Pak. J. Bot., 43(2): 731-741, 2011.

POLLEN MORPHOLOGY OF SOME *ONOSMA* SPECIES (BORAGINACEAE) FROM TURKEY

RIZA BİNZET*

Department of Biology,
Science and Arts Faculty, Adıyaman University, 02040, Adıyaman, Turkey

Abstract

The pollen morphology of nine taxa three of which are endemics belonging to *Onosma* L. (Boraginaceae); *O. orientale, O. halophilum, O. bourgaei, O. chlorotrichum, O. heterophyllum, O. ambigens, O. oreodoxum, O. sintenisii* and *O. bulbotrichum* from Turkey has been investigated by light microscopy and scanning electron microscopy. The shape of pollen grains of *Onosma* is frequently prolate, subprolate and sphaeroidea. The outline in polar view is subrounded and subtriangular. The dimension for the polar length ranges between 13.01-21.57 µm, equatorial width 10.56-20.30 µm, colpi length 8.19-16.58 µm, colpi width 1.69-4.36 µm, pori length 1.05-4.28 µm and pori width 2.52-10.71 µm. The pollen morphology of studied taxa are taxonomically of significant characters. The main pollen morphology differences have been found at the section level, especially in pollen type.

Introduction

The *Onosma* L. (Boraginaceae) genus with c.150 species having its center of distribution and maximum concentration of species in Iran and westwards into Syria and Turkey, eastwards and northeastwards to Turkestan, Altai, China, Afghanistan, Pakistan, Kashmir, Tibet and India to Burma (Qureshi & Qaiser, 1987; Al-Shehbaz, 1991; El-Shazly, 2003; Naz *et al*., 2006). The genus of *Onosma* is represented by about 102 taxa (97 species) in Turkey and the rate of endemism among native species is 50%. *Onosma* has about 50 endemic species and one endemic variety (Riedl, 1978; Davis *et al*., 1988; Yıldırımlı, 2000; Riedl *et al*., 2005 and Binzet & Orcan, 2007).

The genus has been divided into three sections: *Protonosma, Podonosma* and *Onosma*. *Protonosma* and *Podonosma* sections are represented by one species; the others *Onosma* specimens belong to *Onosma* section. This section is separated into two subsections according to solely indumentum types: *Asterotricha* (Boiss.) Gürke., and *Haplotricha* (Boiss.) Gürke.

Some *Onosma* species are used as herbs, folk medicines and dyes. In folk medicine, these plants are employed for burns, wounds and ailments (Khajuria & Jain, 1993; Özgen *et al*., 2003). The flowers of some species are eaten (Öztürk & Özçelik, 1991). Various local names are used for *Onosma* species as emzik otu (Eskişehir), emcek (Kemaliye-Erzincan) and yalancı havacıva, emzik (Kemah-Erzincan) in Turkey (Baytop, 1994).

Boraginaceae is an eurypalynous family (Clarke, 1977; Diez, 1984) in which a large number of species can be recognized by their pollen characters (Diez & Valdes, 1991). The first palynological studies in the Boraginaceae were that of Geoffery (Erdtman, 1952). Later the pollen morphology of the family Boraginaceae have been studied by Erdtman (1952), Marticorena (1968), Huynh (1971; 1972), Nowicke & Ridgway (1973), Nowicke & Skvarla (1974), Diez (1984) and Perveen, (1995).

*E-mail: rbinzet@gmail.com, rbinzet@posta.adiyaman.edu.tr

The pollen morphology of the *Onosma* species which is the difficult genus from systematic and taxonomic point of view. In order to solve these problems, detailed pollen morphological studies of some species of the genus *Onosma* have been investigated. Five of *Onosma* species (*O. mutabile, O. bracteosum, O. giganteum, O. roussaei* and *O. mersinana*) have been examined for pollen morphology by Binzet & Orcan (2003a, 2003b, 2009). Based upon the literature search, we could find no pollen characteristics about the nine taxa viz., (*Onosma* L. (Boraginaceae); *O. orientale, O. halophilum, O. bourgaei, O. chlorotrichum, O. heterophyllum, O. ambigens, O. oreodoxum, O. sintenisii* and *O. bulbotrichum*) from Turkey.

In this study, we have carried out a detailed analysis of pollen size and morphology of nine taxa of the genus *Onosma* growing in the different are of Turkey in order to correlate the palynological characteristics with the taxonomy of the genus.

Material and Methods

Onosma pollen specimens were collected from field in 2002-2006. The voucher specimens are listed in Table 1. Pollen for light microscopy (LM) was prepared using the method described by Wodehouse (1935) and Acetolysis by Erdtman (1952). For light microscopy, the pollen grains were mounted in unstained glycerine jelly and observations were made with Olympus BX 40 research microscope (x10; x100). Measurements and taking photographs of pollens were done after allowing one month for the specimens to reach normal dimensions and forms. Measurements were taken on at least 30 pollen grains. For each of the measured pollen grains the ratio of polar to equatorial axis length (P/E) was determined and the mean P/E ratio was calculated from these individual values. For scanning electron microscopy (SEM) studies, pollen grains were first treated with 70% alcohol and then dried before mounting on stubs with gold. The photomicrographs were taken with LEO 44O electron microscopes. The palynological terminology mainly follows Erdtman (1952), Faegri & Iversen (1964) and Punt *et al*., (1994).

Results and Discussion

The *Onosma* genus is a difficult group among dicotyledon plants for systematical and taxonomical reasons. Although a better understanding of exine sculpturing of the pollen of Turkish *Onosma* taxa studied previously with LM by Binzet & Orcan (2003a, 2003b), some points are not clear. In order to supply more morphological data to solve some of systematical and taxonomical problems, a detailed pollen morphological study of 9 species (Table 2) of the genus *Onosma* has been investigated by light microscopy and scanning electron microscopy.

The results of light microscopy data on the pollen grains of specimens are summarized in Table 3 and LM images of some Turkish *Onosma* species are given in Fig. 1. The outline in polar view is subrounded and subtriangular (Figs. 1-3). The dimension ranges for investigated pollen are found as the polar axis is 13.01-21.57 μm long, equatorial diameter is 10.56-20.30 μm long, colpus length 8.19-16.58 μm, colpus width 1.69-4.36 μm, porus length 1.05-4.28 μm and porus width 2.52-10.71 μm, exine 0.33-1.15 μm thick, intine 0.57-0.80 μm thick, length of polar triangular edge 4.78-7.87 μm. SEM micrographs of pollen grain (polar view, profile view and surfaces) are given in Figs. 2,3. The obtained pollen characteristics are summarized as:

***O. orientale*:** Isopolar, pollen grains sphaeroidea in shape and tricolporatae in type, the exine is intectate, sculpture rugulate-granulate, colpi margin undulated.

***O. chlorotrichum*:** Heteropolar, pollen grains sphaeroidea-subprolatae in shape, the exine is intectate, sculpture granule, 2-5 granule number per 1 μm^2 in narrow pole, sculpture of apertures are verrucate, colpi and pori margin are undulated and swollen which is characteristics.

***O. sintenisii*:** Heteropolar, pollen grains subprolatae-sphaeroidea in shape, the exine is intectate, sculpture granule, 10-11 granule number per 1 μm^2 in narrow pole, sculpture of apertures are verrucate, colpi and pori margin undulated.

***O. oreodoxum*:** Heteropolar, pollen grains subprolatae in shape, the exine is intectate, sculpture granule, 10-11 granule number per 1 μm^2 in narrow pole, sculpture of apertures are verrucate, colpi and pori margin straight-undulated.

***O. halophilum*:** Heteropolar, pollen grains subprolatae in shape, the exine is intectate, sculpture granule, sculpture of narrow colpies are psilat-granulate and 12-14 granule number per 1 μm^2, sculpture of apertures are verrucate, colpi and pori margin straight.

Table 1. The locality of the investigated *Onosma* Species.

Taxa	Locality
O. orientale (L.) L.	C6 Hatay, Musa dağı, rocky slopes, 07.V.2004, 500 m Binzet 125.
O. chlorotrichum Boiss. & Noe	B10 Van, Doğubeyazıt-Van 40 km, stoney and volcanic lands, 13.VI.2002, 1900 m, Binzet 80
O. sintenisii Hausskn. ex Bornm.	B7 Sivas, Divriği-Kemaliye 44 km, slopes, 26.V.2006, 1060 m, Binzet 99
O. oreodoxum Boiss.	C3 Antalya, Kaş around, macchie, 22.III. 2004, Binzet 123
O. halophilum Boiss. & Heldr.	C4 Konya, Konya-Aksaray 8 km, 13.VI. 2006, roadside, 1000 m, Binzet 130
O. bulbotrichum DC.	C7 Adıyaman, Karahöyük village, slopes, 24.V.2006, 650 m, Binzet 85
O. bourgaei Boiss.	A5 Amasya, Akdağ, Çalardı plateau, 28.V. 2006, Binzet 128
O. heterophyllum Griseb.	A4 Bartın, Bartın entrance, roadside and slopes, 09.VI.2007, 30m, Binzet 129
O. ambigens Lacaita	B7 Sivas, Sincan–Divriği 7 km, roadside and steppe, 26.V.2006, Binzet 127

Table 2. Section of the investigated species.

***Podonosma* section**	***Onosma* section**	
	***Haplotricha* subsection**	***Asterotricha* subsection**
O. orientale	*O. halophilum**	*O. bourgaei*
	O. chlorotrichum	*O. heterophyllum*
	O. oreodoxum	*O. ambigens**
	O. bulbotrichum	
	*O. sintenisii**	

* Endemic

Table 3. Measurements and exine characteristics of pollen grains of *Onosma* L. (M ±SD, µm)*.

Taxa	Pollen shape	P	E	plg	plt	clg	clt	ex	i	t
O. orientale (N)	Sphaeroidea	13.87±0.73	12.49±0.81	3.33±0.37	2.52±0.34	10.15±0.84	1.69±0.42	0.46	0.57	4.99
O. orientale (A)	Sphaeroidea	14.17±0.89	13.12±0.77	1.05±0.30	8.77±0.97	-	-	0.72	-	-
O. chlorotrichum (N)	Sphaeroidea	16.15±0.84	15.41±0.89	3.98±0.52	5.2±0.62	12.97±1.03	3.40±0.57	0.51	0.73	6.25
O. chlorotrichum (A)	Subprolatae	16.34±0.97	13.77±1.01	1.15±0.44	8.49±1.29	13.66±0.76	3.12±0.45	0.81	-	6.91
O. sintenisii (N)	Subprolatae	15.25±0.72	13.32±0.41	4.28±0.37	3.60±0.58	12.70±0.76	3.09±0.34	0.33	0.59	5.24
O. sintenisii (A)	Sphaeroidea	21.35±1.15	18.85±1.33	1.99±0.34	10.01±0.67	16.58±0.98	4.30±0.65	1.07	-	7.87
O. oreodoxum (N)	Subprolatae	14.59±0.70	11.28±0.53	2.98±0.64	3.46±0.33	10.57±0.72	2.71±0.38	0.61	0.77	6.12
O. oreodoxum (A)	Subprolatae	13.01±0.86	11.17±0.85	-	-	8.19±0.83	3.51±0.43	0.88	-	6.28
O. halophilum (N)	Subprolatae	15.36±0.45	12.27±0.42	3.08±0.39	3.74±0.44	10.44±0.84	3.52±0.31	0.52	0.80	7.09
O. halophilum (A)	Subprolatae	17.35±0.98	14.23±0.87	2.56±0.45	9.32±0.76	12.87±0.93	4.36±0.65	0.78	-	7.02
O. bulbotrichum (N)	Sphaeroidea	14.82±0.42	14.36±0.43	3.67±0.37	4.20±0.58	11.81±0.68	3.21±0.36	0.40	0.74	4.78
O. bulbotrichum (A)	Sphaeroidea	21.57±1.05	20.30±0.98	2.26±0.34	10.71±0.67	16.38±0.86	3.52±0.54	1.15	-	7.26
O. bourgaei (N)	Prolata	14.36±0.53	10.56±0.76	2.67±0.40	2.87±0.49	11.13±0.84	2.89±0.52	0.48	0.74	5.60
O. bourgaei (A)	Prolata	15.97±0.78	11.73±0.74	2.92±0.41	3.18±0.47	12.19±0.92	3.68±0.52	0.60	-	5.74
O. heterophyllum (N)	Prolata	16.32±0.79	11.73±0.74	2.84±0.34	3.75±0.38	11.79±0.81	2.98±0.40	0.46	0.66	7.42
O. heterophyllum (A)	Subprolatae	14.37±0.90	12.11±0.84	1.53±0.48	7.14±0.99	10.35±0.88	3.39±0.51	0.81	-	6.66
O. ambigens (N)	Prolata	17.03±0.75	11.32±0.81	2.63±0.50	3.06±0.53	12.03±0.77	1.98±0.38	0.49	0.69	7.2
O. ambigens (A)	Prolata	19.47±0.89	13.23±0.97	2.89±0.44	9.21±0.77	14.58±0.93	2.98±0.64	0.86	-	7

* N- Non acetolysed pollen grains (LM); A-Acetolysed (LM); M- Mean value; SD-Standart deviation; P- Length of polar axis; E- Width of equatorial axis; plg- Length of pores (pori); plt- Width of pores (pori); clg- Length of colpus (colpi); clt- Width of colpus (colpi); ex.- Exine thickness; i- intine thickness; t- Length of polar triangular edge (The ratio of the distance between the apices of two ectocolpi of a zonocolpate pollen grain to its equatorial diameter).

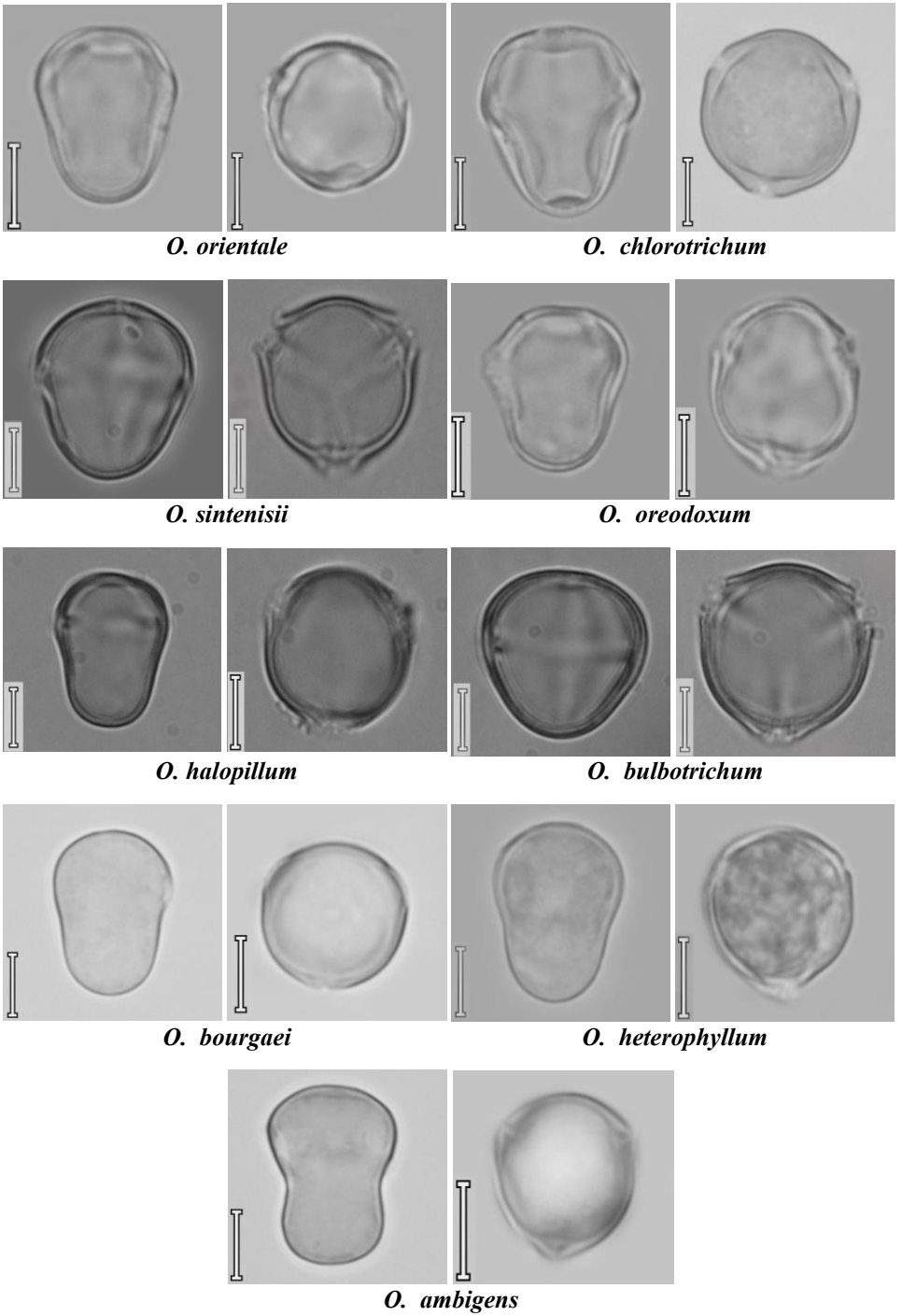
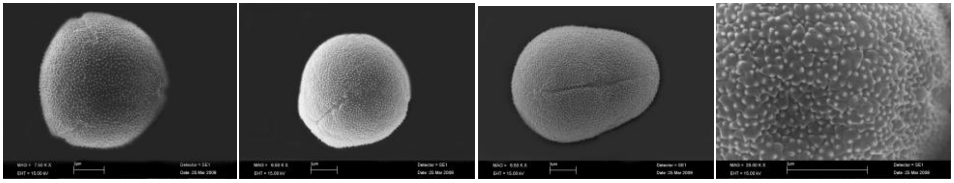
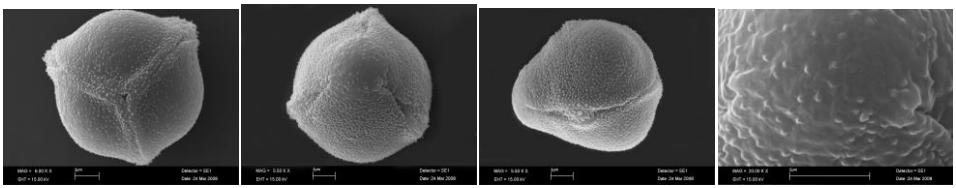


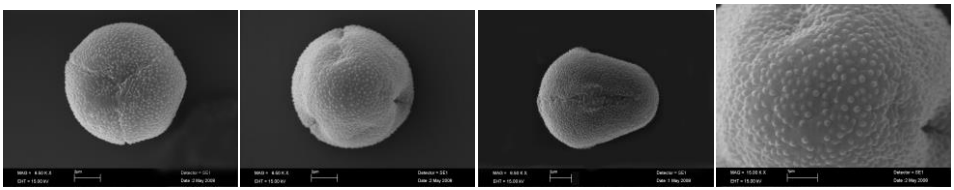
Fig. 1. LM images of some Turkish *Onosma* species, scale 6 μm.



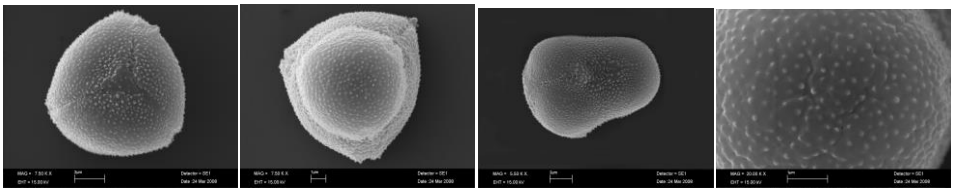
O. orientale



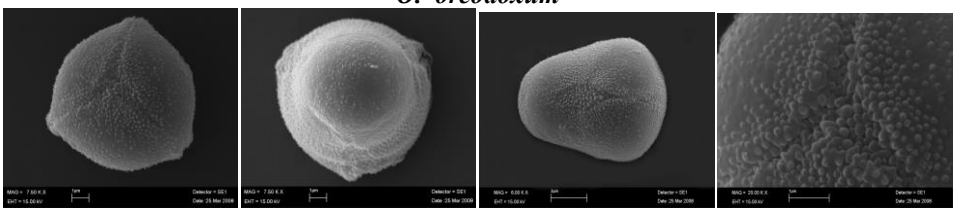
O. chlorotrichum



O. sintenisii



O. oreodoxum



O. halopillum

Fig. 2. SEM micrographs of pollen grain: polar view, profile view and surfaces.

***O. bulbotrichum*:** Heteropolar, pollen grains sphaeroidea in shape, the exine is intectate, sculpture granule, sculpture of apertures are verrucate, colpi and pori margin straight-undulated.

***O. bourgaei*:** Heteropolar, pollen grains prolata in shape, the exine is intectate, sculpture psilate-granulate, 6-8 granule number per 1 μm^2 in narrow pole, sculpture of apertures are verrucate, colpi and pori margin undulated.

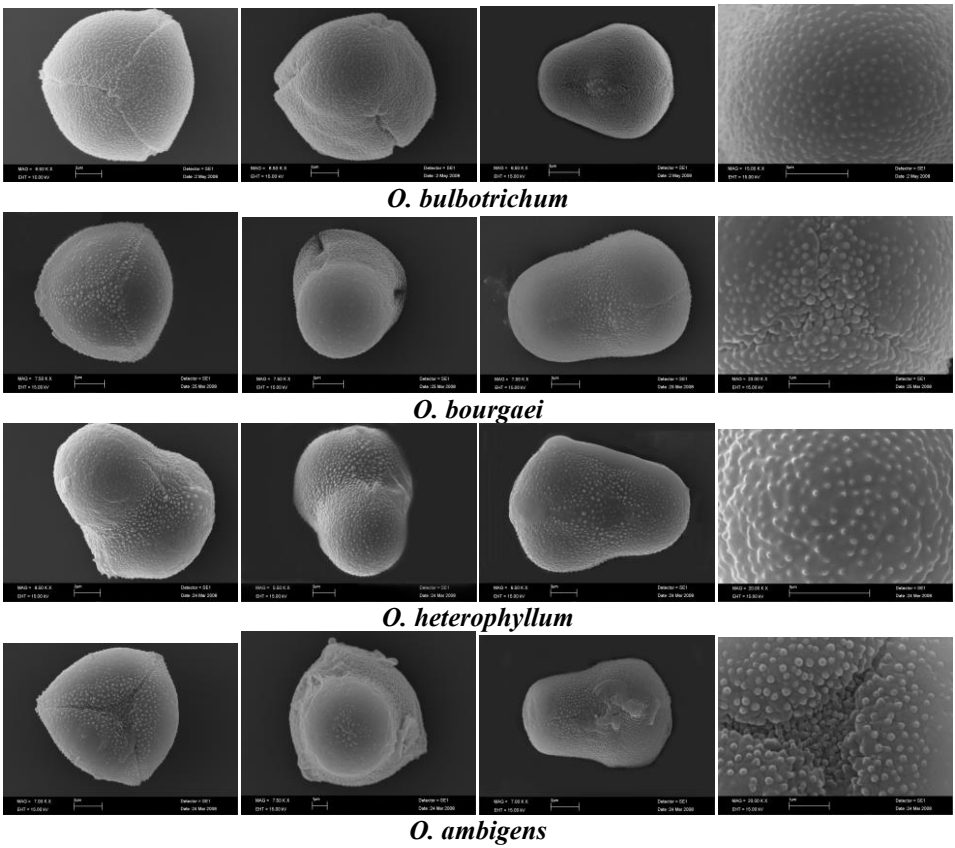


Fig. 3. SEM micrographs of pollen grain: polar view, profile view and surfaces.

***O. ambigens*:** Heteropolar, pollen grains prolata in shape, the exine is intectate, sculpture psilate-granulate, 10-14 granule number per 1 μm^2 in narrow pole, sculpture of apertures are verrucate-psilate, colpi and pori margin undulated and dense granulate.

It was previously reported that the three colpi of syncolporatae pollen in some members of the genus *Onosma* converge in one pole or even two poles (Ning *et al*., 1995). However, in the present study, we found that all *Onosma* taxa except *O. orientale* were observed to have three colpies of syncolporatae pollen at large pole and colpies are joined at the only large poles (heteropolar). In addition to the colpies of *O. orientale* do not converge in one or two poles and isopolar.

Wodehouse (1935) and Lee (1979) have reported that a direct correlation between flower and pollen size. However, Pandey (1971) demonstrated in the genus *Nicotiana* that there was no correlation between flower and pollen size and the species with the largest flowers had small pollen grains. We found that the pollen of *O. sintenisii* and *O. bulbotrichum* are smaller (15.25 μm (N) and 14.82 μm (N), respectively) but with larger flowers whereas, in *O. heterophyllum* are larger (16.32 μm (N)) with smaller flowers in this study. These obtained results are in agreement with the results Pandey (1971).

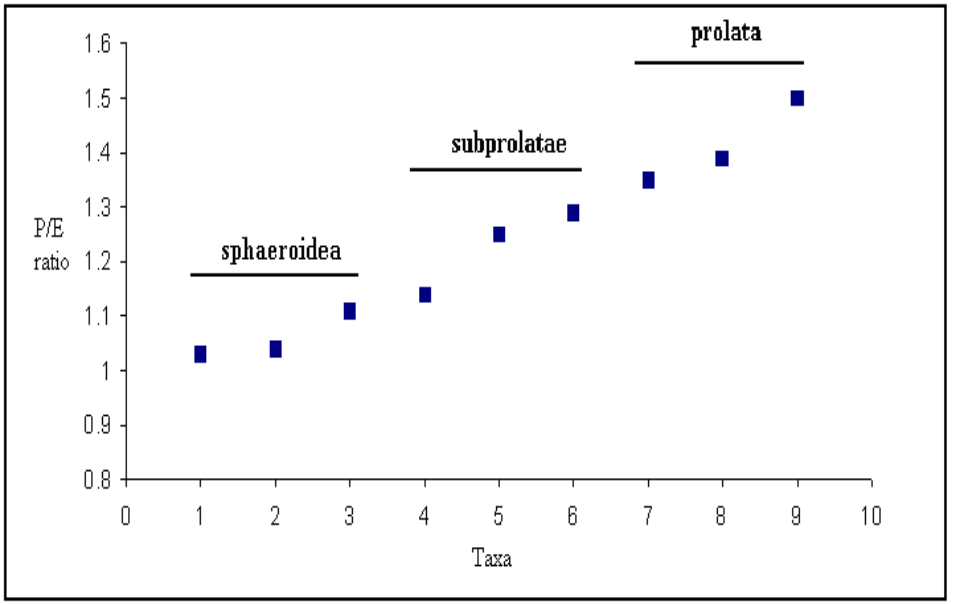


Fig. 4. P/E ratio (N), 1-*O. bulbotrichum*, 2-*O. chlorotrichum*, 3-*O. orientale*, 4-*O. sintenisii*, 5-*O. halophilum*, 6-*O. oreodoxum*, 7-*O. bourgaei*, 8-*O. heterophyllum*, 9-*O. ambigens*.

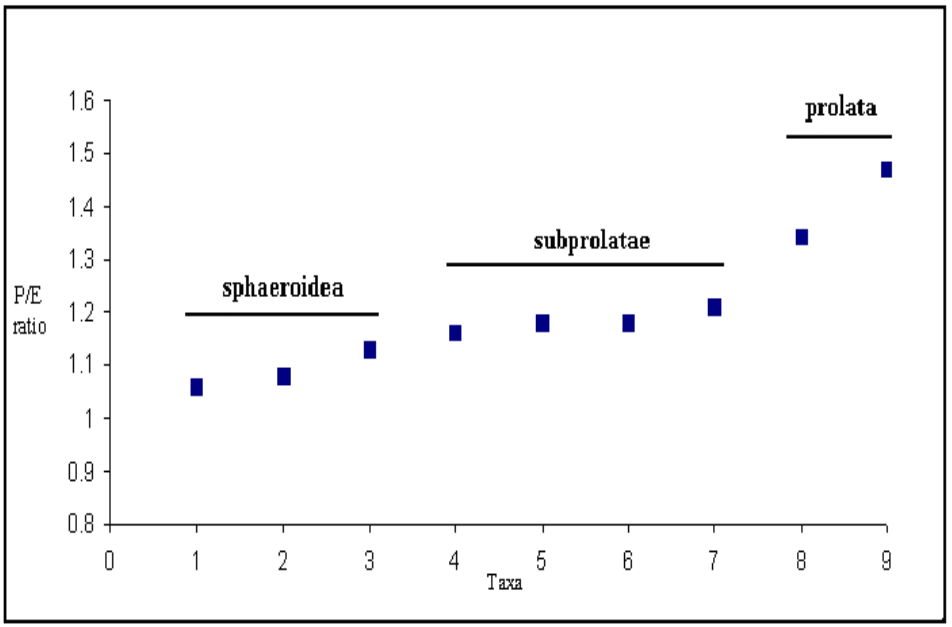


Fig. 5. P/E ratio (A). 1- *O. bulbotrichum*, 2- *O. orientale*, 3- *O. sintenisii,* 4- *O. oreodoxum,* 5- *O. chlorotrichum*, 6- *O. heterophyllum,* 7- *O. halophilum*, 8- *O. bourgaei,* 9- *O. ambigens*.

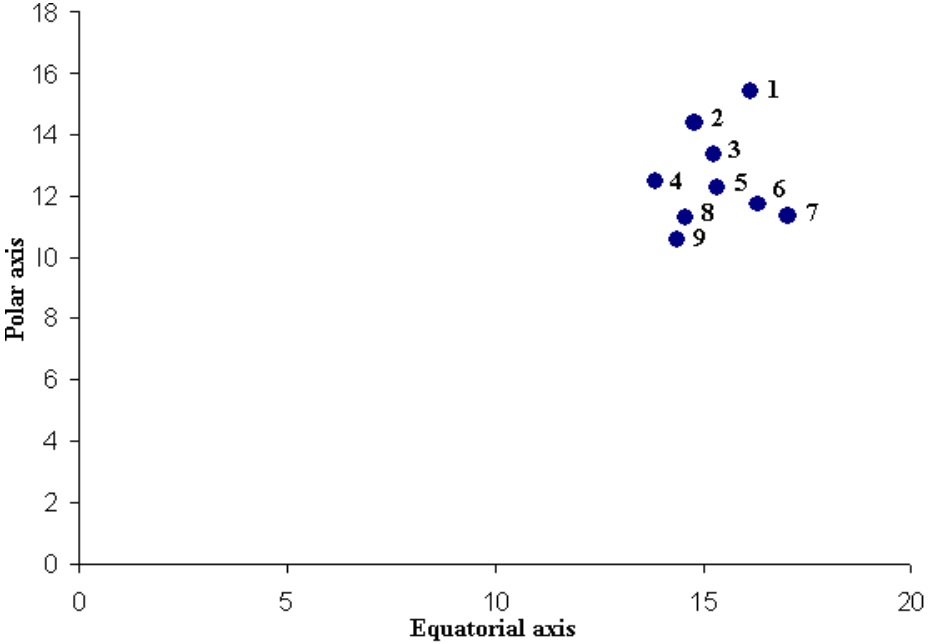


Fig. 6. Distribution of *Onosma* pollen taxa with respect to polar and equatorial axes (N).
1- *O. chlorotrichum,* 2- *O. bulbotrichum*, 3- *O. sintenisii,* 4- *O. orientale*, 5- *O. halophilum* 6- *O. heterophyllum,* 7- *O. ambigens,* 8- *O. oreodoxum,* 9- *O. bourgaei*.

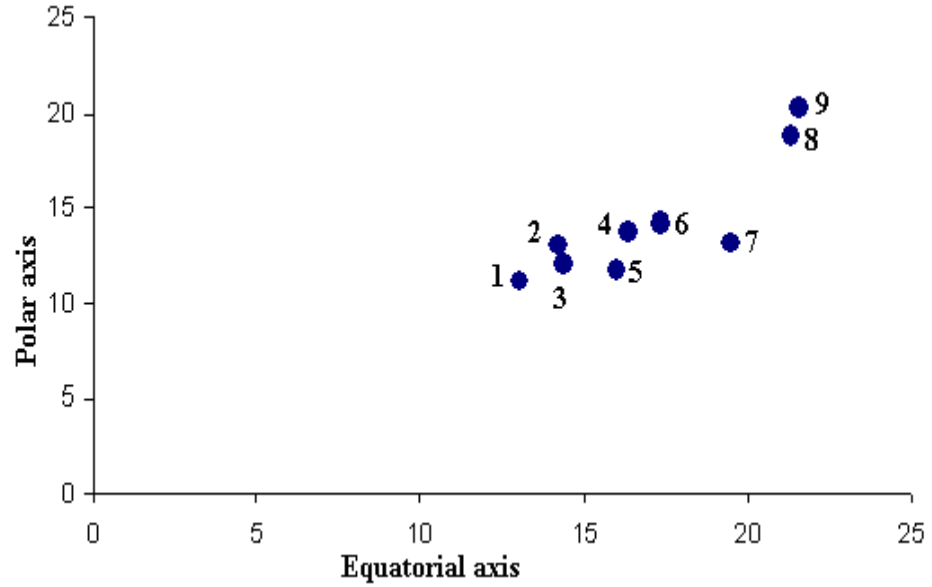


Fig. 7. Distribution of *Onosma* pollen taxa with respect to polar and equatorial axes (A). 1- *O. oreodoxum,* 2- *O. orientale*, 3- *O. heterophyllum,* 4- *O. chlorotrichum,* 5- *O. bourgaei,* 6- *O. halophilum,* 7- *O. ambigens,* 8- *O. sintenisii,* 9- *O. bulbotrichum*

Although taxonomically significant differences, exception of the pollen size, between the investigated *Onosma* taxa for micromorphology of pollen grains were not found by Maggi *et al*., (2008), our data clearly indicated that isopolar and heteropolar characters are taxonomically significant differences between the *Onosma* sections.

In Table 3 and Figs. 4 and 5 clearly showed that according to Wodehouse method, there are three different groups (sphaeroidea, subprolatae and prolata) on the basis of polar and equatorial axis among pollen grains belonging to the investigated taxa. Pollen grains are heteropolar in both *Haplotricha* and *Asterotricha* subsections. While pollen shape of Asterotricha subsection is prolate, pollen shape of Haplotricha subsection is sphaeroidea and subprolata.

Distribution of examined pollen of taxa with respect to polar and equatorial axes are given in Fig. 6 and Fig. 7.

Acknowledgements

I would like to thank Ihsan Akşit for his skilful technical assistance with the scanning electron microscopy and photography.

References

Al-Shehbaz, I.A. 1991. The Genera of Boraginaceae in the Southeastern United States: *Journal of The Arnold Arboretum*, 1: 1-169.

Baytop, T. 1994. *A Dictionary of Vernacular Names of Wild Plants of Turkey*. Ankara: Publication of the Türk Dil Kurumu (The Turkish Language Society) No. 578.

Binzet, R. and N. Orcan. 2003a. Morphological, anatomical and palynological study of *Onosma bracteosum* Hausskn. & Bornm. and *Onosma mutabile* Boiss. (Boraginaceae): *Phytologia Balcanica,* 9: 97-111.

Binzet, R. and N. Orcan. 2003b. Morphological and palynological studies on *Onosma roussaei* DC. and *Onosma giganteum* Lam. (Boraginaceae): *The Herb Journal of Systematic Botany,* 10: 57-76.

Binzet, R. and N. Orcan. 2007. A new species of *Onosma* L. (Boraginaceae) From Southern Turkey: *A journal for Botanical Nomenculature,* 17: 8-10.

Binzet, R. and N. Orcan. 2009. Anatomical and Palynological investigations on endemic *Onosma mersinana* Riedl, Binzet & Orcan, *Pakistan Journal of Botany,* 41: 503-510.

Clarke, G.C.S. 1977. *The Nortwest European Pollen Flora*, 10. Boraginaceae: *Review of Paleobotany and Palynology,* 24: 50-101.

Davis, P.H., R.R. Mill and K. Tan. 1988. *Flora of Turkey and the East Aegean Islands*, Vol. 10. University press, Edinburgh, pp. 184-185.

Diez, M.J. 1984. Contribucion al Atlas Polinico de Andalucia Occidental, I. Boraginaceae: *Lagascalia,* 13: 147-171.

Diez, M.J. and B. Valdes. 1991. Pollen morphology of the tribes Eritrichieae and *Cynoglosseae* (Boraginaceae) in the Iberian Peninsula nd its taxonomic significance: *Botanical Journal of The Linnean Society,* 101: 49-66.

El-Shazly, A., A. Abdel-Ghani and M. Wink. 2003. Pyrrolizidine Alkaloids from *Onosma arenaria* (Boraginaceae): *Biochemical Systematics and Ecology,* 31: 477-485.

Erdtman, G. 1952. *Pollen Morphology and Plant Taxonomy*, First Publ. 1952 Upsala.

Faegri, K. and J. Iversen. 1964. *Textbook of Pollen Analysis*, Munksgaard, Copenhagen, pp. 263.

Huynh, K.L. 1971. Le Pollen de Genera *Arnebia* Forssk. Et du genre, *Macromedia* DC. (Boraginaceae), et la position taxonomique particuliere du *M. echioides* (l.)Boiss. *Candollea* 26: 165-171.

Huynh, K.L. 1972. The original position of the generative nucleus in the pollen tetrads of *Agropyron*, *Jtea, Linanthes* and *Onosma* and its phylogenetic significance in the Angiosperms. *Grana,* 12: 105-112.

Khajuria, R.K. and S.M. Jain. 1993. Two new napthhoquinones from the roots of *Onosma hispidum*: *Indian Journal of Chemistry,* 32B: 390-391.

Lee, S. 1979. A factor analysis study of the functional significance of Angiosperm pollen. *Systematic Botany,* 3: 1-19.

Maggi, F., V. Kolarcık and P. Martonfi. 2008. Palynological analysis of five selected *Onosma* taxa: *Biologia,* 63: 183-186.

Marticorena, P.C. 1968. Granos de pollen de plantas chilenas, Gayana, Instituto Central de Biologia: *Botanica,* 17: 36-60.

Naz, S., S. Ahmad, S. Ajaz Rasool, S. Asad Sayeed and R. Siddiqi. 2006. Antibacterial activity directed isolation of compounds from *Onosma hispidum*: *Microbiological Research,* 161: 43-48.

Ning, J.C., Y.Z. Xi and Y.L. Zhang. 1995. A comparative palynological Study on the *Maharanga* and *Onosma* (Boraginaceae): *Acta Phytotaxonomica Sinica,* 33: 52-57.

Nowicke, J.W. and J.E. Ridgway. 1973. Pollen studies in the genus *Cordia* (Boraginaceae): *American Journal of Botany,* 60: 584-591.

Nowicke, J.W. and J.J. Skvarla. 1974. A palynological investigation of the genus *Tournefortia* (Boraginaceae): *American Journal of Botany*, 61: 1021-1036.

Özgen, U., Y. Houghton, Y. Ogundipe and M. Coşkun. 2003. Antioxidant and antimicrobial activities of *Onosma argentatum* and *Rubia peregrine*: *Fitoterapia,* 74: 682-685.

Öztürk, M. and H. Özçelik. 1991. *Benefical Plants of East Anatolia*. SİSKAV, Ankara.

Pandey, K.K. 1971. Pollen Size and incompatibility in *Nicotiana* pp. 317-322. *Pollen: Development and Physiology*. (Ed.): J. Heslop-Harrison. Butterworths, London.

Perveen, A., U.S. Qureshi and M. Qaiser. 1995. Pollen flora of Pakistan-IV Boraginaceae: *Pakistan Journal of Botany,* 27: 327-360.

Punt, W., S. Blackmore, S. Nilson and A. Le Thomas. 1994. *Glossary of Pollen and Spore Terminology*. Utrecht, LPP Foundation.

Qureshi, U.S. and M. Qaiser. 1987. Palynological study of *Onosma* (Boraginaceae) From Pakistan: *Pakistan Journal of Botany,* 19: 99-105.

Riedl, H. 1978. "*Boraginaceae*" In: *Flora of Turkey and the East Aegean Islands.* (Ed.): P.H. Davis. University press, Edinburgh. Vol. 6. pp. 237-437.

Riedl, H., R. Binzet and N. Orcan. 2005. A new species of *Onosma* (Boraginaceae-Lithospermeae) from Southern Turkey: *Edinburgh Journal of Botany,* 61: 127-130.

Wodehouse, R.P. 1935. *Pollen Grains*.– McGraf-Hill, New York

Yıldırımlı, Ş. 2000. The chorology of the Turkish species of Boraginaceae family: *The Herb Journal of Systematic Botany,* 7: 257-272.

(Received for publication 8 March 2010)