Maxim. | " | Italy |
| Spiraea salicifolia L. | " | Italy |
| Spiraea sargentiana K. Koch. | " | Italy |
| Exochorda korolkowii Lav. | " | Switzerland |
| Exochorda racemosa Lindl. | " | France |
| Filipendula ulmaria (L.) Maxim. | Rosoideae | Switzerland |
| Filipendula vulgaris Moench. | " | Austria |
| Rhodotypos scandens (Thunb.) Makino. | " | France |
| Fragaria nipponica Makino. | " | Germany |
| Fragaria vesca L. | " | France |
| Geum rivale L. | " | Switzerland |
| Geum urbanum L. | " | Switzerland |
| Potentilla argyrophylla Wall. ex. Lehm. | " | Germany |
| Potentilla concinna A. Gray. | " | Germany |
| Rubus grayanus Maxim. | " | Japan |
| Rubus parvifolius L. | " | Japan |
| Acaena saccaticupula Bitter | " | Germany |
| Alchemilla fissa Gunth. and Schumm. | " | Germany |
| Sanguisorba minor Scop. | " | Germany |
| Rosa canina L. | ,, | Italy |
| Rosa gallica L. | " | Italy |
| Rhaphiolepis ovata Briot. | Maloideae | Austria |
| Rhaphiolepis umbellata (Thunb.) Makino. | Waterone cae | Austria |
| Cotoneaster salicifolius Franch. | ,, | England |
| Cotoneaster simonsii Baker. | ,, | England |
| | ,, | _ |
| Crataegus cuneata Siebold. and Zucc. | ,, | Japan England |
| Crataegus monogyna Jacq. | ,, | England |
| Pyracantha angustifolia Franch. | ,, | England |
| Pyracantha coccinea M. Roe | ,, | England |
| Pyracantha crenulata D. Don Roem. | " | England |
| Sorbus commixta. Hedl. | | Japan |
| Sorbus sorbifolia (Poir.) Hedl. | " | Japan |
| Photinia wrightiana Maxim. | " | Japan |
| Mespilus germanica L. | " | England |
| Malus sylvestris Mill. | " | Egypt |
| Amelanchier ovales Medik. | " | France |
| Amelanchier rotundifolia Dum. Cours. | " | Spain |
| Prunus laurocerasus L. | Prunoideae | Egypt |
| Prunus domestica L. C. K. Schneid. | " | Egypt |
| Prunus amygdalus Batsch. | " | England |
| Prunus armeniaca L. | " | France |
| Pyrus communis L. | Maloideae | Egypt |

The general features of the large–sized seeds were examined by binocular stereomicroscope. This microscope belongs to the National Research Center, Dokki, Cairo. The magnification power was expressed by (x) for each photograph. The detailed morphological features of the other seeds were examined by SEM using magnification of x = 750. The seeds were mounted with colloidal silver or carbon on copper stubs and coated with a thin layer of gold in polaron E 5000 sputter coater. The specimens were then examined by a JEOL. Scanning microscope (SEM) at the National Research Center Dokki, Cairo. The terminology used was after Stearn (1966) and Corner (1976).

For the data analysis, the total number of the recorded characters (176) in each taxon, were scored, combined together in three sets of data and coded for creating the data matrix of computation:

- (a) Morphological characters of whole plant.
- (b) Morphological characters of seed or achene coats (SEM).
- © All characters combined.

The presence or absence of each 176 different characters was treated as a binary character in a data matrix i.e. coded 1 and 0 respectively. The relationships between the taxa studied, expressed by average taxonomic distance (dissimilarity), have been demonstrated as phenograms, based on the analysis of the recorded characters using the NTsys program package for IBM-pc as described by Rohlf (1993).

RESULTS AND DISCUSSION

The traditional suprageneric classification of the *Rosaceae* into minor categories was based on only few characters as fruit type and base chromosome numbers (Focke, 1894 and Heywood, 1993). In the present study, the incorporation of additional morphological criteria in the numerical analysis did not alter the basic traditional scheme of the four subfamilies and the taxa included in them, except in minor details. These additional macromorphological criteria included; leaf form and venation, inflorescence type, flower structure and hypathium shape.

Rhodotypos scandens (Rosoideae) was separated from the remaining taxa at the average taxonomic distance of 2.00 due to its possession of some characteristic features (a tetramerous flower with a discoid annular hypanthium and a drup type of fruit). Focke (1894), separated this genus in a separate tribe (Kerrieae) mainly on the basis of its possessing distinct stipules, an axis not forming part



Fig. 1: Seed and/or achene shape of the sutdied taxa of subfamily spiraeodeae (1-10) and subfamily Rosoideae (11-24)



Fig. 2: Seed and/or acghene shape of the studied taxa of subfamily *Rosoideae* (25-26) and subfamily *Maloideae* (27-42& 47) and subfmily *Prunoideae* (43-46)

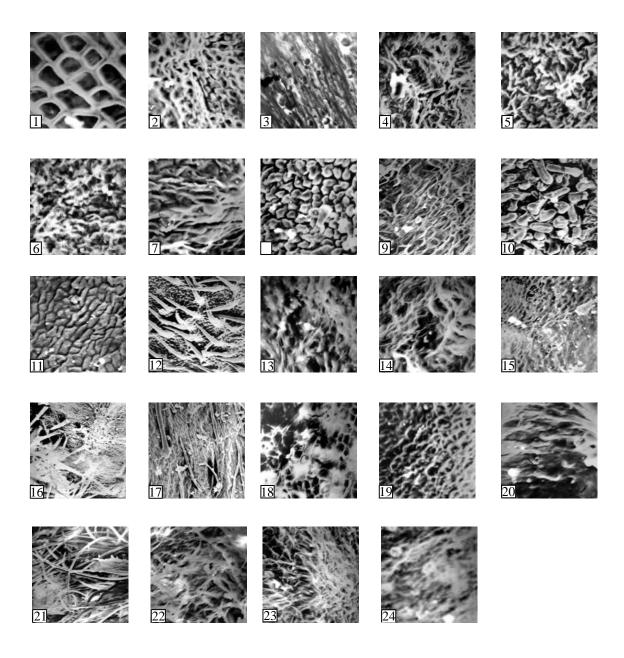


Fig. 3: Seed and/or achene micromorphology of the studied taxa of subfamily *Spiraeoideae* (1-10) and subfamily *Rosoideae* (11-24)

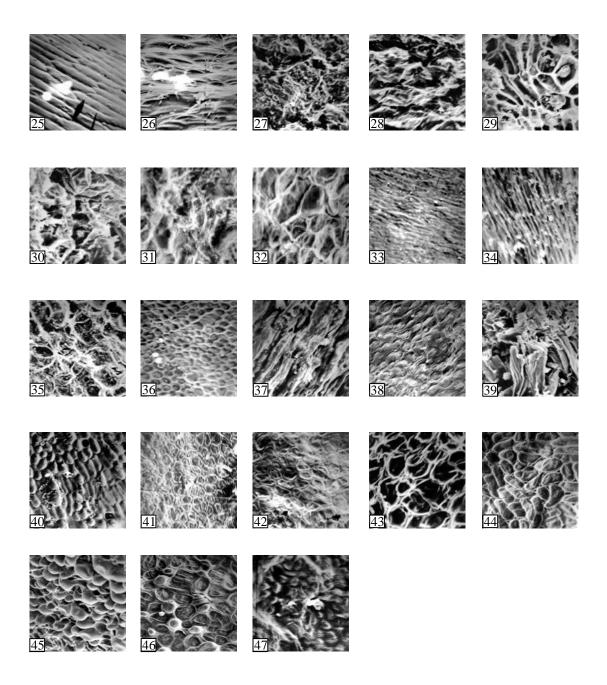


Fig. 4: Seed and/or achene micromorphology of the studied taxa of subfamily *Rosoideae* (25-26), subfamily *Maloideae* (27-42 & 47) and subfamily *Prunoideae* (43-46)

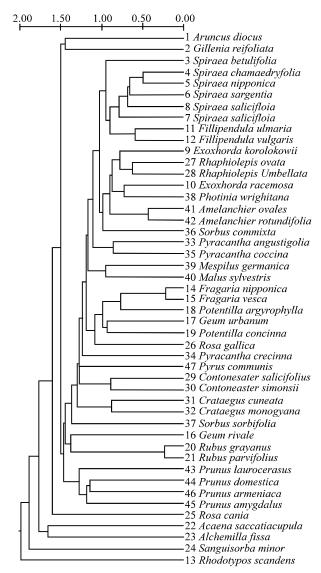


Fig. 5: UPGMA-phenogram based on 79 macromorphological characters illustrated the average taxonomic distance (dissimilarity) between the studied species of the *Rosaceae*

of the fruit, stamens tapering up words from a broad base and carpels that are few and whorled.

The constructed phenogram (Fig. 7) showed also that five of the studied species of *Spiraea* had some relationships with both species of *Filipendula* (*Rosaceae-Ulmareae*). This was mainly due to their sharing some characters as their racemose inflorescences, their pentamerous flowers each with a bell-shaped hypanthium bearing a disk. Each flower in these taxa also had numerous stamens and few carpels. On the other hand, *Spiraea albiflora* was segregated from the

Filipendula - Spiraea group mainly because it differed from them in the following aspects; its possessing a subshrubby habit, a pubescent texture and a lanceolate leaf blade. This result shows that Spiraea L. as currently delimited, may be a heterogenous assembly as suggested by Judd et al. (1999). The delimitation of Spiraea still posses many problems and the exact number of species may be between 80-100 (Mabberley, 1997).

A relationship was recorded between *Exochorda korolkowii* (*Spiraeoideae-Exochordeae*) and the two studied species of *Rhaphiolepis* (*Maloideae*). Both clustered at the dissimilarity level of 0.77 mainly due to their shrubly habit, glabrous texture and simple obovate leaf blade, racemose inflorescence each with 10-20 stamens and 3-5 carpels, while the other studied species *Exochorda racemosa* are clustered with *Photinia wrightiana* (*Maloideae*) mainly due to their shruby habit, simple oblong to obovate leaf blade with serrate margin, pentamerous flower in racemose inflorescence. Each flower with 10-20 stamens and 3-5 carpels.

The relationship of the Maloideae with members of the Spiraeoideae were shown earlier by Campell et al. (1995). According to Evans and Dikinson (1999), a study based on cladistic analysis showed that the members of the Spiraeoideae might be sister taxa to an enlarged Maloideae. clade. However, the survey on the additional macro-morphological characters included in this study, did not locate any significant synapomorphies that can unite or be shared by any of the studied taxa, to support an alien classification of the family apart from the traditional one. The few shared characters by some taxa that were discussed earlier, can be considered as merely simplesiomorphic ones (from a cladistic point of view) and cannot be used to deduce relationships between distantly related taxa. However, the additional macromorphological characters helped in supporting the relatively isolated nature of Rhodotypos (Rosoideae). This view was held earlier by Focke (1894). The study also added some additional evidence for the heterogenetly of some genera as Spiraea.

It is worth mentioning that the micromorphology of the seed coat or achene characters showed a marked degree of similarity and/or overlap among the studied taxa. Barthlott (1981 and 1990), stated that the micromorphological features of seed epidermal cells are highly significant at the subgeneric to the subfamilial level. The results seem to support strongly the monophyletic nature of the *Rosaceae*, a fact that was shown by Judd *et al.* (1999), who stated that morphological and molecular characters support the monophyly of the family. However, in the present study, the four traditional subfamilies of the *Rosaceae* viz.;

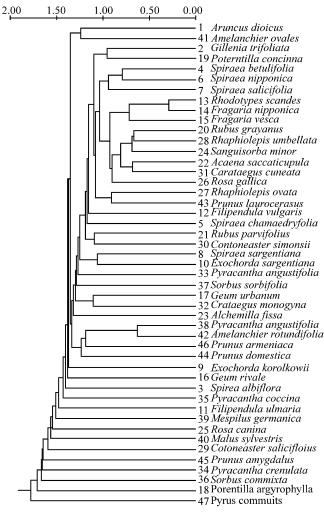


Fig. 6: UPGMA-phenogram based on 97 micromorphological characters of seed or achene coat illustrated the average taxonomic distance (dissimilarity) between the studied species of the *Rosaceae*

Maloideae, Prunoideae, Rosoideae and Spiraeoideae, will be taken into consideration, while discussing the results. This is because these traditional subfamilies were accepted by many authors (Robertson, 1974 a,b,c; Cronquist, 1981, Mabberley, 1997; Reveal, 1998 and Judd et al., 1999) merely because this classification is relatively simple and practical to a great extent (Fig. 6). However, only the most remarkable relationships within the four subfamilies will be considered here.

Subfamily *Maloideae*: The *Maloideae* appears to be a monophyletic group within the *Rosaceae*, united together by distinct synapomorphies as the pome type of fruit and

the basic chromosome number of the 17. In the present study, some taxa of the *Maloideae* had relations with several taxa in the other three subfamilies. These will be presented in the following:

Amelanchier ovales and Aruncus dioicus (Spiraeoideae) were both clustered at the dissimilarity level of 1.24 mainly due to their possessing monomorphic reticulate pattern of seed coat epidermal cells with raised and thick anticlincal walls (Fig. 1and2). On the other hand, Sanguisorba minor was separated with Rhaphiolepis umbellata and Rubus grayanus at the dissimilarity level of 0.68 due to their possessing reticulate, polymorphic irregularly shaped overall pattern of seed coat epidermal cells. They are also characterized by very thick, wavy, highly raised and highly striated anticlinal walls. But the other taxon clustered at the dissimilarity level of 0.65 due to their possessing a reticulate to faviulariate overall epidermal pattern, with irregularly shaped polymorphic epidermal cells and very thick, wavy, highly raised and highly striated anticlinal walls. Also, a relation was recorded between the following:

Cotoneaster simonsii and Rubus parvifolius (Rosoideae) both taxa are clustered around the 1.09 dissimilarity level (Fig. 7). Crataegus monogyna and Geum urbanum (Rosoideae) both taxa are clustered at (1.09) due to their possession of epidermal cells with slightly wavy and highly raised and highly striated anticlinal walls (Fig. 3).

Evans and Dickinson (1999), reported that several authors have suggested that the origin of the *Maloideae* was within the *Spiraoideae*. In their opinion, the fleshy pome fruit was derived from the expansion of the hypanthium (floral cup). They also stated that the incorporation of the ovaries by the enlarged hypanthium resulted in the inferior ovaries present in the majority of *Maloideae* genera. Data from seed morphology in the present study, give some support to the previous hypothesis about the *Maloideae* origin as many studied taxa had relationships with members of the *Spiraeoideae*.

Regarding the relationships with members of some *Maloideae* taxa with members of the *Rosoideae*, Heywood (1993), stated that the *Maloideae* origin can be within members of the *Rosoideae* with basic chromosome numbers of 8 and 9, i.e. any of the other three subfamilies and not within the *Spiraeoideae* in particular, as mentioned above. The present study also, showed that some taxa of the *Maloideae* had relationships with members of the *Prunoideae* regarding their seed coat morphology. The most remarkable were:

Rhaphiolepis ovata and Prunus laurocerasus at the dissimilarity level of 0.91 due to their possessing a rough globose seed with a reticulate overall seed coat pattern.

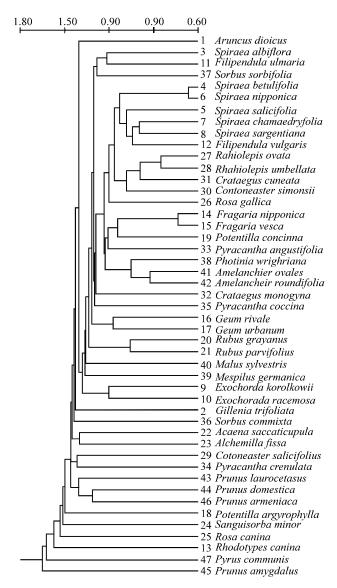


Fig. 7: UPGMA-phenogram based on 176 macro and micromorphological characters illustrated the average taxonomic distance (dissimilarity) between the studied species of the *Rosaceae*

Epidermal cells possess wavy and striated anticlinal walls. *Amelanchier rotundifolia* and *Photinia wrightiana* are clustered with *Prunus armeniaca* at the dissimilarity level of 1.18 mainly due to their seed and/or achene possessing convex periclinal epidermal cells.

These results may again give some support to an alternate hypothesis about the *Maloideae* origin stated by Stebbins (1950 and 1974), that the subfamily originated from the hybrid polyploids involving primitive representatives or ancestors of the subfamilies *Spiraeoideae* and *Prunoideae*.

Subfamily *Prunoideae*: Regarding the *Prunoideae*, *Prunus laurocerasus* and *P. amygdalus* are widely separated from the remaining two species of *Prunus* i.e. *P. armeniaca* and *P. domestica*. *Prunus laurocerasus* clustered with *Rhaphiolepis ovata* (*Maloideae*) at dissimilarity level (0.91), while *Prunus amygdalus* had relationships with most of the investigated taxa.

Prunus armeniaca clustered with Photiniawrightiana and Amelanchier (Maloideae) at the dissimilarity level of 1.18. The relation of Prunus to the Maloideae were discussed formerly. Concerning the generic delimitation, the taxonomy of Prunus has been contraversial (Lee and Wen, 2001). The most widely adopted classification is that of Rehder (1940), who divided it into five sub-genera. Viz.: Cerasus. Amygdalus, Padus. Laurocerasus, Prunus and Hutchinson (1964),recognized three genera (Laurocerasus, Padus and Prunus sensu stricto) within Prunus sensu lato. Others recognized six to ten genera within the generic concept of Prunus sensu lato (Yu et al., 1986 and Takhtajan, 1997). In the present study, the investigated species of Prunus falls according to Rehder's classification as follows: Sub-genus Laurocerasus (P. laurocerasus), Sub-genus Amygdalus (P. amygdalus) and Sub-genus Prunus (P. armeniaca and P. domestica). In all these classifications, P. laurocerasus is placed in a separate sub-genus (Laurocerasus).

A phylogenetic analysis of *Prunus* on molecular basis (its sequences of nuclear ribosomal DNA) recognizes two major groups in the genus: (a) The *Amygdalus - Prunus* group, (b) The *Cerasus - laurocerasus* and © *Padus* group (Lee and Wen, 2001). The present study is in accordance with the phylogenetic analysis presented by Lee and Wen (2001) in regarding *Prunus laurocerasus*, which was separated from the remaining studied taxa of *Prunus*. However, it does not agree with the splitting of *Prunus amygdalus* from *P. armeniaca* and *P. domestica*.

The relationship between *P. laurocerasus* and *Rhaphiolepis ovata* is supported by some vegetative macromorphological features, as both taxa are evergreen with glossy leaves (contrary to most *Prunus* species which are generally deciduous), both taxa have similar growth patterns as both possess dormant buds on the main stem and branches. These buds often produce recurrent flushes of epicormic shoots (an angiosperm phenomena described by Koslowski and Pallardy, 1997).

Subfamily *Rosoideae*: The relations of certain *Rosoideae* with members of the other three subfamilies were discussed formerly. However, the following taxa were shown to have relations with most of the studied taxa including the former three subfamilies:

Potentilla argyrophylla, Rosa canina, Filipendula ulmaria, Geum rivale, Alchemilla fissa and Filipendula vulgaris.

Mourad and Al-Nowaihi (2001), reported that *Geum rivale* and *Filipendula vulgaris* share a similar type of ramification from the ventral funicular vascular strand of the achene, while another type of ramification from the dorsal strand was shared between *Filipendula ulmaria* and *Fragaria nipponica*.

Their observations along with the results of the present study may suggest that the *Rosoideae* as currently circumscribed may prove to be a paraphyletic group (Eriksson and Donoghue, 1995). Moreover, Morgan *et al.* (1994), showed that the *Rosoideae* was polyphyletic as traditionally delimited, but when some aberrant groups were removed, the rest showed good support as being monophyletic. On the other hand, a close relationship was recorded between *Rhodotypos scandens* and *Fragaria nipponica*, both taxa clustered at a dissimilarity level of 0.30 due to their possessing a globose to pear-shaped achene and/or seed, with an epidermal overall pattern reticulate to favulariate, with wavy, very thick, highly raised and striated anticlinial walls, periclinial walls concave and highly striated (Fig. 3).

The widely separated taxa of *Potentilla* along the dendrogram, supports a study by Eriksson *et al.* (2001), that stated that the *Potentilla* as currently delimited may be grossly paraphyletic and can be treated as a mere assemblage of closely related taxa, with affinities to other taxa in the *Rosoideae*.

Subfamily *Spiraeoideae*: The most remarkable observation was that some investigated taxa had relationships with both the *Maloideae* and the *Rosoideae* in particular, while the two *Spiraea* species (*S. chamaedryfolia* and *S. albiflora*) had relationships with most of the studied taxa, the same was for *Exochorda korolkowii*. The relationship of the *Spiraeoideae* taxa with some members of the *Maloideae* were discussed formerly. However, the following additional relationships were recorded:

Spiraea sargentiana with Exochorda racemosa at the dissimilarity level of 1.06 due to their possessing overall achene or seed coat pattern reticulate to colleculate. Epidermal cells that are irregularly shaped with the anticlinal walls slightly wavy, very thick, highly raised and smooth. The periclinal walls that are concave and smooth.

Gillenia trifoliata with Potentilla concinna (Rosoideae) at 0.97 mainly due to their possessing an achene or seed that is longitudinal to pear-shaped, with a rough texture. The epidermal cells are rounded with highly raised anticlinal walls (Fig. 4).

The remaining taxa of *Spiraea* (S. betulifolia, S. nipponica and S. salicifolia) were closely related, the former two splitted at 0.80, while the latter clustered with the former two species of *Spiraea* at 0.94 due to the presence of pointed hairs at some parts of the seed and the irregularly shaped polymorphic epidermal cells.

The wide distribution of the *Spiraeoideae* across the constructed dendrograms (from seed morphology data) shows that this subfamily may be a polyphyletic assemblage as suggested by Morgan *et al.* (1994), Evans (1999), Evans and Dickinson, 1999 and 2001) and Judd *et al.* (1999).

Finally and after reviewing the relationships between the studied taxa, as revealed by the addition of 79 macromorphological and 97 morphological criteria, the following remarks can be drawn:

The constructed phenogram did not fit with the traditional suprageneric classification of the family as presented by Judd et al. (1999) and others. The studied taxa were widely separated and distributed all over the phenograms. Moreover, the relationships between the studied taxa showed some conflict and/or overlap between those based on seed micromorphological aspects. These results may appear frustrating for the first time, but as regarding an old, large and diverse family as the Rosaceae, these results agree with the previous studies on the family using other criteria as that of Morgan et al. (1994), Campell et al. (1995), Eriksson, et al. (1998), Evans (1999) and Eriksson et al. (2001). These authors stated that the taxa of the Rosaceae showed some conflict and were widely distributed on dendrograms based on molecular data from the chloroplast gene ndhF and similar molecular data. Moreover, non-molecular data as petal and stamen micromorphology showed a similar conflict with molecular ones (Evans and Dickinson, 1999 and 2001).

The two data sets were both not in accordance with the macromorphological criteria that separated the family into its subfamilies and taxa. (Evans, 1999). However, Mishler (2000) cautioned that observing a particular data set exhibiting serious conflict with another is not a sufficient reason to reject combining them. In their opinion, there must also be additional evidence, outside the phylogenetic analysis of lineage sorting and that all the characters or data must be used and combined together to achieve more relation to the constructed dendrograms. They also stated that this view rests on the fact that organellar genomes may have different phylogenies than those associated with nuclear genomes and morphologies. The same view is also stated by Smith and Sytsma (1990), Rieseberg and Soltis (1991) and Doyle (1992). And so after constructing a dendrogram according combination of data from vegetative macromorphology and seed macro and micromorphology data, the following relations were observed:

In the subfamily *Maloideae*; the wide variation between the two studied taxa of *Amelanchier* suggests that these two taxa may be separate species as was previously known and thus are not in agreement with Valentine and Chater (1972) and GRIN (2002), who stated that *Amelanchier rotundifolia* Dum. Cours. is a synonym of *Amelanchier ovales* Medik.

A relation was recorded between Sorbus sorbifolia and both of Spiraea albiflora (Spiraeiodeae) and Filipendula ulmaria (Rosoideae). It is splitted from them at the dissimilarity level of 1.28. The other studied species of Sorbus (S. commixta) had relations with most of the studied taxa. This result agrees with the synopsis of Maloideae genera presented by Robertson et al. (1991). In their opinion, Sorbus sensu lato is considered polyphyletic and the species have been reasigned to five genera (Sorbus, s. str., Cormus, Chamaesphilus, Aria and Torminalis).

The two studied species of *Rhaphiolepis* (*R. ovata* and *R. umbellata*) were split from each other at 0.85. however, they had relations with *Crataegus cuneata* and *Cotoneaster simonsii*. *Rhaphiolepis* clustered with the former taxon at 0.99 and with the latter at 1.08 (Fig. 5).

The whole group of *Rhaphiolepis* and both *Crataegus cuneata* and *Cotoneaster simonsii* had relations with five of the studied taxa of *Spiraea* (*Spiraoideae*) at the dissimilarity level of 1.16. This result may support the view of Evans (1999), Evans and Dickinson (1999) and Evans and Dickinson (2001), who stated that results from *ndhF*, non-molecular and *rbcl* analysis place members of the *Spiraoideae* as sister taxa to an enlarged *Maloideae* clade.

In the present study, the two investigated species of Crataegus were separated on the phenogram Crataegus cuneata had relations with Cotoneaster simonsii at 1.08 and with Rhaphiolepis umbellata and R. ovata at 0.99, while the other studied taxon (Crataegus monogyna) had relations with many taxa in the other three subfamilies. This result may agree with previous studies on this genus (Dickinson 1986; Dickinson and Phipps, 1986; Campell and Dickinson, 1990; Evans and Dickinson, 1996; Dickinson et al., 1996; Dickinson and Love, 1997 and several others). These authors reported a wide variation in the phenotypes of Crataegus and reported that its taxonomic complexity is bound up with the occurrence of gametophytic apomixis and polyploidy.

The phenogram or dendrogram based on the combination of all characters studied, again showed that the two studied species of *Cotoneaster* were widely distributed along the dendrogram. The first species

(Cotoneaster smonsii) had relations with Cratageus cuneata and Rhaphiolepis species at 1.08. The relation of Cotoneaster with Crataegus is in agreement with Mabberley (1997), who reported that the two genera Crataegus and Cotoneaster are closely related.

The other studied species of *Cotoneaster* (*C. salicifolus*) had relations with *Pyracantha coccinea* at 1.42. This result can give more support to the view of Rohrer *et al.* (1991), that *Cotoneaster* and *Pyracantha* are part of a monophyletic group branching from the base of a cladogram based on fruit anatomical structure.

In the two investigated species of *Pyracantha*, the first one (*P. angustifolia*) had relations with both *Fragaria* and *Potentilla concinna* (*Rosoideae*) at 1.20 average taxonomic distance. While the recorded species (*P. coccinea*) had relations with *Cotoneaster salicifolius* (as mentioned before) (Fig. 7).

The studied species of *Photinia* (*P. wrightiana*) had relations with the two species of *Amelanchier* at 1.04 level.

Malus sylvestris was shown to have relations with most taxa from the four subfamilies. The same for Mespilus germanica. However, the present study does not support a close relationship between Mespilus and Crataegus as presented by Dickinson et al. (2001). These authors stated that the data based on rDNA sequence variation showed that Mespilus and Crataegus along with Amelanchier form a monophyletic clade along with other genera as Peraphyllum and Malacomeles. In their opinion, Mespilus can be nested within Crataegus.

Pyrus communis was split from the rest of the studied taxa (Prunus amygdalus excluded) at 1.63. The taxonomy of Pyrus has been controversial, De-Candolle (1825) included Malus, Photinia and Sorbus in Pyrus. Decaisne (1874) and Koehne (1890) treated Pyrus in a more restricted sense and splitted off Photinia, Malus and Sorbus. A comprehensive review of taxonomic treatments applied to these genera was provided by Robertson et al. (1991).

In the subfamily *Prunoideae*; *Prunus armeniaca* is grouped with *P. domestica* at the level 1.32. This result agrees with the supraspecific classification of the genus (Rehder, 1940 and Lee and Wen, 2001) as both taxa are in sub-genus *Prunus*. However, the grouping of *P. laurocerasus* with *P. armeniaca* and *P. domestica* does not fit to the previous supraspecific classification as *P. laurocerasus* belong to the somewhat distant subgenus *P. laurocerasus* (Rehder, 1940) or *Cerasus laurocerasus* group (Lee and Wen, 2001). The splitting of *Prunus amygdalus* from the rest of the studied taxa seems somewhat aberrant and inconsistent with the previous studies. Further research is still needed on this taxon in particular.

In the subfamily Rosoideae; the constructed phenogram according to the combination of all characters studied, again stressed on the relations between Filipendula and Spiraea. Filipendula ulmaria had relations with Spiraea albiflora (Spiraeoideae) at 1.22 and Filipendula vulgaris had relations with Spiraea salicifolia and S. sargentiana at 1.05 level.

The two species of *Rosa* (*R. gallica* and *R. canina*) were widely distributed across the phenogram. This result may seem somewhat aberrant as the monophyly of roses is now fully documented (Evans and Dickinson, 2001). More studies are still needed on this genus utilizing other criteria

A close relationship was recorded between the two studied species of *Fragaria* as was expected.

The two studied species of *Potentilla* were widely distributed across the dendrogram. The first species *P. coccinea* had relationships with two species of *Fragaria* at 1.14. The record species (*P. argyrophylla*) had relations with most of the studied taxa. It is worth mentioning that *Potentilla*, the largest genus in the *Rosoideae* is variable in morphological feature (Mabberley, 1997). The previous result give more support to Eriksson *et al.* (2001) who stated that *Potentilla* may be grossly paraphyletic. The same result was also presented by Eriksson and Donoghue (1995) and Eriksson *et al.* (1998).

The two studied taxa of *Geum* clustered together at 1.16. They also possessed some relationships with some of the *Maloideae* and *Spiraeoideae* at the level 1.32 (Fig. 7). The two studied taxa of *Rubus* clustered together at 1.05. *Acaena saccaticupula* and *Alchemilla fissa* clustered together at 1.40. The two genera *Sanguisorba* and *Rhodotypos* had relations with most of the studied taxa.

In the subfamily *Spiraeoideae*; *Aruncus dioicus* had relationships with members of the other subfamilies. *Spiraea albiflora* had relationships with *Filipendula ulmaria* at the level 1.22 while, a close relationship was recorded between *S. betulifolia* and *S. nipponica* at the level 0.68. *Spiraea salicifolia* clustered with *S. sargentiana* at the level 1.00.

The two taxa split from *S. chamaedryfolia* at 1.08. The latter five taxa of *Spiraea* had relationships with *Filipendula vulgaris* at 1.13. The status of *Spiraea* as a monophyletic taxon was discussed formerly. The relations of the taxa of *Spiraea* to various members of the *Maloideae* is worth mentioning: *Spiraea albiflora* with *Sorbus sorbifolia*. The rest of the *Spiraea* with *Rhaphiolepis* sp., *Crataegus cuneata* and *Cotoneaster simonsii*. This may give extra widence to the hypothesis that *Maloideae* may have originated within members of the *Spiraeoideae* (Evans, 1999).

The two species of *Exochorda* (*E. korolkowii* and *E. racemose*) clustered at 1.19. The *Exochorda* also had relationships with most of the studied taxa at 1.37. *Gillenia trifoliata* was shown to have relations with most of the studied taxa.

Finally, this study was only a preliminary investigation on the available taxa of the *Rosaceae* representing the accepted four subfamilies, helping only in throwing some light on the relationships between the taxa studied. It would be by no means be considered as a final output for a diverse and old family as the *Rosaceae*. Also, the study showed that the present suprageneric classification of the family and also the relations between the *Rosaceae* taxa may be in need of revision and so gives support to previous studies urging on the same issue (Morgan *et al.*, 1994; Evans, 1999; Judd *et al.*, 1999; Eriksson *et al.*, 2001 and Evans and Dickinson, 2001).

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