

Insight on the Taxonomy of Caryophyllaceae Juss. Based on Pollen Grain Diversity

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Abstract

This work has been carried out on the pollen grains of 63 species representing the major taxonomic division of the family using both the light and scanning electron microscopes.

Objective: The aim of this study is to elucidate the pollen grain diversity within the Caryophyllaceae taxa, and to limit these variations to coordinate with the taxonomic division of the family.

Material and Methods: The data obtained from the pollen examinations by both light and SEM subjected to PAST program and resulted in grouping the taxa under three main groups and nine subgroups. These grouping did not coordinate with the classical morphological division of the family.

Results: This work demonstrates that aperture type, number of pores and colpi, the pollen surface ornamentation beside the density of microechini have important systematic significance in the discrimination of the taxa.

Conclusion: The results support the taxonomic classification of the family into four subfamilies as mentioned by previous works by the separation of the Paronychieae in its own subfamily Paronychioideae, and the three tribes; Polycarpeae, Corrigioleae and Sperguleae; in the forth subfamily Polycarpoideae. The two genera *Gypsophylla* and *Minuartia* need reclassification. The pollen morphological characters showed differences within the three main subfamilies and within the genera as well as the species under each subfamily and can be used in the identification of the studied taxa. The pollen diversity within the studied Caryophyllaceae supports the molecular phylogenetic works in suggesting that the traditionally recognized subfamilies are non-monophyletic and reclassification of the genera and tribes must be considered and in agreement with the taxonomic division of the family into three subfamilies with many tribes.

Keywords: Alsinoideae, Caryophyllaceae, Caryophylloideae, Paronychioideae, Pollen grains.

1. INTRODUCTION

Caryophyllaceae considered from the large families of angiosperms. It includes about 2200 species and 86 genera, Members of the family are mainly centered in the Mediterranean area with great distribution in Northern Hemisphere and exhibits great diversity in the habitat and growth form [1]. Many members of the family are popular ornamentals commonly planted in gardens for their beautiful flowers as *Dianthus* L. and *Gypsophila* L. or for their medicinal resources as *Cerastium* L., *Silene* L., and *Stellaria* L. [2]. The family has traditionally divided into three subfamilies, Caryophylloideae, Alsinoideae and Paronychioideae [3-5]. More recently, new classification systems have been proposed based on molecular and morphological evidence in Caryophyllaceae, and eleven tribes were recognized [6-8]. Rabeler and Hartman [5] had another opinion about the taxonomy and classification of the genera. He divided the family into four subfamilies: Alsinoideae, Caryophylloideae, Paronychioideae, and Polycarpoideae. Generally all members of the Caryophyllaceae have common morphological features as swollen nodes, with simple, sessile, opposite leaves, solitary flowers or flowers aggregated in dichasial cymes. Flowers are glorious actinomorphic, pentamerous with distinct beautiful clawed petals. In spite of the common vegetative and floral characters, the relationships and grouping of the genera have been under argument and dispute. In the traditional classification of the family the presence or absence of the stipules was the base in distinguishing subfamily

Paronychioideae from the other two subfamilies, Alsinoideae and Caryophylloideae (table 1). Many taxonomical and palynological works have been done to clarify the relation between the species under the same genus. From these works are those [9-17]. In fact the carnation family has great diversity in its morphological characters and life forms. The family has plants which are annual, biennial or perennial herbs with swollen nodes and opposite or alternate leaves. The flowers are either solitary or aggregated in different kinds of inflorescences. Thus [6] announced that the relationships within the genera of the Caryophyllaceae have been difficult, because many of them are not well defined morphologically and are difficult to distinguish.

[1, 6, 18] Mentioned that the subscription of the three caryophyllaceae subfamilies is difficult due to the morphological homoplasy. [1] suggested that both the two subfamilies Alsinoideae and Caryophylloideae form a monophyletic group based on caryophyllad-type embryology, comparing with the solanad embryology in the third subfamily Paronychioideae. Accordingly [6, 7, 18] made molecular phylogenetic studies within the three traditional subfamilies disregarding their monophyly. [19] Found great conflicts between the Caryophyllaceae genera. She proposed new opinions about the relation between the genera and mentioned that genus *Polycarpon* is polyphyletic and that both *Spergula* and *Spergularia* are monophyletic. She suggested that Polycarpaea is polyphyletic and should be split into three larger and several smaller genera and members of *Paronychia* need to be transferred to *Herniaria* to maintain monophyly. Caryophyllaceae which is a large and cosmopolitan flowering plant family the systematics of many of its basal groups has been unclear till now, due to a lack of unambiguous morphology. Some members of Caryophyllaceae are used medicinally. Monitoring the trade in medicinal plants is complicated due to the absence of useful and precise identification characters.

Therefore, new taxonomic revisions of the whole family have been conducted according to molecular phylogenetic data [20-27] and even so the relation between members under the three subfamilies still unclear. Most of the pollen grain studies focused on the genus *Minuartia* and *Stellaria* beside other related species to explore the pollen morphological features in trials to understand their taxonomic importance within the family.

The family is worldwide distributed, and native to the Mediterranean area. In Egypt, the family is represented by 28 genera with 92 species [28]. The largest genera are *Silene* (27 sp.), *Dianthus* (6 sp.), *Minuartia* (6 sp.), *Herniaria* (5 Sp.) and *Paronychia* (5 sp.). The rest of the genera are represented by one to four species only. This work has carried out on 63 species representing the major taxonomical division of the family. The aim of this study is to elucidate the pollen grain diversity within the Caryophyllaceae taxa, and to which limit these variations coordinate with the morphological and molecular taxonomical opinions of the genera and subfamilies. Also the work aims to provide clear variations between the taxa under the three main subfamilies.

2. MATERIALS AND METHODS

This work includes 63 species belonging to 22 genera representing the different classical tribes under the three subfamilies (table 1). The species were examined as its own without regarding to its taxonomic division. They were arranged alphabetically to be treated without any bias to its division. Mature flowers of the studied species were taken from herbarium sheets allocated in Alexandria University herbarium beside some fresh specimens collected from the Mediterranean coastal strip in Alexandria city, Egypt. The flowers were carefully opened under Stereo-microscope to collect the anthers in test tubes, smeared by glass rod and acetolyzed according to [29]. Qualitative and quantitative characters were taken according to 30 pollen grain examination from each taxa. The pollen grains were examined, measured and photographed using Zeiss light microscope with a pre-calibrated eye-piece micrometer. The mean and standard deviations were calculated for each measured character. The apocolpium index (Polar area index, AI) was calculated as the ratio of the mean distance between the apices of two colpi to its equatorial diameter. All measurements are in micrometer and stated as means and standard deviations.

Non acetolyzed pollen grains from the different taxa were sputtered onto copper stubs, coated with 30 nm gold using fine coat ion sputter JEOL JFC 1100E, examined and photographed at 30 KV using JEOL JSM-3500 scanning electron microscope (SEM) allocated in the Faculty of Science, Alexandria

University. The terminology used in this study was according to [30]. The studied taxa and their taxonomical position are summarized in table 1.

2.1. Data Analyses

Fifteen pollen morphological characters have been subjected for cluster analyses using PAST 3 programs V.3. Designed by Hammer (1999-2013)³¹ to estimate the relationship between the characters as well as between the studied taxa.

Table 1. Classification of the Caryophyllaceae according to Bittrich (1993) and the studied taxa.

Subfamily	Common Character	Tribe	Genera	Species studied		
Alsinoideae A. Braun	No stipules, petals free	Alsineae Lam. & DC. Syn. Pl. Fl. Gall.: 392. 1806	<i>Cerastium</i> L.	<i>C. dichotomum</i> L. <i>C. viscosum</i> L.		
			<i>Pseudostellaria</i> Pax	-----		
			<i>Stellaria</i> L.	<i>S. media</i> (L.) Vil. <i>S. pallida</i> (Dum.) Pire		
		Arenarieae Kitt., Taschenb. Fl. Deutschl., ed. 2, 2: 981. 1844	<i>Arenaria</i> L.	<i>A. deflexa</i> Decne <i>A. serpicifolia</i> L.		
			<i>Moehringia</i> L.	-----		
Caryophylloideae Arn.	No stipules, petals connate	Caryophylleae Lam. & DC., Syn. Pl. Fl. Gall.: 386. 1806	<i>Dianthus</i> L.	<i>D. cyri</i> Fish & Mey. <i>D. strictus</i> Banks et Sol.		
			<i>Gypsophilla</i> L.	<i>G. capillaris</i> (Forssk.) C.Chr. <i>G. viscosa</i> Murr.		
			<i>Vaccaria</i> Medicus	<i>V. oxyodonta</i> Boiss. <i>V. pyramidata</i> Medicus		
		Scleranthae DC.	<i>Scleranthus</i> L.	-----		
		Geocarpeae	<i>Geocarpon</i>	-----		
		Pycnophylleae	<i>Lychnis</i>	-----		
		Habrosieae	<i>Habrosia</i>	-----		
		Sagineae J.Presl in Nowočeská Bibl. [Wšobecný Rostl.] 7: 1609, 1621. 1846	<i>Bufonia</i> L.	<i>B. multiceps</i> Decne		
			<i>Minuartia</i> L.	<i>M. geniculata</i> (Poiret)Thell <i>M. meyeri</i> Bornm. <i>M. picta</i> (Sbth.& Sm.)Bornm. <i>M. procumbens</i> Grarbn. <i>M. tenuifolia</i> (L.)Hiern		
				<i>Sagina</i> L.	-----	
				Sileneae DC., Prodr. 1: 351. 1824	<i>Silene</i> L.	<i>S. aegyptiaca</i> (L.) L.f. <i>S. apetala</i> Wil. <i>S. arabica</i> Boiss. <i>S. behen</i> L. <i>S. biappendiculata</i>

				Rohrb. <i>S. colorata</i> poiret <i>S. conoidea</i> L. <i>S. gallica</i> L. <i>S. linearis</i> Decne <i>S. longipetala</i> Vent. <i>S. nocturna</i> L. <i>S. oliveriana</i> Oth. <i>S. palestina</i> Bois. <i>S. pseudotocion</i> Desf. <i>S. rubella</i> L. <i>S. setacae</i> Viv. <i>S. succulenta</i> Forssk., <i>S. tridentata</i> Desf.	
Paronychioideae Vierh.,	Fleshy stipules, petals separate or united	Paronychieae Dumort., Fl. Belg.: 86. 1827	<i>Herniaria</i> Tourn. ex L.	<i>H. fontanesii</i> J. <i>H. glabra</i> L. <i>H. hirsuta</i> L. <i>H. nemistemon</i> J. Gay	
			<i>Gymnocarpos</i> Forssk.	<i>G. decandrum</i> Forssk.	
			<i>Paronychia</i> Mill.	<i>P. argentea</i> Lam. <i>P. nivea</i> DC.	
		Polycarpeae DC., Prodr. 3: 373. 1828	<i>Cometes</i> L.	<i>C. abyssinica</i> R. Br. <i>C. surattensis</i> L.	
			<i>Loeflingia</i> L.	<i>L. hispanica</i> L.	
			<i>Polycarpeae</i> Lam.	<i>P. corymbosa</i> (L.) Lam. <i>P. repens</i> (Forssk.)Asch & Schweinf. <i>P. spicata</i> Wight ex Arn.	
				<i>Polycarpon</i> Loefl.	<i>P. prostratum</i> (Forssk.) Pax <i>P. succulentum</i> (Del.)J.Gray <i>P. tetraphyllum</i> (L.) L.
					<i>Pteranthus</i> Forssk.
			<i>Sphaerocoma</i> T. Anders.		<i>S. hookeri</i> T. Anders.
			Corrigioleae Dumort., Fl. Belg.: 86. 1827	<i>Telephium</i> L.	<i>T. sphaerospermum</i> Boiss.
				<i>Holosteum</i> L.	<i>H. umbellatum</i> L.
			Sperguleae Dumort.	<i>Spergula</i> L.	<i>S. arvensis</i> L. <i>S. fallax</i> E.H.L.
				<i>Spergularia</i> (Pers.) J.Presl & C.Presl	<i>S. bocconii</i> (Sol.ex Scheele) Ash. et Graebn., <i>S. diandra</i> Boiss. <i>S. marina</i> (L.) Gresib. <i>S. media</i> (L.) C. Presl ex Griseb. <i>S. rubra</i> (L.) J. & C. Presl

3. RESULTS

All the investigated taxa were treated regardless their taxonomic category of the genera and examined unbiased to their rankings as shown in table 2. According to the groupings of the studied taxa the description of the pollen grains was based on the common characters of the species and divided mainly into three groups and nine subgroups.

3.1. Group A Includes 29 Taxa and Divided into Three Subgroups

3.1.1. Common features of group A

Taxa belonging to group A were apolar isodiametric with spheroidal pollen grains with different diameters with pantoporate apertures. The number of pores was varied from 6 to 22 pores/pollen differed within the subgroups. The pore diameter and interporal distances varied as well. Exine was thin, tectate echinate or tectate microechinate perforate ornamentation.

3.1.2. Subgroup A I

Seven taxa; *Arenaria defexa*, *A. serpifolia*, *Silene aegyptiaca*, *S. arabica*, *S. noctirna*, *S. oliveriana* & *Vaccaria oxyodonta*; were in this subgroup share the pollen features, as their sizes were moderate from 32.6 to 46.4 μm . The number of pores varied from 12 in *Arenaria serpifolia* and *Vaccaria oxyodonta* to 20 in *Silene aegyptiaca* and *S. arabica*. The pore diameter varied from 2.8 μm in *S. noctirna* to 6.0 μm in *S. aegyptiaca*. The interporal distance varied from 3.8 μm in both *S. aegyptiaca* and *S. arabica* to 8.2 μm in *Arenaria serpifolia*. The pores were densely covered by granules. Exine thickness from 2.2 μm to 2.8 μm with tectate echinate or tectate microechinate perforate ornamentation.

3.1.3. Subgroup A II

Twelve *Silene* species; *Silene biappendiculata*, *S. conoidea*, *S. colorata*, *S. gallica*, *S. linearis*, *S. longipetala*, *S. palestina*, *S. pseudotocion*, *S. rubella*, *S. setacea*, *S. succulent* & *S. tridentate*; were grouped together in this subgroup. These species has pollen diameter range from 38.8 μm to 48.2 μm . The number of pores varied from 12 in *S. longipetala*, 16 in *S. linearis*, 20 in *S. tridentate* to 22 in the rest of the species. The pore diameter is moderate as it ranges from 3.5 μm in *S. colorata* to 5.5 μm in *S. longipetala*. The interporal distance varied from 3.8 μm *S. biappendiculata* to 5.4 μm in *S. succulenta*. The pores are sparsely ornamented by rounded granules. The exine thickness varied from 2.3 μm in *S. biappendiculata* to 3.0 μm in *S. rubella*, *S. succulent* and *S. tridentata*, with tectate microechinate perforate tectum in all the subgroup species, except *S. palestina* the tectum is without perforation.

3.1.4. Subgroup A III

Ten taxa; *Cerastium dichotomum*, *C. viscosum*, *Minuartia meyeri*, *M. picta*, *M. tenuifolia*, *Silene apeta*, *S. behen*, *Stellaria media*, *S. pallida*, *Vaccaria pyramidata*; were clustered together sharing most of the pollen features. The pollen diameter varied from 30.8 μm in *Minuartia meyeri* to even 42.8 μm in *Silene behen*. The number of pores varied from 10 in *Minuartia meyeri*, 14 in *Cerastium dichotomum*, 16 in *C. viscosum* and reach to 20 in *S. behen*, while it was 12 in the rest of the species. The pore diameter is small or moderate in the subgroup taxa as it ranges from 2.5 μm in *Minuartia meyeri*, 2.8 μm in the two *Cerastium* species to 6.2 μm in *S. behen*. The interporal distance varied from 3.2 μm *Minuartia tenuifolia* to 6.8 μm in *M. meyeri*. The pores are sparsely ornamented by rounded granules in all of the species, except the two *Cerastium* species the pores ornamented by moderate rounded granules. The exine thickness varied from 3.0 μm in *Vaccaria pyramidata* to 3.8 μm in *S. behen*, with sparsely tectate echinate or tectate granulate tectum.

3.2. Group B Includes 12 Taxa and Divided into Two Subgroups

All the taxa belonging to group B have apolar, isodiametric with spheroidal pollen grains and pantoporate apertures. The pollens were small in diameters in subgroup I and moderate in subgroup II. The pores were few in the two subgroups from 6 to 12, their diameter and interporal distances were variable. Exine thin with tectate foveolate, echinate, granulates or microechinate perforate ornamentation.

3.2.1. Subgroup B I

Nine taxa; *Bufonia multiceps*, *Gymnocarpus decandrum*, *Gypsophilla viscosa*, *Herniaria fontanesii*, *H. glabra*, *H. hirsuta*, *H. hemistemon*, *Paronychia argentia* & *P. nivea*; were grouped together in this subgroup sharing small pollen grains diameters less than 20 μm in all of the species, except in *Gypsophilla viscosa* it was 25.2 μm . *Bufonia multiceps* the pollens were subprolate with small polar axis length (20.2 μm). The number of pores were few, from 6 to 10 pore/pollen. The pore diameter was small in most of the species; less than 3.0 μm in *Bufonia multiceps*, *Herniaria fontanesii*, *H. glabra*, *H. hirsuta*, *H. hemistemon* and *Paronychia nivea*; while it reached 6.8 μm in *Gymnocarpus decandrum*. The interporal distances varied from 2.8 μm in *Paronychia argentea* to 4.5 μm in both *Herniaria hirsuta* and *H. hemistemon*. The pores were covered by considerably dense rounded granules. Exine thickness from 2.2 μm to 3.2 μm with foveolate, tectate echinate, and tectate granulate or tectate microechinate perforate ornamentation.

3.2.2. Subgroup B II

Three taxa only; *Dianthus cyri*, *D. strictus* & *Holosteum umbellatum*; were found in this subgroup having moderate pollen diameters from 35.0 to 47.2 μm . The number of pores was few, from 8 to 12 pore/pollen. The pores were wide in the two *Dianthus* species, reach more than 6.0 μm , while they are moderate in *Holosteum umbellatum* (3.8 μm). The interporal distances were from 5.8 to 6.2 μm . The pores were covered by sparse rounded granules. Exine thickness varied from 2.5 μm to 3.2 μm with reticulate or tectate microechinate perforate ornamentation.

3.2.3. Group C includes 22 taxa and divided into four subgroups

Taxa belonging to group C were divided into four subgroups. In general the pollen grains in the four subgroups were isopolar, isodiametric with oblate, subprolate or spheroidal with tricolpate apertures. The pollens differed in size within the four subgroups. The colpi were either wide or narrow with different lengths, their membrane were either smooth or ornamented by rounded granules. The apocolpium distances and mesocolpium regions varied between the four subgroups. Exine thin or very thin with tectate echinate, tectate granulates or tectate microechinate perforate ornamentation.

3.2.4. Subgroup C I

Three taxa were found in this subgroup; *Cometes abyssinica*, *C. surattensis* & *Pteranthus dichotomous*; Having moderate sized subprolate pollen grains with polar axis from 27.9 to 36.2 μm and equatorial diameter from 27.2 to 35.8 μm . The colpi were wide or narrow, long reaches near the equators with apocolpium regions 0.13 or 0.14 and mesocolpim distances from 7.8 to 10.4 μm . The colpi membranes were densely or moderately granulate. Exine thickness was from 2.3 to 2.8 μm with tectate echinate or tectate microechinate perforate ornamentation.

3.2.5. Subgroup C II

Three taxa were found in this subgroup; *Gypsophilla capillaries*, *Loeflingia hispanica* & *Telephium sphaerospermum*; this subgroup have very small size pollen grains from 14.2 to 20.5 μm , with different shapes from the oblate to spheroidal or even subprolate. The colpi were narrow, shorter than the polar axis with apocolpium regions 0.28 or 0.30 and mesocolpim distances from 4.8 to 6.5 μm . The colpi membrane were densely granulate in *Gypsophilla capillaries* and smooth in the two species. Exine was thin from 1.5 to 2.5 μm with tectate echinate or tectate granulate ornamentation.

3.2.6. Subgroup C III

Nine taxa were found in this subgroup; *Minuartia geniculata*, *Minuartia procumbens*, *Polycarpeae corymbosa*, *P. repens*, *P. spicata*, *Polycarpon prostratum*, *P. succulentum*, *P. tetraphyllum* & *Sphaerocoma hookeri*; this subgroup have small sized, prolate pollen grains. The polar axis varied from 13.5 μm in *Polycarpon succulentum* to 23.2 μm in *Sphaerocoma hookeri*, while the equatorial diameters varied from 12.2 to 19.8 μm in the same two species. The pollen shape was prolate with tricolpate apertures. The colpi were narrow or wide, shorter than the polar axis with apocolpium regions from 0.11 in *Sphaerocoma hookeri* to 0.26 in *Polycarpeae repens* and mesocolpium distances from 3.5 μm in *Polycarpon tetraphyllum* to 6.6 μm in *Sphaerocoma hookeri*. The colpi membrane was densely granulate in the two *Minuartia* species and sparsely or moderately granulate in the rest

species. Exines thin from 1.5 to 2.5 µm with tectate echinate, tectate granulate or tectate microechinate perforate ornamentation.

3.2.7. Subgroup C VI

Seven taxa were found in this subgroup; *Spergula arvensis*, *S. fallax*, *Spergularia bocconii*, *S. diandra*, *S. marina*, *S. media* & *S. rubra*; the pollen grains in this subgroup were small sized, prolate or subprolate. The polar axis varied from 17.5 µm in *Spergularia media* to 29.8 µm in *Spergula arvensis*, while the equatorial diameters varied from 16.0 to 28.9 µm in the same two species. The apertures were colpate, tetra, and hexa or penta colpate. The colpi were narrow or wide, considerably shorter than the polar axis with apocolpium regions from 0.16 in *Spergularia bocconii* to 0.29 in *Spergularia marina*, *S. media* and *S. rubra*. The mesocolpium distances varied from 4.6 µm in *Spergularia media* to 7.8 µm in *Spergula arvensis*. The colpi membrane was smooth or sparsely except in *Spergularia marina* it was densely granulated. Exine thickness was thin from 1.5 to 2.5 µm with tectate microechinate perforate ornamentation.

Table 2. Pollen morphological characters in the studied species

Key to table 2: AI= Apocolpial index, C/D = Interporal distance / pore diameter, D=Pore diameter, DG.=Density of granules or echinae, D.G./P=Density of granules over pore, += sparse, ++ =moderate, +++ =dense (crowded), D.S= Deeply sunken, Ec= Echinata, FG=Faintly granulate, Fo=Foveolate, G=Granulate, Gr=Group, G1= Diameter < 20 µm, G2= Diameter from 20-30 µm, G3= Diameter > 30 µm Gr=Granulate, HC=Hexacolpate, HpC=Heptacolpate, IP(C)= Interporal distance, L= Length, Med=Mesocolpium distance, MEP=Microechinate perforate, MG=Moderately granulate, Mm=Membrane, N=Narrow, No=Number, Ob=Oblate, Or=Ornamentation, PP= Pantoporate, Pr=Prolate; Re=Reticulate, S= Sunken, SG=Subgroup, SG= Sparsely granulate, Sm=Smooth, Sp= Spheroidal, Spr=Subprolate, Sup=Superficial, TC=Tricolpate, TeC=Tetracolpate, TEc= Tectate echinate, Th=Thickness, TGr=Tectate granulate, TMEP=Tectate microechinate perforate, Ty=Type, W=Wide, Wd=Width.

No	Character → Species↓	Mean±SD			Aperture										Exine			G	S	G
		PAL	EDL	Sha pe	Typ	Pore			Colpus							Th	Or			
					e	No	D	IP	C/ D	DG/ P	W d	L	M m	AI	Med					
1	<i>Arenaria defexa</i>	32.6±2.04	32.6±2.04	Sp	PP	18±2.94	3.8±2.05	7.5±2.92	1.97	+++	--	--	-	--	--	2.2±1.02	TMEP	+++	A	I
2	<i>A. serpiifolia</i>	36.2±1.89	36.2±1.89	Sp	PP	12±3.25	4.2±2.07	8.2±3.22	1.95	++	--	--	-	--	--	2.5±2.32	TMEP	++	A	I
3	<i>Bufonia multiceps</i>	20.2±2.87	20.0±3.22	Sp	PP	6±2.11	2.8±1.19	3.8±1.22	1.36	+++	--	--	-	--	--	3.2±1.09	TMEP	+++	B	I
4	<i>Cerastium dichotomum</i>	32.8±3.02	32.2±3.15	Sp	PP	14±1.04	2.8±2.09	5.4±2.85	1.93	++	--	--	-	--	--	3.4±2.05	TMEP	+	A	III
5	<i>C. viscosum</i>	33.2±1.02	33.2±1.23	Sp	PP	16±1.08	2.8±2.59	4.8±2.18	1.71	++	--	--	-	--	--	3.6±1.98	TEc	+++	A	III
6	<i>Cometes abyssinica</i>	31.5±2.07	30.9±3.01	Spr	TC	--	--	--	--	--	W	26.8±2.18	D	0.14	8.2±1.46	2.3±1.22	TEc	++	C	I
7	<i>C. surattensis</i>	36.2±2.13	35.8±3.42	Spr	TC	--	--	--	--	--	W	27.2±3.81	D	0.13	10.4±2.05	2.6±1.86	TMEP	++	C	I
8	<i>Dianthus cyri</i>	41.2±2.12	41.2±2.12	Sp	PP	8±1.22	6.2±1.22	5.8±2.42	0.94	+	--	--	-	--	--	2.5±1.02	TMEP	+++	B	II
9	<i>D. strictus</i>	47.8±1.76	47.5±2.08	Sp	PP	8±1.06	6.5±1.22	6.2±1.26	0.95	+	--	--	-	--	--	3.2±1.96	Re	-	B	II
10	<i>Gymnocarpus decandrum</i>	18.4±1.02	18.2±1.22	Sp	PP	6±1.02	6.8±1.82	4.2±1.12	0.68	+++	--	--	-	--	--	3.2±1.08	Fo	-	B	I
11	<i>Gypsophilla capillaris</i>	14.2±2.02	18.2±2.02	Ob	TC	--	--	--	--	--	N	6.4±0.43	D	0.30	4.8±1.26	1.5±0.22	TEc	++	C	II
12	<i>G. viscosa</i>	25.2±1.88	25.2±1.88	Sp	PP	8±1.22	5.5±0.85	3.8±1.02	0.69	++	--	--	-	--	--	2.8±0.36	TEr	+++	B	I
13	<i>Herniaria fontanesii</i>	13.2±1.36	13.2±1.36	Sp	PP	8±0.88	2.2±0.34	3.5±0.62	1.59	++	--	--	-	--	--	2.3±0.22	TEc	++	B	I
14	<i>H. glabra</i>	13.6±1.28	13.6±1.28	Sp	PP	8±1.04	2.2±0.66	4.2±0.66	1.91	++	--	--	-	--	--	2.3±0.27	TGr	+	B	I
15	<i>H. hirsuta</i>	14.2±0.33	14.0±0.62	Sp	PP	8±0.22	2.5±1.02	4.5±0.88	1.80	++	--	--	-	--	--	2.2±0.25	TGr	+	B	I
16	<i>H.</i>	13.8	13.8	Sp	PP	8±0.2	2.3±	4.5±1.	1.9	++	--	--	-	--	--	2.5±0.	TGr	+	B	I

Insight on the Taxonomy of Caryophyllaceae Juss. Based on Pollen Grain Diversity

6	<i>hemistemon</i>	±0.83	±0.83			4	1.12	08	6							18				
1	<i>Holosteum</i>	35.2	35.2	Sp	PP	12±	3.8±0.	5.8±2.	1.5	++	--	--	--	--	--	3.2±1.	Re	+	B	II
7	<i>umbellatum</i>	±3.83	±3.83			2.04	48	05	3							88				
1	<i>Loeflingia</i>	16.8±1.	16.2±1.	Spr	TC	--	--	--	--	--	N	11.8±0.	Sm	0.3	4.8±1.2	1.8±0.	TG	+	C	II
8	<i>hispanica</i>	22	82									88		0	2	62				
1	<i>Minuartia</i>	18.5	16.2	Pr	TC	--	--	--	--	--	W	13.6	D	0.2	4.8±2.0	1.8±0.	TEc	++	C	III
9	<i>geniculata</i>	±1.22	±1.06									±1.02	G	6	5	33				
2	<i>M. meyeri</i>	30.8	32.8	Ob	PP	10±0.	2.5±0.	6.8±0.	2.2	+	--	--	--	--	--	3.2±0.	TGr	+	A	III
0		±2.36	±2.36			44	53	47	7							38				
2	<i>M. picta</i>	31.5	31.2	Sp	PP	12±0.	4.5±0.	5.0±0.	1.1	+	--	--	--	--	--	3.2±0.	TGr	+	A	III
1		±2.58	±2.58			75	53	33	1							63				
2	<i>M. procumbens</i>	21.5±1.	17.8±1.	Pr	TC	--	--	--	--	--	N	16.2±1.	D	0.2	5.1±2.0	2.0±0.	TGr	+	C	III
2		88	12									22	G	1	2	33				
2	<i>M. tenuifolia</i>	30.8±2.	30.8±2.	Sp	PP	12±0.	3.8±0.	3.2±0.	0.8	+	--	--	--	--	--	3.2±0.	TGr	+	A	III
3		44	44			46	26	24	4							79				
2	<i>Paronychia</i>	13.5±1.	13.4±1.	Sp	PP	6±2.0	3.2±0.	2.8±0.	0.8	+++	--	--	--	--	--	2.2±0.	TGr	+	B	I
4	<i>argentea</i>	03	03			0	22	22	9							24				
2	<i>P. nivea</i>	15.2±1.	15.2±1.	Sp	PP	10±2.	2.2±0.	3.0±0.	1.3	+++	--	--	--	--	--	2.2±0.	TEc	++	B	I
5		34	22			00	16	25	6							22				
2	<i>Polycarpeae</i>	21.4±0.	16.3±2.	Pr	TC	--	--	--	--	--	W	18.8±1.	M	0.2	5.1±0.6	1.7±0.	TEc	++	C	III
6	<i>corymbosa</i>	32	40									22	G	5	6	24				
2	<i>P. repens</i>	18.2±1.	14.0±1.	Pr	TC	--	--	--	--	--	W	14.2±2.	M	0.2	3.8±1.2	2.0±0.	TEc	++	C	III
7		02	22									04	G	6	5	52				
2	<i>P. spicata</i>	20.5±1.	15.5±0.	Pr	TC	--	--	--	--	--	W	18.2±1.	M	0.2	4.8±1.2	2.4±0.	TEc	++	C	III
8		00	72									00	G	5	2	15				
2	<i>Polycarpon</i>	17.6±1.	15.2±0.	Pr	TC	--	--	--	--	--	W	16.0±0.	M	0.1	4.6±1.6	2.0±0.	TEc	++	C	III
9	<i>prostratum</i>	74	44									62	G	6	6	15				
3	<i>P. succulentum</i>	13.5±1.	12.2±1.	Pr	TC	--	--	--	--	--	W	11.8±1.	SG	0.2	3.8±0.5	1.8±0.	TEc	++	C	III
0		48	12									22		3	5	03				
3	<i>P. tetraphyllum</i>	16.5±1.	12.6±1.	Pr	TC	--	--	--	--	--	N	13.2±1.	SG	0.1	3.5±0.6	1.5±0.	TEc	++	C	III
1		00	21									11		9	6	11				
3	<i>Pteranthus</i>	27.9±1.	27.3±1.	Spr	TC	--	--	--	--	--	N	21.2±1.	M	0.1	7.8±1.9	2.8±0.	TEc	++	C	I
2	<i>dichotomous</i>	52	66									31	G	4	5	22				
3	<i>Silene</i>	45.2±2.	45.2±2.	Sp	PP	20±2.	6.0±0.	3.8±0.	0.6	+++	--	--	--	--	--	2.5±0.	TEc	++	A	I
3	<i>aegyptiaca</i>	32	32			00	05	08	3							02				
3	<i>S. apetala</i>	34.5±3.	34.5±3.	Sp	PP	16±1.	4.5±0.	6.8±0.	0.9	+	--	--	--	--	--	3.5±0.	TEc	++	A	III
4		11	11			00	22	22	5							60				
3	<i>S. arabica</i>	35.8±4.	35.8±4.	Sp	PP	20±0.	4.2±0.	3.8±0.	0.8	+++	--	--	--	--	--	2.3±0.	TEc	++	A	I
5		22	22			52	33	22	1							22				
3	<i>S. behen</i>	42.8±2.	42.8±2.	Sp	PP	20±0.	6.2±0.	4.5±0.	0.7	+	--	--	--	--	--	3.8±0.	TEc	+	A	III
6		82	82			22	45	52	3							38				
3	<i>S. biappendiculata</i>	48.2±1.	48.2±1.	Sp	PP	22±0.	4.5±1.	3.8±0.	0.8	+	--	--	--	--	--	2.3±0.	TME	++	A	II
7		88	88			25	22	63	4							38	P	+		
3	<i>S. conoidea</i>	42.2±1.	42.2±1.	Sp	PP	22±0.	4.5±0.	3.8±0.	0.8	+	--	--	--	--	--	2.8±0.	TME	++	A	II
8		62	62			55	50	58	4							52	P			
3	<i>S. colorata</i>	42.2±3.	42.2±3.	Sp	PP	22±0.	3.5±0.	4.2±0.	1.2	+	--	--	--	--	--	2.5±0.	TME	++	A	II
9		82	82			50	52	22	0							32	P			
4	<i>S. gallica</i>	40.3±2.	40.3±2.	Sp	PP	22±0.	4.5±0.	5.2±0.	1.1	+	--	--	--	--	--	2.3±0.	TME	+	A	II
0		22	22			22	72	35	2							33	P			
4	<i>S. linearis</i>	33.8±2.	33.8±2.	Sp	PP	16±0.	4.2±0.	4.5±0.	1.0	+	--	--	--	--	--	2.8±0.	TME	++	A	II
1		50	50			62	44	22	7							25	P			
4	<i>S. longipetala</i>	41.2±1.	41.2±1.	Sp	PP	12±0.	5.5±0.	4.8±0.	0.8	+	--	--	--	--	--	2.6±0.	TME	++	A	II
2		30	30			33	52	33	7							38	P			
4	<i>S. nocturna</i>	39.5±2.	39.5±2.	Sp	PP	16±0.	2.8±0.	6.8±0.	0.4	+++	--	--	--	--	--	2.8±0.	TME	++	A	I
3		80	80			22	45	72	1							42	P			
4	<i>S. oliveriana</i>	46.4±3.	46.4±3.	Sp	PP	18±0.	3.8±0.	5.8±0.	0.6	+++	--	--	--	--	--	2.3±0.	TEc	+	A	I
4		62	62			18	45	77	6							22				
4	<i>S. palestina</i>	42.5±2.	42.5±2.	Sp	PP	22±1.	4.2±0.	5.2±0.	0.8	+	--	--	--	--	--	2.6±0.	TEc	+	A	II
5		62	62			02	45	52	1							38				
4	<i>S. pseudotocion</i>	38.2±1.	38.2±1.	Sp	PP	16±2.	3.8±0.	5.2±0.	0.7	+	--	--	--	--	--	2.3±0.	TME	+	A	II
6		82	82			02	72	22	3							22	P			
4	<i>S. rubella</i>	43.5±1.	43.5±1.	Sp	PP	22±1.	4.2±0.	4.5±0.	0.9	+	--	--	--	--	--	3.0±0.	TME	+	A	II
7		52	52			05	35	45	3							28	P			
4	<i>S. setacea</i>	40.2±3.	40.2±3.	Sp	PP	22±1.	4.8±0.	5.2±0.	0.9	+	--	--	--	--	--	2.5±0.	TME	+	A	II
8		82	82			22	75	48	2							22	P			
4	<i>S. succulenta</i>	42.8±2.	42.8±2.	Sp	PP	22±1.	5.2±0.	5.4±0.	0.9	+	--	--	--	--	--	3.0±0.	TME	++	A	II

9		42	42			02	85	85	6							28	P			
5	<i>S. tridentata</i>	38.8±2.72	38.8±2.72	Sp	PP	20±1.05	4.2±0.82	4.8±0.88	0.8+6	+	--	--	--	--	--	3.0±0.32	TME	++	A	II
5	<i>Spergula arvensis</i>	29.8±1.52	28.9±0.36	Spr	TeC	--	--	--	--	--	N	21.8±3.12	Sm	0.25	7.8±1.05	2.3±0.32	TME	+	C	IV
5	<i>S. fallax</i>	24.6±2.22	23.4±2.22	Spr	HC	--	--	--	--	--	N	13.6±1.44	Sm	0.23	7.2±0.55	2.5±0.11	TME	++	C	IV
5	<i>Spergularia bocconii</i>	23.8±2.22	19.8±1.22	Pr	HpC	--	--	--	--	--	W	21.2±2.82	SG	0.16	6.2±1.02	2.2±0.22	TME	++	C	IV
5	<i>Spergularia diandra</i>	20.2±1.25	19.2±1.55	Spr	HC	--	--	--	--	--	N	12.3±0.22	Sm	0.28	5.8±1.02	2.2±0.12	TME	++	C	IV
5	<i>S. marina</i>	24.2±1.11	18.8±1.22	Pr	HpC	--	--	--	--	--	W	13.5±1.66	D	0.29	5.6±1.22	2.3±0.22	TME	++	C	IV
5	<i>S. media</i>	17.5±0.22	16.0±0.52	Spr	HpC	--	--	--	--	--	N	14.0±0.52	Sm	0.29	4.6±1.02	1.5±0.15	TME	++	C	IV
5	<i>S. rubra</i>	24.2±2.82	21.5±1.12	Pr	HpC	--	--	--	--	--	N	13.5±0.55	Sm	0.29	6.8±1.05	2.2±0.28	TME	++	C	IV
5	<i>Sphaerocoma hookeri</i>	23.2±1.92	19.8±2.02	Pr	TC	--	--	--	--	--	N	22.8±2.05	D	0.11	6.6±1.95	2.5±0.92	TME	++	C	III
5	<i>Stellaria media</i>	35.2±1.82	35.2±1.82	Sp	PP	12±1.00	4.5±0.82	4.2±0.55	1.07	++	--	--	--	--	--	3.3±0.88	TEc	++	A	III
6	<i>S. pallida</i>	36.2±1.55	36.2±1.55	Sp	PP	12±1.02	4.6±0.33	4.8±0.56	0.96	++	--	--	--	--	--	3.3±1.00	TGr	++	A	III
6	<i>Telephium sphaerospermum</i>	20.5±1.22	20.5±1.54	Sp	TC	--	--	--	--	--	N	16.8±1.22	Sm	0.28	6.5±1.02	2.5±0.35	TEc	+	C	II
6	<i>Vaccaria oxydonta</i>	39.2±0.82	39.2±0.82	Sp	PP	12±1.82	4.2±0.75	5.2±0.85	0.81	++	--	--	--	--	--	2.3±0.22	TEc	++	A	I
6	<i>V. pyramidata</i>	40.5±1.22	40.5±1.22	Sp	PP	12±1.55	4.2±0.25	5.4±1.04	0.78	++	--	--	--	--	--	3.0±0.22	TEc	+	A	III

3.3. Taxonomic Evaluation

3.3.1. I-Subfamily Alsinoideae

a-Tribe Alsineae

This tribe was represented by two genera *Cerastium* and *Stellaria* with four species. The genus has apolar symmetric spheroidal pantoporate pollen grains with moderate pollen sizes. The pores were from 12 to 16 small or medium in diameter, with moderately granulated pore membrane. The exine thick with tectate microechinate perforate, tectate echinate or tectate granulate ornamentation. These four species clustered together in group AIII.

b-Tribe Arenarieae

This tribe was represented by the genus *Arenaria* with two species. The genus has apolar symmetric spheroidal pantoporate pollen grains with moderate pollen sizes. The number of pores was from 12 to 18 moderate in diameter, with densely granulated membrane. The exine thickness was thin with tectate microechinate perforate ornamentation. These two species clustered together in group AI.

3.3.2. II-Subfamily Caryophylloideae

a-Tribe Caryophylleae

This tribe was represented by the three genera *Dianthus*, *Gypsophilla* and *Vaccaria* with six species. The genera have apolar or isopolar symmetric oblate or spheroidal pollen grains. The pollen size was moderate in the five species, while in *Gypsophilla capillaris* it was small. Apertures were pantoporate from 8 to 12 moderate or large in diameters with sparse granules over the pores in the five species, while in *G. capillaris* they were tricolpate. The exine moderate in the five species, very thin in *G. capillaris*, with tectate microechinate perforate, tectate echinate or reticulate ornamentation. The six species clustered in various groups and subgroups. The two *Dianthus* species came in group BII, the two *Vaccaria* species in group AI and AIII mainly for their different exine features. The two *Gypsophilla* species came apart from each other *G. capillaris* in group CII and *G. viscosa* in group BI for their great variations in all the pollen features.

b-Tribe Sagineae

This tribe was represented by the two genera *Bufonia* and *Minuartia* with six species. The genera have apolar or isopolar symmetric oblate, spheroidal or prolate pollen grains. The pollen size was small or moderate. Apertures were of two types, four species have pantoporate apertures and two have tricolpate apertures. The pantoporate ones have from 6 to 12 moderate or large in diameters with sparse granules over the pores in the three species, while in *Bufonia multiceps* the granules over the pore membrane were dense. The two tricolpate species belonging to genus *Minuartia* have long colpi with apocolpial index from 0.21 to 0.26 with dense granulated membrane. The exine moderate in the four pantoporate species, thin in tricolpate species, with tectate microechinate perforate, tectate echinate or tectate granulate ornamentation. The six species clustered in various groups and subgroups. Genus *Bufonia* came in group BI as it has 6 pores/pollen covered by dense granules and have tectate microechinate perforate exine. The five *Minuartia* species came in two groups, AIII and CIII. The two *Minuartia* species with tricolpate apertures clustered in group CIII, while the three with pantoporate apertures clustered in group AIII.

C-Tribe Sileneae

This tribe was represented by one genus *Silene* with eighteen (18) species. The genus has apolar symmetric spheroidal pantoporate pollen grains with moderate pollen sizes. The number of pores varied from 16 to 22 moderate or large in diameter, with sparse or densely granulated membrane. The exine was moderate with tectate microechinate perforate or tectate echinate ornamentation. The eighteen species clustered within the three subgroups under group A according to variations in pollen size, number of pores and exine ornamentation

3.3.3.III-Subfamily Paronychioideae

a-Tribe Paronychieae

This tribe was represented by the three genera *Herniaria*, *Gymnocarpos* and *Paronychia* with seven species. The genera have apolar symmetric spheroidal pollen grains. The pollen size was small. Apertures were pantoporate from 6 to 10 moderate in diameters within six species and very large in *G. decandrum*. The pores ornamented by moderate to dense granules. The exine moderate in thickness with tectate microechinate or tectate granulate ornamentation in six species while it was foveolate in *G. decandrum*. These seven species were clustered together in group BI.

b-Tribe Polycarpeae

This tribe was represented by six genera *Cometes*, *Loeflingia*, *Polycarpeae*, *Polycarpon*, *Pteranthus* and *Sphaerocoma* with eleven species. The genera have isopolar symmetric subprolate or prolate pollen grains. The pollen size was small or moderate. Apertures were tricolpate with long or even short colpi and apolar indices varied 0.11 to 0.30. The exine thin or moderate in thickness with tectate microechinate perforate, tectate echinate or tectate granulates ornamentation. These eleven species were clustered together in group C within three subgroups (I, II & III) according to variations in pollen size and shapes beside colpi length and exine ornamentation.

c-Tribe Corrigioleae

This tribe was represented by two genera *Telephium* and *Holosteum* with two species. The two genera were completely different in their pollen morphological features. They shared their spheroidal shapes with apolar symmetric appearance. The pollen size was small with tricolpate aperture in *Telephium sphaerospermum* and moderate with pantoporate (12) aperture in *Holosteum umbellatum*. The exine moderate in thickness with tectate echinate ornamentation in *Telephium sphaerospermum* or reticulate in *Holosteum umbellatum*. These two species were clustered in two different groups, *Holosteum umbellatum* in group BII while *Telephium sphaerospermum* in group CII based on their pollen features variations.

d-Tribe Sperguleae

This tribe was represented by two genera *Spergula* and *Spergularia* with seven species. The two genera were similar in their pollen morphological features. They have isopolar, symmetric, subprolate or prolate shapes with tetra- hexa- or even penta-colpate apertures. The pollen size was small to

medium in size with short or moderate colpi with apocolpial index from 0.16 to 0.29 with smooth or granulated colpi membranes. The exine thin to moderate in thickness with tectate microechinate ornamentation. These two genera were clustered in group CIV as its own subgroup for their common pollen features.

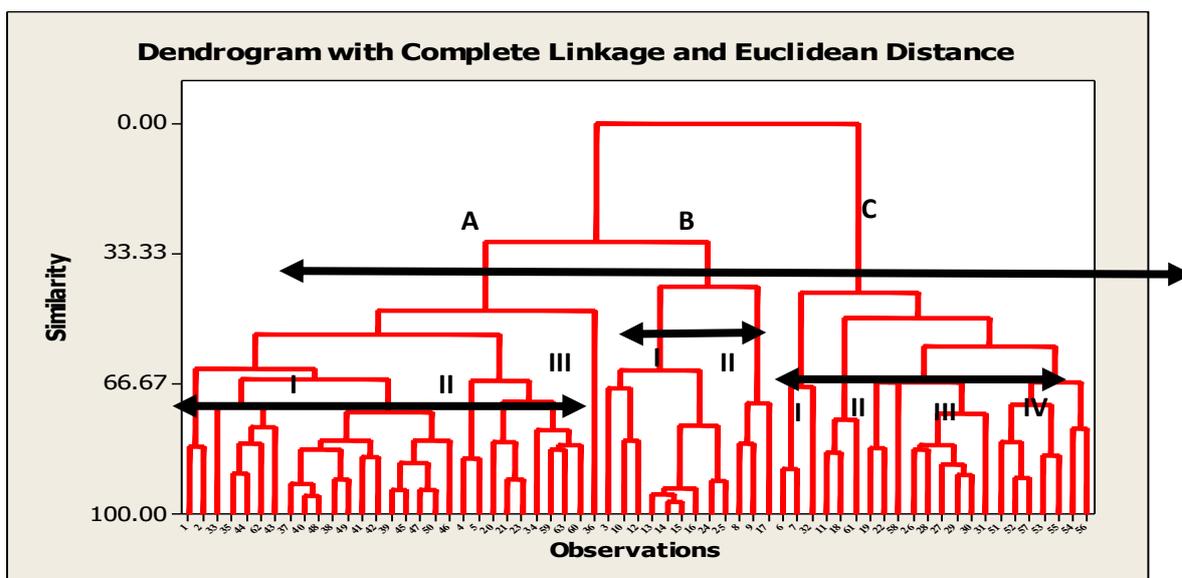
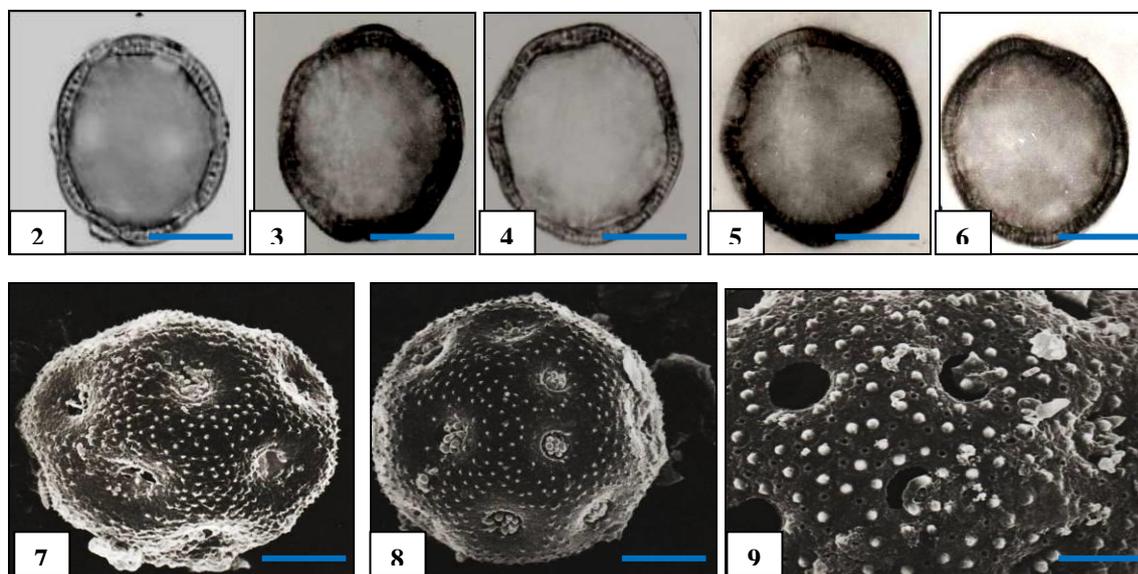


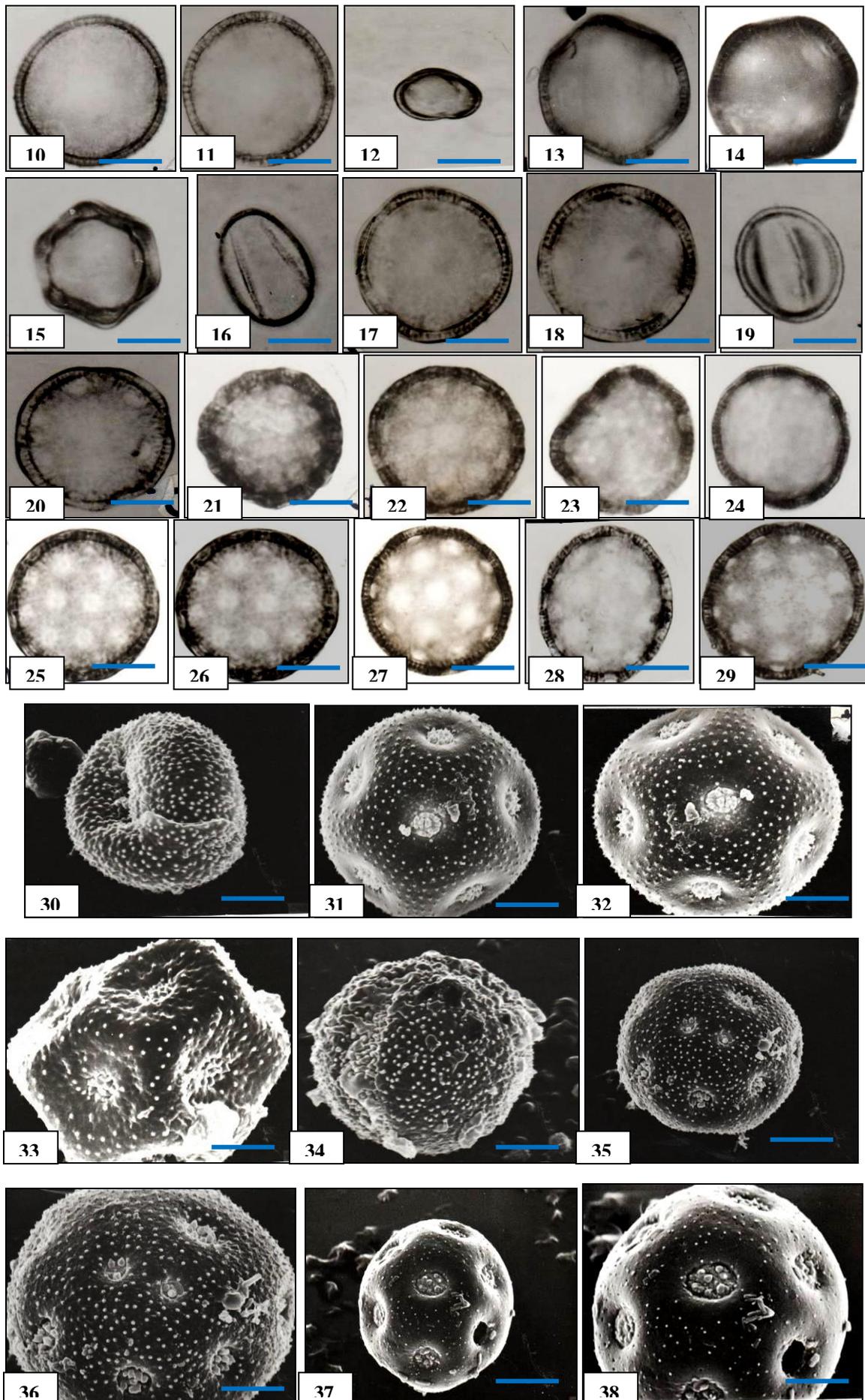
Fig1. clustering dendrogram of the studied taxa according to their pollen characters

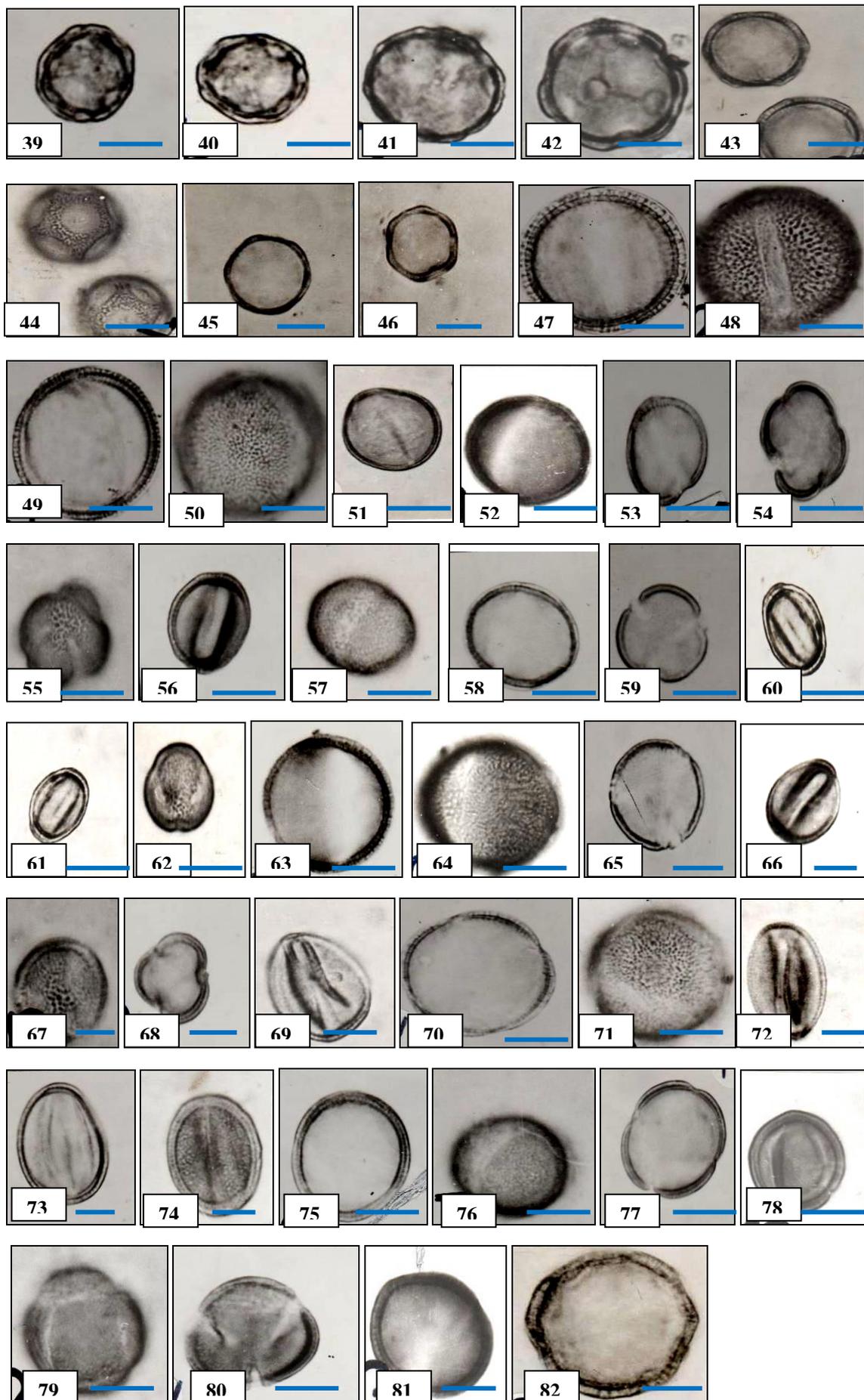
Group A I includes seven taxa *Arenaria deflexa*, *A. serpicifolia*, *Silene aegyptiaca*, *S. arabica*, *S. nocturna*, *S. oliveriana* & *Vaccaria oxyodonta*, Subgroup A II includes twelve taxa *S. biappendiculata*, *S. conoidea*, *S. colorata*, *S. gallica*, *S. linearis*, *S. longipetala*, *S. palestina*, *S. pseudotocion*, *S. rubella*, *S. setacea*, *S. succulent* & *S. tridentate*, Subgroup A III includes ten taxa *Cerastium dichotomum*, *C. viscosum*, *Minuartia meyeri*, *M. picta*, *M. tenuifolia*, *Silene apeta*, *S. behen*, *Stellaria media*, *S. pallida*, *Vaccaria pyramidata*

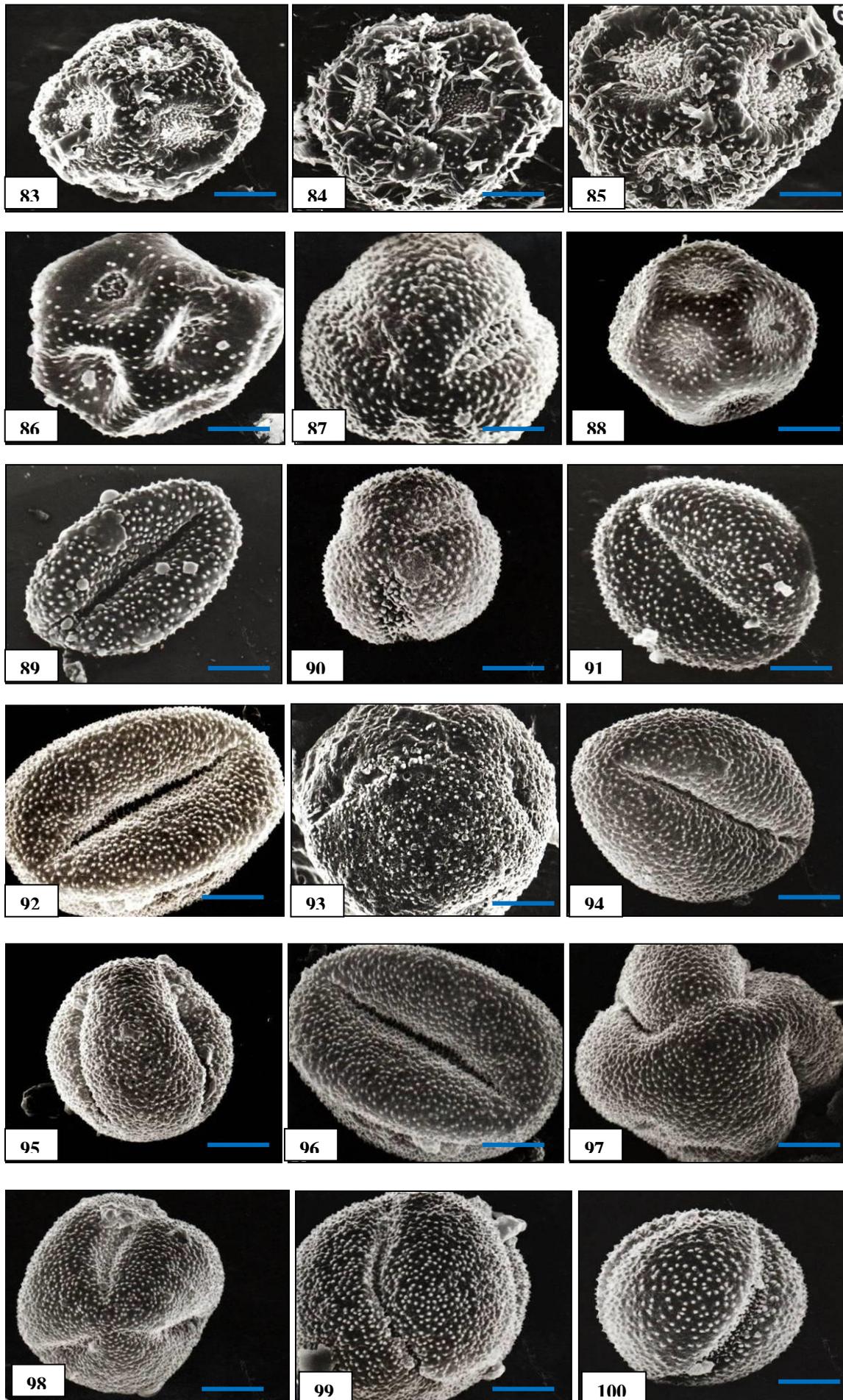
Subgroup B I includes nine taxa *Bufonia multiceps*, *Gymnocarpus decandrum*, *G. viscosa*, *Herniaria fontanesii*, *H. glabra*, *H. hirsuta*, *H. hemistemon*, *Paronychia argentea* & *P. nivea*, Subgroup B II includes three taxa *Dianthus cyri*, *D. strictus* & *Holosteum umbellatum*

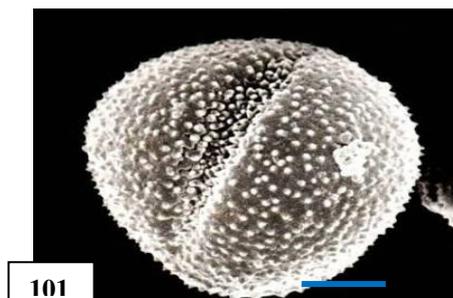
Subgroup C I includes three taxa *Cometes abyssinica*, *C. surattensis* & *Pteranthus dichotomus*, Subgroup C II includes three taxa *Gypsophilla capillaris*, *Loeflingia hispanica* & *Telephium sphaerospermum*, Subgroup C III includes nine taxa *Minuartia geniculata*, *Minuartia procumbens*, *Polycarpeae corymbosa*, *P. repens*, *P. spicata*, *Polycarpon prostratum*, *P. succulentum*, *P. tetraphyllum* & *Sphaerocoma hookeri*, Subgroup C VI includes seven taxa *Spergula arvensis*, *S. fallax*, *Spergularia bocconii*, *S. diandra*, *S. marina*, *S. media* & *S. rubra*











Figs. 2-9 Photographs of the studied taxa from subfamily Alsinoideae, Figs.2-6 LM photographs 2- *Arenaria deflexa* , 3- *Cerastium dichotomum* , 4- *C. viscosum*, 5- *Stellaria media*, 6- *S.pallida*, Figs. 7-9 SEM photographs 7-*Stellaria media*, 8 &9- *S. pallida*, Figs. 10-38 Photographs of the studied taxa from subfamily Caryophylloideae, Figs. 10-29 LM photographs ,10- *Dianthus cyri*, 11- *D. strictus*, 12- *Gypsophilla capillaries*, 13- *G. viscosa*,14- *Vaccaria pyramidata*, 15- *Bufonia multiceps*, 16- *Minuartia geniculata*, 17- *M. meyeri* , 18- *M.picta*, 19- *M. procumbens*, 20- *M. tenuifolia*,21- *Silene apetala*, 22- *S. gallica* , 23-*S. linearis*, 24- *S. palestina*, 25- *S. pseudotocion* 26- *S. rubella*, 27-*S. setaceae*, 28- *S.succulenta*, 29- *S. tridentata*, Figs. 30-39 SEM photographs, 30- , SEM 30- *Gypsophilla capillaris*, 31-*Vaccaria oxyodonta*, 32- *V. pyramidata*, 33- *Bufonia multiceps*, 34- *Minuartia geniculata*, 35- *M. meyeri*, 36- *M. picta*, 37 & 38- *M. procumbens*, Figs. 39-38 Photographs of the studied taxa from subfamily Paronychioideae, Figs 39-82 LM photographs, 39- *Herniaria fontanesii* , 40- *H. glabra*, 41- *H. hirsuta*, 42- *H. nemistemon*, 43 & 44- *Gymnocarpos decandrum*, 45- *Paronychia argentea*, 46- *P. nivea* , 47 & 48-*Cometes abyssinica*, 49 & 50- *Cometes surattensis*, 51 & 52- *Loeflingia hispanica*, 53, 54, 55 & 56- *Polycarpon tetraphyllum*, 57 & 58- *P. prostratum*, 59- *P. succulentum*, 60, 61 & 62- *Sphaerocoma hookeri*, 63 & 64- *Pteranthus dichotomous*, 65 & 66- *Polycarpaea corymbosa*, 67 & 68- *P.repens* 69- *P.spicata* , 70 & 71- *Spergularia arvensis*, 72- *Spergularia bocconii*, 73& 74- *S. marina*, 75 & 76- *S. media*, 77, 78 & 79- *S. rubra* , 80- *Telephium sphaerospermum*, 81 & 82- *Holosteum umbellatum*, Figs 83-101 SEM photographs, 83, 84 & 85- *Gymnocarpos decandrum*, 86- *Herniaria hemistemon*, 87- *Loeflingia hispanica*, 88- *Paronychia argentea*, 89 & 90- *Polycarpon repens*, 91-*Polycarpon tetraphyllum*, 92- *Spergularia arvensis*, 93- *Spergularia bocconii*, 94 & 95- *S. media*, 96 & 97- *S. rubra*, 98 & 99- *S. marina*, 100 & 101- *Sphaerocoma hookeri*, Magnification bar

Bar= 15 µm in 2, 3, 4, 5, 6, 10, 11, 14, 17, 18, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29. 47, 48, 49, 50, 63, 64, 70, 71, 81, 82.

Bar= 12 µm in 13, 11, 16, 19,,36, 43, 44.

Bar= 10 µm in 7, 8, 15, 31, 32, 35, 37, 39, 40, 41, 42, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 65, 66, 67, 68, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81 , 82.

Bar=5 µm in 12, 30, 83, 84, 91, 93, 96, 97, 98, 99, 100, 101.

Bar= 3 µm in 31, 33, 34, 38, 86, 87, 88, 89, 90, 92, 94, 95.

Bar= 2 µm in 9, 85.

4. DISCUSSION

Pollen morphological features proved to be of great importance for plant identification and solving taxonomic confusion since the bigening of the last century. Family Caryophyllaceae with its great diversity in its morphological characters and the confusion in the division of its taxa has drawn the attention of many taxonomists to clarify the relation between the genera and its species. All the taxonomic grouping of the Caryophyllaceae taxa conducted on the morphological and molecular investigations based on certain genera and species. Few works have been done on most of the genera within the three proposed subfamilies as well as the pollen characters of most of the genera [9-17, 20, 22, 24-26, 32-34] beside others. [35] Considered the diversity of the whole orde, Caryophyllales with less consideration of the family groups. This work was conducted on the pollen grain features of most of the genera belonging to the three subfamilies; Alsinoideae, Caryophylloideae and Paronychioideae; with 63 species with representatives of the main tribes. These species were representing most of the Egyptian Caryophyllaceae in the Flora of Egypt [28]. In this study, the important characters of the pollen grain features were compared and treated as self identity without the consideration of their taxonomic ranking. The characters of the pollen grains in each species were subjected to PAST program to illucidate the relationship between the studied species and resulting in clustering dendrogram of the studied species (Fig.1). The dendrogram grouped the studied taxa into three main groups and nine subgroups. These groups are in partial agreement with [19]. The pollen characters of the studied taxa were varied greatly in their shapes; oblate, spheroidal, subprolate or even prolate; sizes; small, medium or even large and pollen aperture types and numbers. Accordingly the

taxonomic evaluation of the pollen grains within the studied taxa according to their traditional and recent taxonomic status will be discussed.

Subfamily Alsinoideae; with its two tribes; showed great symmilarities between its studied species. This subfamily has apolar spheroidal pantoporate pollen grains with tectate microechinate perforate, tectate echinate or tectate granulate ornamentation. Tribe Alsineae came in group AIII while tribe Arenarieae came in group AI and the two tribes are greatly correlated to each other. This result support [14,16,33,34], in considering the pollen morphological features of limited use to be the first step in the systematic discrimination within the three taxonomic groups, but it can help in their phylogenetic position. They considered the taxa within subfamily Alsinoideae have advanced palynological characters and have similarities in their pollen grain features. They reported that the two genera; *Arenaria* and *Cerastium*; related to the genus *Stellaria* and they considered *Stellaria* in a middle evolutionary position in the Caryophyllaceae family. This merging of the genera was mentioned before by [19].

Subfamily Caryophylloideae; with its three tribes; mostly have apolar symmetric moderate size spheroidal pantoporate pollen grains with different numbers of pore/pollen. Except in *Gypsophilla capillaris*, tribe Caryophylleae and two *Minuartia* species; *M. geniculata* and *M. procumbens*; tribe Sagineae, they have isopolar symmetric small size oblate or prolate tricolpate pollen grains. These three species were clustered in the C group while the rest of the studied Caryophylloideae were clustered in the A group. The eighteen studied species tribe Sileneae has homogenous pollen characters with variations in pollen diameters and number of pores and all grouped in cluster A. This result coordinates with that of [36] as they considered the number of pores and exine ornamentation have important systematic significance within the *Silene* species. [25] Found that pollen wall ornamentation, pore characteristics, and size variations within the Iranian *Minuartia* species can be of use to understand their taxonomic significance within the family. This observation approved that of [37] who recorded two types of pollen grains within *Minuartia* species. This finding coordinate with the *Minuartia* species studied in this investigation, as two types of pollen grain apertures were recorded within the studied *Minuartia* species. [11] Found that the variations in the pollen grains within the *Minuartia* species were more than two types as they considered both the aperture types and pollen shapes and sizes in their division. According to [38] an infrageneric classification of *Minuartia* included four subgenera and 12 sections within subg. *Minuartia*. [7] found that the *Minuartia* species are polyphyletic and has different origins; this can be valuable finding when considering the aperture types and number in the discrimination of the species. The two tricolpate *Minuartia* species, beside *Gypsophilla capillaris* have pollen characters similar to those of subfamily Paronychioideae. The same variations in the pollen characters within the *Minuartia* species had been recorded by [38] who pointed to subgenus *Rhodalsine* has trizonocolpate with a relatively thin exine and it differs from that of all other species of the subfamily Alsinoideae. Species of this subfamily have pollen grains that are pantoporate with a thick exine. They concluded that pollen characters of the species of subgenus *Rhodalsine* showed relationship with those of *Spergularia* (subfamily Paronychioideae) as pointed by [19]. [39] Recorded significant variations within the seeds of *Minuartia* species which cannot fit with the taxonomical sections of the genus. [40] Concluded that the pollen grain characters of *Dianthus* species were variable and did not coordinste with the sectional classification of the genus. This conclusion is in agreement with the pollen grain results obtained in this study.

Subfamily Paronychioideae; with its four tribes; have two different types of pollen grains. Tribe Paronychieae has pollen grains similar to the above mentioned two subfamilies with spheroidal pantoporate apertures, but differs in their pollen grain size and number of pores. The porate pollen grains within the *Paronychia* species have been recorded by [9] in the Turkish species, this tribe clustered in the B group. The other three tribes, Polycarpeae, Corrigioleae and *Sperguleae* have isopolar symmetric pollen grains with oblate, subprolate or prolate shapes. The pollen grains were small or moderate in size and have colpate aperture. Tribe Corrigioleae has two types of pollen grain, the *Holosteum* type which is similar to the subfamily Alsinoideae and the *Telephium* type which is similar to the subfamily Paronychioideae. The pollen data obtained in this investigation are in acceptance to those obtained by [19] who suggested merging the two genera *Spergula* and *Spergularia*, splitting tribe Polycarpeae into three larger and several smaller genera and transferring members of *Paronychia* to *Herniaria*. Tribe *sperguleae* characterized by the multiple numbers of

colpi which made this tribe distinguished from the whole family and clustered in separate subgroup under the C group (CIV).

4.1. Significance Statement

The taxonomy of the Caryophyllaceae taxa is still confusing and needs more scientific works. Pollen grain characters have aid in solving many taxonomic problems and used in the identification of many species. This work has been done to investigate the pollen grains of 63 species representatives of the three taxonomic subfamilies to clarify their features and to which degree they can help in the grouping of the family.

5. CONCLUSION

This work demonstrates that aperture type, number of pores and colpi, the pollen surface ornamentation beside the density of microechini have important systematic significance. The results support the taxonomic classification of the family into four subfamilies as mentioned by [5]. He divided the family into four subfamilies: Alsinoideae, Caryophylloideae, Paronychioideae, and Polycarpoideae. The separation of the Paronychiea from subfamily paronychioideae was supported by the pollen characters and the two genera *Gypsophylla* and *Minuartia* need reclassification. [6] Proposed new classification according to molecular study of the family with eleven clades named as tribes. This proposal approved by⁷ and most recent molecular works done by [36, 41, and 42]. The pollen morphological characters showed differences within the three subfamilies and within the genera and species under each subfamily and can be used in the identification of the studied taxa. These differences were not coordinate with the vegetative and floral taxonomical classifications. The pollen diversity within the studied Caryophyllaceae supports the molecular phylogenetic works in suggesting that the traditionally recognized subfamilies are non-monophyletic and reclassification of the genera and tribes must be considered. Meanwhile the present study supports Rabeler and Hartman [5] in the taxonomic division of the family into three subfamilies and many tribes.

6. CONFLICT OF INTEREST

The author declared that this work has no conflict of interest.

7. FUNDING

No funding agencies support this work.

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Citation: Wafaa Kamal Taia. *Insight on the Taxonomy of Caryophyllaceae Juss. Based on Pollen Grain Diversity. International Journal of Advanced Research in Botany.* 2025; 9(1):26-44. DOI: <https://doi.org/10.20431/2455-4316.0901003>.

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