

**POLLEN MORPHOLOGY OF *ONOSMA* L.
(BORAGINACEAE) TAXA DISTRIBUTED IN NE ANATOLIA
KUZEY-DOĞU ANADOLU'DA YAYILIŞ GÖSTEREN
ONOSMA L. (BORAGINACEAE) TAKSONLARININ POLEN
MORFOLOJISI**

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ABSTRACT:

Pollen morphology of seven endemic *Onosma* L. taxa (Boraginaceae) from Turkey was investigated by using light (LM) and scanning electron microscopy (SEM). As a result of LM and SEM observations, it was found that the pollen types are generally syncolporate, anisopolar, and circular in polar view and the thickness of peritectate exine is 0.93-1.15 µm. Exine ornamentation was found as generally granulate-perforate, psilate, verrucate in polar area, mostly granulate and verrucate in equatorial area. While pollen shape is prolate-spheroid in *O. circinnatum* H. Riedl, subprolate in the rest of the examined taxa. The results also indicated that pollen characters are effective in separating the examined taxa.

Key words: Light Microscopy, *Onosma*, Palynology, Scanning Electron Microscopy, Turkey

ÖZET:

Bu çalışmada ışık ve electron mikroskobu kullanılarak yedi (7) endemik *Onosma* taksonunun polen morfolojileri çalışılmıştır. Genellikle polen tipleri sinkolporat, anizopolar ve polar görünüşte yuvarlak ve ekzin kalınlıkları 0.93-1.15 µm arasında değişmektedir. Ekzin süslenmeleri

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genellikle granulat-perforat ve psilae polar kutupta ve ekvatoralde granulat ve verrukattır. *O. cirninnatum* polen şekli prolat-siferoid iken diğerleri subprolattır. Sonuçlar çalışılan taksonları ve cinsin diğer üyelerini ayırmada polen karakterlerinin etkin olduğunu göstermiştir.

Anahtar Kelimeler: Işık Mikroskobu, *Onosma*, Palinoloji, Taramalı Elektron Mikroskobu, Türkiye

1. INTRODUCTION

Onosma (Boraginaceae) is represented with 150 species in Asia and Europe (El-Shazly et al 2003). In Turkey, the genus *Onosma* has 103 taxa (97 species, four varieties, one hybrid species and 50 endemic species, one endemic variety) and the rate of endemism is 50% according to native species (Yıldırım 2000, Riedl et al., 2005, Turkmen 2006, Binzet and Orcan 2007). Most of the Turkish representatives are endemic to Turkey and many of them have been studied with respect to their bioactive chemical compounds because of the usages in the folk medicine (Ozgen et al. 2003; Ozgen et al. 2004). In recent years the genus *Onosma* has been the subject of anatomical (Azizian et al 2000; Akcin and Engin 2004), chemical (Mellidis et al. 1993) and karyological (Teppner 1981; Teppner 1988; Teppner and Tuzlaci 1994) studies that improved our understanding of the systematic of this genus. The presence or absence of stellate setae and their characteristics are widely used as major character in order to solve the taxonomic difficulties in the genus (Pignatti 1982). Binzet and Orcan (2003 a) investigated the anatomical and palynological characteristics of *O. roussaei* DC. and *O. giganteum* Lam. and stressed its taxonomic importance in the genus.

Erdtman (1969) reported that detailed pollen morphological studies are very important in the taxonomy of Boraginaceae. Pollen morphology, therefore, *Onosma* has great potential as a means of classification, and is frequently utilized to clarify taxonomic problems (Scheel et al. 1996).

There are many taxonomic problems in the *Onosma* genus, particularly in C. & S.E Europe and Turkey (Riedl 1978). *Onosma* species are not investigated well enough by a taxonomist that's why many taxonomic difficulties remain unsolved. Up to now there were very limited studies carried out on Turkish representatives in order

to solve their taxonomic confusion by means of palynological studies. Palynological data which are extremely sparse at present, may provide useful reference points in future studies. Thus the objective of this study for determining the value of palynological features among *Onosma* taxa distributed in NE Anatolia.

2. MATERIAL AND METHODS

Specimens: Plants were collected between 2003 and 2006 from Northeast Anatolia and dried according to standard herbarium techniques. The vouchers are stored in the Herbarium of Karadeniz Technical University, Department of Biology (KTUB).

Palynological study the pollen grains at light microscopy analysis were treated with the standard method described by Erdtman (1952), and the observations were made with a microscope under 100x objectives with 10x eye piece. For SEM studies, the untreated pollen grains were mounted onto a metallic stub with double-sided adhesive tape and coated with gold in a sputtering chamber with restriction to 150A. The SEM examination was carried out using a Jeol-JSM 6060 scanning electron microscope. The size of the pollen of each species was measured based on 30 pollen grains from each specimen and described in terms of the longest, shortest, and average length and width, respectively. Pollen terminology follows mainly Punt et al (2007).

3. RESULTS

Pollen Characters and Figure List has been given below according to Flora of Turkey.

Table 1. Locality information of the examined *Onosma* taxa

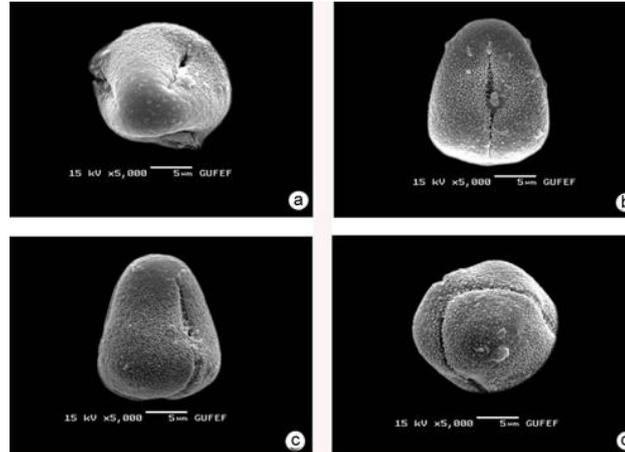
No	Taxa	Locality
1	<i>Onosma liparioides</i> DC.	A7 Bayburt: Kop Mount, 2700 m, Türkmen 090, KTUB
2	<i>Onosma isauricum</i> Boiss. & Heldr.	A8 Artvin: Yusufeli-Barhal Road, 680 m, Türkmen 070, KTUB
3	<i>Onosma bracteosum</i> Hausskn. & Bornm.	A7 Gümüşhane: Köse Mount, 1800 m, Türkmen 07, KTUB
4	<i>Onosma circinnatum</i> H. Riedl	A7 Gümüşhane: Köse Mount, Köse Barrage Bed, 1650 m, Türkmen 04, KTUB
5	<i>Onosma bornmuelleri</i> Hausskn.	A7 Trabzon: Maçka, 531 m, , Türkmen 01, KTUB
6	<i>Onosma armenum</i> DC.	A7 Gümüşhane: Tersun Mount, 2065 m, Türkmen 039, KTUB
7	<i>Onosma trapezunteum</i> Boiss. & Huet ex Hand.-Mazz.	A7 Trabzon: near Şinik, 150 m, Türkmen 030, KTUB

Table 2. Measurements and exine characteristics of pollen grains of *Onosma* L. taxa

Taxon	Pollen shape	Polar axis (μm)			Equatorial diameter (μm)			Exine (μm)		
		M	S	Var.	M	S	Var.	M	S	Var.
<i>O. liparioides</i>	Subprolate	20.62	± 1.03	19.00-22.80	17.58	± 0.96	16.15-19.00	0.96	± 0.10	0.85-1.15
<i>O. isauricum</i>	Subprolate	22.61	± 0.84	20.90-23.75	17.83	± 0.59	17.10-19.00	0.92	± 0.11	0.85-1.15
<i>O. bracteosum</i>	Subprolate	24.29	± 0.92	22.80-25.65	21.44	± 0.89	19.95-22.80	0.97	± 0.11	0.85-1.15
<i>O. circinnatum</i>	Prolate-spheroid	25.43	± 0.89	23.75-26.60	23.24	± 0.92	21.85-24.70	1.11	± 0.08	0.95-1.15
<i>O. bornmuelleri</i>	Subprolate	25.08	± 1.05	22.80-26.60	21.47	± 0.81	19.95-22.80	1.05	± 0.07	0.95-1.10
<i>O. armenum</i>	Subprolate	20.59	± 1.04	18.05-22.80	16.94	± 1.00	15.20-19.00	1.04	± 0.07	0.95-1.10
<i>O. trapezunteum</i>	Subprolate	25.65	± 0.86	23.75-26.60	21.95	± 1.07	19.95-23.75	1.09	± 0.08	0.95-1.15

***Onosma liparioides* DC. (Fig. 1a-d)**

The pollen grains subprolate in equatorial view, polar axis 20.62 μm , equatorial axis 17.58 μm . Circular in polar view, 18.46 μm in diameter. Colpi quietly long and thin, 13.36 μm x 2.40 μm ; pore 3.26 μm x 4.07 μm . Ornamentation in polar area perforate, in equatorial area granulate; exine 0.97 μm thick.

**Figure 1.** Pollen micrographs of *O. liparioides*; a-Polar view (SEM x5000), b-Equatorial view (SEM x5000), c-Aperture (SEM x5000), d- Apocolpium (SEM x5000)

***Onosma isauricum* Boiss. & Heldr. (Fig. 2a-d)**

The pollen grains subprolate in equatorial view, polar axis 22.61 μm , equatorial axis 17.83 μm . Circular in polar view, 18.53 μm in diameter. Colpi quietly long and thin, 15.80 μm x 3.74 μm ; pore 4.00 μm x 4.45 μm . Ornamentation in polar and equatorial area granulate; exine 0.93 μm thick.

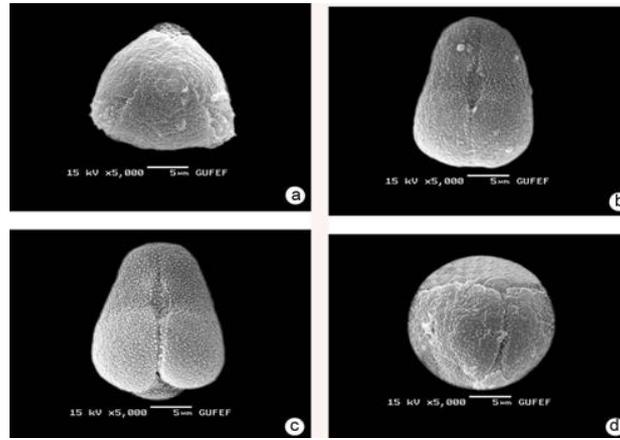


Figure 2. Pollen micrographs of *O. isauricum* a-Polar view (SEM x5000), b-Equatorial view (SEM x5000), c-Aperture (SEM x5000), d-Apocolpium (SEM x8500)

***Onosma bracteosum* Hausskn. & Bornm. (Fig. 3a-c)**

The pollen grains subprolate in equatorial view, polar axis 24.29 μm , equatorial axis 21.44 μm . Circular in polar view, 21.85 μm in diameter. Colpi quietly long and thin, 14.53 μm x 3.34 μm ; pore 3.34 μm x 4.37 μm . Ornamentation in polar and equatorial area granulate; exine 1.05 μm thick.

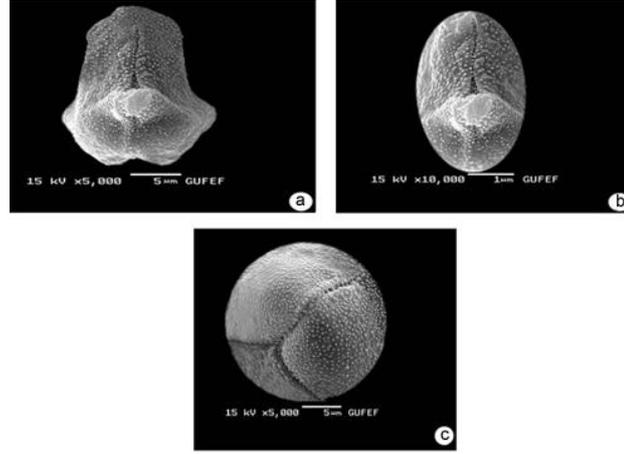


Figure 3. Pollen micrographs of *O. bracteosum*; a-Equatorial view (SEM x5000), b- Aperture (SEM x10000), c-Apocolpium (SEM x5000)

Onosma circinnatum H. Riedl (Fig. 4a-d)

The pollen grains prolate-sphaeroid in equatorial view, polar axis 25.43 μm , equatorial axis 23.24 μm . Circular in polar view, 23.78 μm in diameter. Colpi quietly long and thin, 15.07 μm x 4.13 μm ; pore 4.78 μm x 5.34 μm . Ornamentation in polar area psilate, in equatorial area granulate; exine 1.11 μm thick.

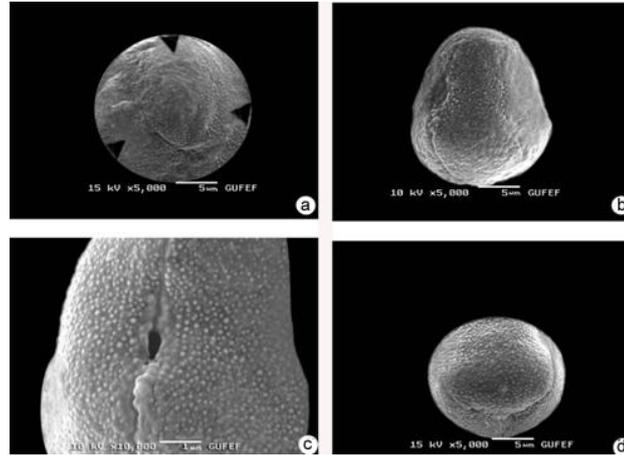


Figure 4. Pollen micrographs of *O. circinnatum*; a-Polar view (SEM x5000), b-Equatorial view (SEM x5000), c-Aperture (SEM x10000), d-Apocolpium (SEM x5000)

***Onosma bornmuelleri* Hauskn. (Fig. 5a-c)**

The pollen grains subprolate in equatorial view, polar axis 25.08 μm , equatorial axis 21.47 μm . Circular in polar view, 21.03 μm in diameter. Colpi quietly long and thin, 17.07 μm x 3.75 μm ; pore 3.82 μm x 4.29 μm . Ornamentation in polar area granulate, in equatorial area verrucate; exine 1.05 μm thick.

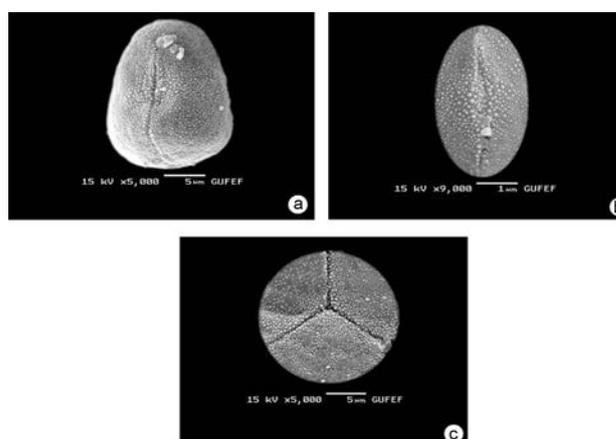


Figure 5. Pollen micrographs of *O. bornmuelleri*; a- Equatorial view (SEM x5000), b-Aperture (SEM x9000), c-Apocolpium (SEM x5000)

***Onosma armenum* DC. (Fig. 6a-d)**

The pollen grains subprolate in equatorial view, polar axis 20.59 μm , equatorial axis 16.94 μm . Circular in polar view, 19.51 μm in diameter. Colpi quietly long and thin, 11.91 μm x 3.13 μm ; pore 2.90 μm x 3.97 μm . Ornamentation in polar area granulate, in equatorial area verrucate; exine 1.10 μm thick.

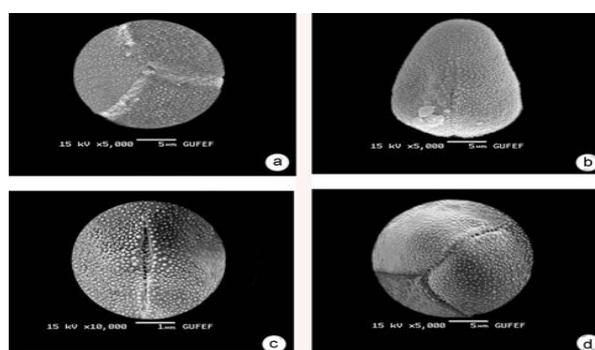


Figure 6. Pollen micrographs of *O. armenum*; a-Polar view (SEM x5000), b-Equatorial view (SEM x5000), c-Aperture (SEM x10000), d-Apocolpium (SEM x8500)

Onosma trapezunteum Boiss. & Huet ex Hand.-Mazz. (Fig. 7a-d)

The pollen grains subprolate in equatorial view, polar axis 25.65 μm , equatorial axis 21.95 μm . Circular in polar view, 21.22 μm in diameter. Colpi quietly long and thin, 16.40 μm x 3.93 μm ; pore 4.46 μm x 5.06 μm . Ornamentation in polar area granulate, in equatorial area verrucate; exine 1.15 μm thick.

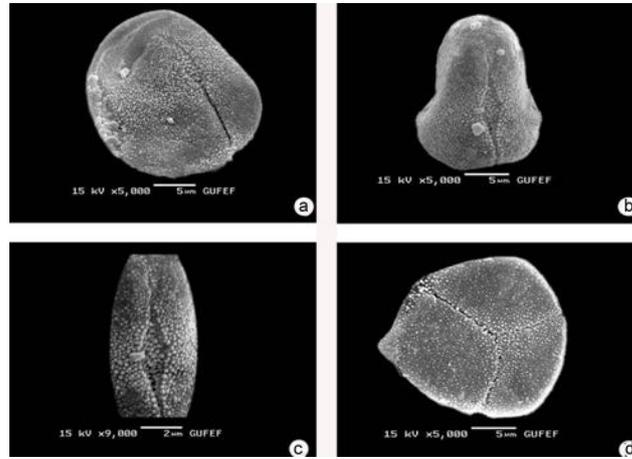


Figure 7. Pollen micrographs of *O. trapezunteum*; a-dry pollen grain (SEM x5000), b-Equatorial view (SEM x5000), c-aperture (SEM x9000), d-apocolpium (SEM x5000)

4. DISCUSSION

The ornamentation type, aperture number, shape and exine stratification are the most important palynological traits in solving taxonomic problems (Kuprianova 1967; Walker 1974a-b; Takhtajan 1980). Palynological data can give very useful information for the taxonomy of *Onosma* in the future and at present; such kinds of information are extremely sparse for the *Onosma* taxa especially distributed in Anatolia (Binzet 2003a, 2003b).

Although there is wide variation in Boraginaceae in terms of the pollen types, aperture number and ornamentation, the pollen grains are generally dispersed monad, zonocolporate and isopolar, polar axis is between 14-55 μm , and equatorial axis is between 11-43

μm (Bigazzi & Selvi 1996). But in our present study, all the examined taxa were observed anisopolar pollen grains (Fig 2b, Fig. 3b, etc).

There are three apertures with clearly three-sided in polar axis in genus *Echium* L., *Lobostemon* Lehm., *Alkanna* Tausch. but apertures in some *Onosma* are syncolporate one pole, they are always free at their ends in *Echium* L., *Lobostemon* Lehm. and *Echiostachys* Levyns (Retief and Van Wyk 1997). In the present study, it was found syncolporate at one pole in all the examined *Onosma* taxa (Fig. 2d, Fig. 3d, etc.)

In our study, Pollen grains of the examined taxa are generally anisopolar, syncolporate, exine structure pertectate and aperture membrane is scabrate or psilate. According to Erdtman (1952), average of the exine thickness was measured between 0.93 μm (*O. isauricum*) and 1.15 μm (*O. trapezunteum*). While the shortest colpus length was observed in *O. armenum* (11.91 μm), the longest was observed in *O. bornmuelleri* (17.07 μm). As seen in the pollen description, the smallest pore lengths were observed in *O. armenum* (2.9 μm) and the biggest is in *O. circinnatum* (4.78 μm).

Perforate ornamentation determined in polar area of *O. liparioides* and psilate ornamentation in *O. circinnatum* granulate in the rest of examined taxa *O. armenum*, *O. isauricum* etc. Prolate-spheroid pollen shape is observed in *O. circinnatum*, subprolate in the rest of the examined taxa e.g *O. liparioides* and *O. armenum*. In contrast, the pollen shape of *O. roussaei* was reported by Binzet (2003 a) as prolate (Wodehouse 1935) and suprolate in *O. giganteum* Lam. which is not included in this study. This means that the most common pollen shapes are suprolate among the examined taxa. Maggi et al. (2008) was reported pollen grains of all 5 species are small sized, 3-syncolporate, subprolate, heteropolar, with ovate equatorial outlines and circular to rounded triangular polar outlines; the tectum is microechinate. Hyunh (1971) was reported *Onosma helveticum*, as well as the other species of this genus, possesses a markedly heteropolar 3-colporate pollen. These pollen traits are in accordance with the findings reported by Qureshi and Qaiser (1987) and Perveen et al. (1995) for both *Onosma* and Boraginaceae.

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REFERENCES

- Akcin ÖE, Engin A. (2004). Endemik *Onosma bornmulleri* Hausskn.'nın Morfolojisi. Anatomisi ve Ekolojisi Üzerine Bir Araştırma. Ekoloji 51: 13-19.
- Azizian D, Khatamsaz M, Kasaian J. (2000). The Taxonomic significance of Leaf anatomy in the genus *Onosma* L. (Boraginaceae) in Iran. Iran. Journ. Bot. 2: 167-180.
- Bigazzi M, Selvi F. (1996). Pollen morphology in the Boragineae (Boraginaceae) in relation to taxonomy of the tribe. Grana 35: 138-153.
- Binzet R, Orcan N. (2003a). Morphological and palynological studies on *Onosma roussaei* DC. and *Onosma giganteum* Lam. (Boraginaceae). Ot Sistematiik Botanik Dergisi 10: 57-76.
- Binzet R, Orcan N. (2003b). Morphological, anatomical and palynological study of *Onosma bracteosum* Hausskn. & Bornm., and *Onosma mutabile* Boiss. (Boraginaceae). Phytologia Balcanica, 9(1): 97-111.
- Binzet, R. and N. Orcan. (2007). A new species of *Onosma* L. (Boraginaceae) From Southern Turkey. *A Journal for Botanical Nomenclature* 17: 8-10.
- El-Shazly A, Abdel-Ghani A, Wink M. (2003). Pyrrolizidine alkaloids from *Onosma arenaria* Waldst. and Kit. (Boraginaceae). Biochemical Systematics and Ecology 31: 477-485.
- Erdtman G. (1952). Pollen Morphology and Plant Taxonomy. Uppsala: Almqvist & Wiksells 133-134.
- Erdtman G. (1969). Handbook of Palynology. New York.
- Huynh KL (1971). The Original Position Of The Generative Nucleus In The Pollen Tetrads Of *Agropyron*, *Itea*, *Luafnanthes*, And *Onoshla*, And Its Phylogenetic Significance In The Angiosperms. Grana 12: 105-112
- Kuprianova A. (1967). Apertures of Pollen Grains and Their Evolution in Angiosperms. Paleobot. Palyn. 3: 73-80.

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- Maggi F, Kolarčik V, Mártonfi P. (2008). Palynological analysis of five selected *Onosma* taxa *Biologia* 63(2): 183-186
- Mellidis AS, Papageorgious VP, Kakkaolu E. (1993). Phenolic constituents from *Onosma heterophylla*. *Journal of Natural Products* 56: 949-952.
- Ozgen U, Houghton PJ, Ogundipe Y, Coskun M. (2003). The Antioxidant and Antimicrobial Activities of *Onosma argentatum* and *Rubia peregrina*. *Fitoterapia* 74: 682-685.
- Ozgen U, Coskun M, Kazaz C, Secen H. (2004). Naphtaquinones from the Roots of *Onosma argentatum* Hub.-Mor. (Boraginaceae). *Turkish Journal of Chemistry*. 28: 451-454.
- Perveen A, Shaheen U, Qureshi US, Qaiser M. (1995). Pollen Flora of Pakistan-IV. Boraginaceae. *Pak J Bot*. 27: 327-360.
- Pignatti S. (1982). *Flora D'Italia*. 2, Edagricole. Bologna.
- Punt W, Hoen PP, Blackmore S, Nilsson S & Le Thomas A. (2007). Glossary of Pollen and Spore Terminology. *Review of Palaeobotany and Palynology* 143: 1-81.
- Qureshi US, Qaiser M. (1987). Palynological study of *Onosma* (Boraginaceae) from Pakistan. *Pak. J. Bot*. 19: 99-105.
- Retief E, Van Wyk AE. (1997). Palynology of Southern African Boraginaceae; the genera *Lobostemon*, *Echiostachys* and *Echium*. *Grana* 36: 271-278.
- Riedl H. (1978). *Onosma* L. in: "Flora of Turkey and the East Aegean Island" ed. P. H. Davis. *Edinburg University Press*. 6: 326-378.
- Riedl H, Binzet R, Orcan N. (2005). A New species of *Onosma* (BORAGINACEAE-LITHOSPERMEAE) from Southern Turkey. *Edinburgh Journal of Botany* 61: 127-130.
- Scheel R, Ybert JP, Barth OM. (1996). Pollen Morphology of The Boraginaceae From Santa Catarina State (Southern Brazil), With Comments on the Taxonomy of the Family. *Grana* 35: 138-153
- Takhtajan AL. (1980). Outline of the Classification of Flowering Plants (Magnoliophyta) *Bot. Rev* 46: 225-359.
- Teppner H. (1981). Karyosystematik von *Onosma stellulatum*, *O. pygmaeum* und *O. leptanthum* (Boraginaceae). *Bot Jahrb. Syst* 102: 297-306.
- Teppner H. (1988). *Onosma kaheirei* spec. nova und *O. erectum* (Boraginaceae) aus Griechenland. *Phyton* 28: 115-131.
- Teppner H, Tuzlaci E. (1994). *Onosma propontica* Aznavour (Boraginaceae-Lithospermae). *Stapfia* 34: 77-83.

- Türkmen Z. (2006). Morphological and palynological features of *Onosma* L. (Boraginaceae) Taxa Distributed in Black Sea Region of Turkey. Unpublished D. Phill. Thesis Karadeniz Technical University. 148 p. Turkey.
- Walker JW. (1974a). Evolution of Exine Structure in the Pollen of Primitive Angiosperms. *Amer J Bot* 61: 891-902.
- Walker JW. (1974b). Aperture Evolution in the Pollen of Primitive Angiosperms. *Amer J Bot* 61: 1112-1137.
- Wodehouse RP. (1935). Pollen Grains. Their Structure, Identification and Significance in Science and Medicine Haffner Publish Company. New York and London. pp 574.
- Yıldırım, Ş. (2000). The chorology of the Turkish species of Boraginaceae Family, *The Herb Journal of Systematic Botany*, 7(2): 257-272.
