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Pollen morphology of *Cornus mas* L. and *Cornus sanguinea* L.

Nurgul Karlioglu Kilic [1*], Gamze Tuttu [2]

[1] Istanbul University, Faculty of Forestry, Department of Forest Botany, 34473, Istanbul, Turkey
[2] Cankiri Karatekin University, Faculty of Forestry, Department of Forest Engineering, 18200, Çankırı

[*] **Corresponding author e-mail (İletişim yazarı e-posta):** nurgulk@istanbul.edu.tr

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Abstract: The pollen morphology of 2 species of the genus *Cornus* L. distributed in Turkey was studied with light and scanning electron microscope. The pollen of genus *Cornus* is 3-colporate. Pollen of the *Cornus mas* is spheroidal and *Cornus sanguinea* is subprolate. Size varies of *Cornus mas* pollen with the polar axis ranging from 21.54 to 27.36 µm and equatorial axis from 21.31 to 26.72 µm. Size varies of *Cornus sanguinea* pollen with polar axis ranging from 54.41 to 71.69 µm and equatorial axis from 46.03 to 63.42 µm. The equatorial outline of *Cornus mas* pollen is triangular and *Cornus sanguinea* pollen is circular. The results showed that pollen of these two species can easily be identified as species level.

Keywords: Pollen morphology, *Cornus mas*, *Cornus sanguinea*

Cornus mas L. ve *Cornus sanguinea* L. Türlerinin Polen Morfolojisi

Özet: Türkiye'de yayılış gösteren *Cornus* L. cinsinin 2 türünün (*Cornus sanguinea* L. ve *Cornus mas* L.) polen morfolojisi ışık ve taramalı elektron mikroskobuyla çalışılmıştır. *Cornus* cinsinin polenleri trikolporat apertüre sahiptir. *Cornus mas*'ın polen şekli sferoid iken *Cornus sanguinea*'nın ise subprolat'dır. *Cornus mas* polenlerinin polar eksen uzunluğu 21.54-27.36 µm ve ekvatoral eksen uzunluğu 21.31-26.72 µm arasında değişmektedir. *Cornus sanguinea* polenlerinde ise polar eksen uzunluğu 54.41-71.69 µm ve ekvatoral eksen uzunluğu 46.03-63.42 µm arasında değişmektedir. *Cornus mas*'ın ekvatoral dış görünüşü üçgenimsi iken *Cornus sanguinea*'nın ise dairemsidir. Elde edilen sonuçlar bu iki türün polenlerinin kolaylıkla ayırt edilebileceğini göstermiştir.

Anahtar Kelimeler: Polen morfolojisi, *Cornus mas, Cornus sanguinea*

1.INTRODUCTION

Cornaceae family has 7 genera and 110 species, most of which can be found in East and Southeast Asia, and growing in temperate and humid regions of the north hemisphere (Kubitzki, 2004; Heywood et al., 2007). Many *Cornus* (dogwood) species are cultivated for their edible fruits or as ornamental plant for their spectacular flower, fruit and leaves. Some species are also utilized to create a decorative effect in the winter season because of their carmine or yellow trunk and branches (Karlioglu, 2014). In our country, there are 2 *Cornus* species which can grow up naturally. These are *C. mas* and *C. sanguinea* (Chamberlain, 1972; Güner et al., 2012).

Although several researches have been conducted about the genus *Cornus* in the field of systematic botany (Chamberlain, 1972; Krüssmann, 1976; Kayacik, 1982; Karlioglu, 2014), there have been mostly studies on the pollen morphology of Cornaceae in the field of palynology (Erdtman, 1952; Chao, 1954; Moore and Webb, 1978; Ferguson, 1966; Perveen and Qaiser, 2002). The pollen shape of Cornaceae changes between spheroidal and prolate (Erdtman, 1952; Stafford and Heath, 1991; Perveen and Qaiser, 2002), and its

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aperture type is tricolporate (Chao, 1954; Wodehouse, 1959). Nevertheless, the number of the measurements proving the difference of the morphology of the pollen in these researches is quite insufficient. Aytug et al. (1971) and Aytug and Merev (2002) have ascertained that *Cornus mas*'s pollen is tricolporate and they have carried out some palynological measurements on modern and fossil pollen grains. Sorkun (2008) has pointed out that only the diameter of tricolporate and colpus of *Cornus mas*'s pollen is bigger than the diameter of its pore. Also, Mert (2009) investigated the pollen length, pollen width, intine and exine thickness of six cornelian cherry (*Cornus mas* L.) cultivars. However many palynological studies have been made about especially Cornaceae, there has been no detailed and comparative study about the pollen morphology of 2 species of *Cornus* which grows naturally in Turkey.

Nowadays, the methods to categorize the plants used by the systematic botanists are mostly based on the morphological characters. The data from pollen morphology can be utilized to detect and categorize the plants generally on family and genera basis while sometimes there can be significant differences pointed out on species basis (Bradley, 1999). A similar study was performed by Akkemik (1995) and has demonstrated the possibility of the discrimination of *Carpinus orientalis* and *Carpinus betulus* in terms of pollen morphology. Hereby, it is possible to discriminate the species in the pollen analysis of the same plant species. In this context, this study aims to present differences of the pollen grains and showing the possibility of the discrimination of the species in the same genus by analyzing the pollen morphology of 2 *Cornus* species differing in their inflorescences color, size and blooming time with the help of light and scanning electron microscope in detail.

2. MATERIAL AND METHOD

The research materials are the pollen grains of *C. mas* picked from Çankırı- Korubaşı Hill neighborhood and the pollen grains of *C. sanguinea* picked from Belgrad Forest. The flower color, inflorescences features (Figure 1) and flowering time are quite different for 2 species of dogwood in this study.

Figure 1. The flowers of *Cornus mas* (a) and *Cornus sanguinea* (b)
Şekil 1. *Cornus mas* (a) ve *Cornus sanguinea*'nın (b) çiçekleri

The pollen grains have been brought to Istanbul University Faculty of Forestry Forest Botany Department Palynology Laboratory and prepared with pollen preparation according to the Wodehouse method (Wodehouse, 1959). The measurement and diagnosis of the number of the pollen grains in the pollen preparations are performed by using x40, x100 immersion objective and 10x ocular in the computer assisted Lecia DM750 light microscope. With the light microscope, at least 50 measurements are performed for each pollen feature (P: Polar axis, E: Equatorial axis, Clg: Length of colpus, Clt: Width of colpus, Plg: Length of porus, Plt: Width of porus, t: Apocolpium and Ex: Exine thickness). A part of this pollen is kept in the fridge in +4˚C for the measurement with Scanning Electron Microscope (SEM). The size of the pollen, aperture type and the features of ornamentation are studied in detail by taking photos with both SEM and light microscope.

3. RESULTS AND DISCUSSION

The pollen grains of *Cornus* species have tricolporate aperture. While the shape of *Cornus mas*'s pollen is spheroidal, *Cornus sanguinea*'s is subprolate (Figure / Şekil 2; Table / Tablo 1). The polar axis of *Cornus mas*'s pollen grains ranges from 21.54 to 27.36 µm and their equatorial axis ranges from 21.31 to 26.72 µm. The polar axis of *Cornus sanguinea*'s pollen grains ranges from 54.41 to 71.69 µm and their equatorial axis ranges from 46.03 to 63.42 µm (Table 1).

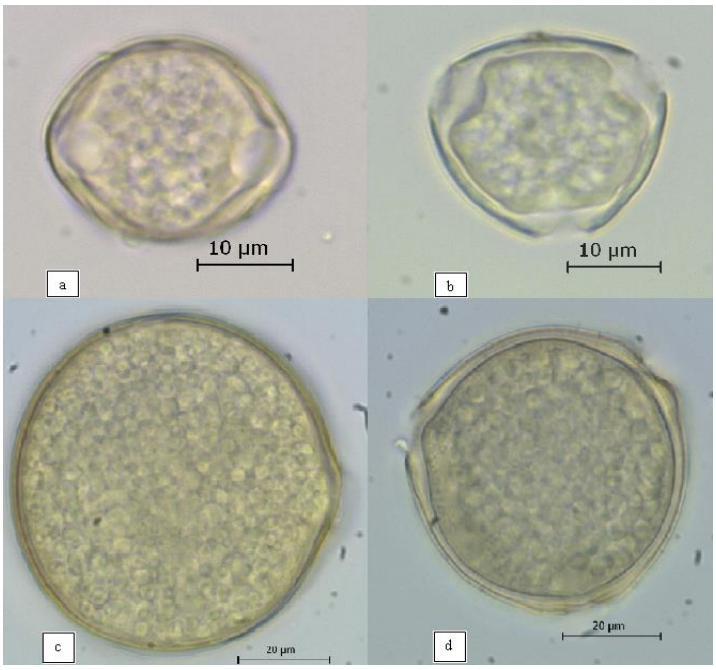

Figure 2. The equatorial and polar view of *Cornus mas* (a, b) and *Cornus sanguinea* (c, d)
Şekil 2. *Cornus mas* polenlerinin ekvatoral (a) ve polar (b) görünüşü ile *Cornus sanguinea* polenlerinin ekvatoral (c) ve polar (d) görünüşü

Table 1. The polar and equatorial axis, length and width of colpus and porus, apocolpium and exine thickness of *C. mas* and *C. sanguinea*
Tablo 1. *C. mas* ve *C. sanguinea* polenlerinin polar eksen ve ekvatoral eksen uzunlukları, kolpus uzunluğu ve genişliği, por uzunluğu ve genişliği, polar üçgenin bir kenarı ve ekzin kalınlığı

Plant species		***Cornus mas***	***Cornus sanguinea***
Polar axis (P) (µm)		(21.54-27.36)	(54.41-71.69)
Equatorial axis (E) (µm)		(21.31-26.72)	(46.03-63.42)
P/E rate ve pollen shape		0,99 (sferoidal)	1,15 (subprolate)
Aperture		Tricolporate	Tricolporate
Ornamentation		Granulate	Granulate
Colpus (C) (µm)	**Clg**	(12.34-18.82)	(41.53-61.86)
	Clt	(3.72-6.19)	(7.32-14.23)
Porus(P) (µm)	**Plg**	(4.29-8.83)	(7.96-12.80)
	Plt	(3.72-6.19)	(7.32-14.23)
Apocolpium (t) (µm)		(3.97-5.85)	(8.83-15.70)
Exine thickness (Ex) (µm)		(0.99-1.77)	(1.30-3.49)

The colpus length (Clg), colpus width (Clt), porus length (Plg), porus width (Plt), apocolpium (t) and exine thickness (Ex) of both the pollen of two species have been measured and found that they are different (Table / Tablo 1; Figure / Şekil 3). The colpus length of *C. mas*'s pollen grains ranges from 12.34 to 18.82 µm and their colpus width ranges from 3.72 to 6.19 µm. On the contrary, the colpus length of *C. sanguinea*'s pollen grains ranges from 41.53 to 61.86 µm and their colpus width ranges from 7.32 to 14.23 µm. The porus length of *C. mas*'s pollen grains ranges from 4.29 to 8.83 µm and their porus width ranges from 3.72-6.19 µm. However, the porus length of *Cornus sanguinea*'s pollen grains ranges from 7.96 to 12.80 µm and their colpus width ranges from 7.32 to 14.23 µm (Table 1; Figure 3).

Figure 3. The equatorial and polar view of *C. mas* (a, b) and *C. sanguinea* (c, d) in Scanning Electron Microscope
Şekil 3. *C. mas* ve *C. sanguinea* polenlerinin SEM'de ekvatoral ve polar görünüşü

Moreover, when both of the plant species' pollen ornamentation is analyzed with SEM, it has been detected that it is granulate and the aperture type is tricolporate (Figure 4).

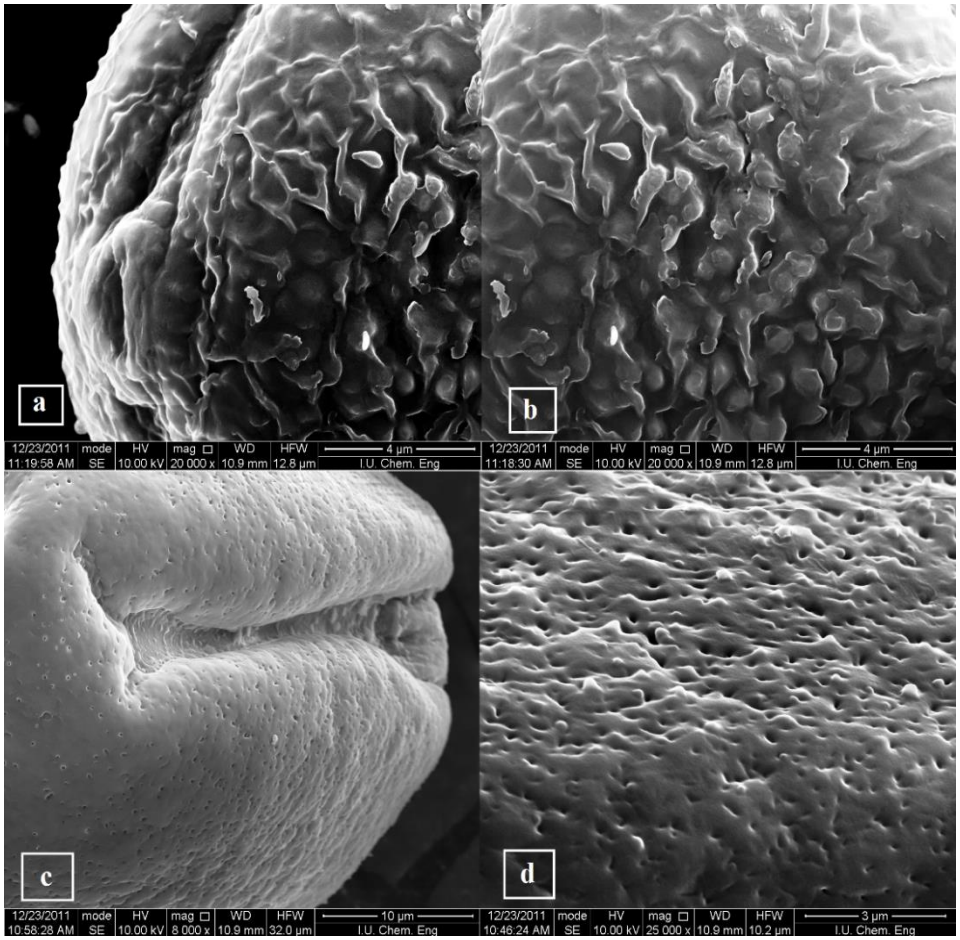

Figure 4. The aperture type and sculpture forms of *C. mas* (a, b) and *C. sanguinea* (c, d) in SEM
Şekil 4. *C. mas* ve *C. sanguinea* polenlerinin SEM'de apertür tipi ve skulptur formları

The apocolpium (t) of *Cornus mas*'s pollen grains ranges from 3.97 to 5.85 µm and their exine thickness (Ex) ranges from 0.99 to 1.77 µm. On the contrary, the apocolpium of *C. sanguinea*'s pollen grains ranges between 8.83 and 15.70 µm, and their exine thickness ranges from 1.30 to 3.49 µm (Table 1). There is a clear difference between these features.

Whether there is a considerable difference between two species or not in terms of the variables P, E, Clg, Clt, Plg, Plt, t, and exine thickness has been examined by using t-test which compares independent samples. The results of t-test reveals the considerable differences between two species on the level of significance p=0,001 in terms of all those variables (Table 2).

Table 2. The results of t-test for independent samples (P: Polar axis, E: Equatorial axis, Clg: The length of colpus, Clt: The width of colpus, Plg: The length of porus, Plt: The width of porus, t: Apocolpium and Ex: Exine thickness)
Tablo 2. Bağımsız örnekler için t- testi sonuçları (P: Polar eksen uzunluğu; E: Ekvatoral eksen uzunluğu; Clg: kolpus uzunluğu; Clt: kolpus genişliği; Plg: por uzunluğu, Plt: por genişliği, t: polar üçgenin bir kenarı ve Ex: Ekzin kalınlığı)

	P	**E**	**Clg**	**Clt**	**Plg**	**Plt**	**t**	**Ex**
t- value	**63,042*****	**56,524*****	**52,495*****	**22,859*****	**17,088*****	**22,859*****	**30,369*****	**17,141*****

*** P=0,001 it shows significance level

Many plants can be distinguished by pollen morphology with the pollen diagnosis keys (Erdtman, 1952; Erdtman, 1957; Faegri and Iversen, 1964; Iwanami et al., 1988; Moore et al., 1991; Hesse et al., 2009) formed on genera level. This study has shown that the identification of 2 dogwoods which grows naturally in our country would be in the species level by using palynological parameters.

According to Aytug et al. (1971), the length of the polar axis is (P): 62.42 µm and equatorial axis length (E) is 56.22 µm for the pollen grains of *C. mas*. The colpus length (Clg) for fresh pollen grains of *C. mas* has not been measured. The colpus width (Clt) is 11.18 µm, the porus length (Plg) is 9.30 µm and porus width is (Plt) 11.18 µm. The apocolpium (t) is 11.74 µm and exine thickness (Ex) is detected as 1.17 µm. The pollen type is sphaeroidae and the sculpture is detected as granula. When all these measurement results are considered, there is a sampling error for *C. mas*' pollen grains. According to the data of pollen morphology obtained from Aytug et al. (1971), their results should belong to the *C. sanguinea* because polar and equatorial axis lengths of its pollen grains are significantly bigger than those of *C. mas*.
Mert (2009) also described the pollen length, pollen width, intine and exine thickness of pollen grains of six cornelian cherry (*C. mas*) cultivars. The pollen grain length and width for the studied cultivars ranged from 23.63 to 25.13 µm and 24.25 to 27.13 µm respectively. In this study, pollen shapes determined as oblate spheroidal and prolate spheroidal. When (Mert, 2009) pollen grain results compared with our study, only pollen length values found similar.

Moreover, some pollen characters of Cornaceae was investigated by Erdtman (1952), Chao (1954), Moore et al. (1991), and it is found similar to the pollen morphology data of *C. mas* in our research.

4. CONCLUSIONS

A considerable difference between *C. mas* and *C. sanguinea* was found in terms of the variables (P, E, Clg, Clt, Plg, Plt, t and Ex) on the level of significance p=0,001 by using t-test which compares independent samples. The polar and equatorial axis, the length of colpus and porus, apocolpium and exine thickness of *C. mas* and *C. sanguinea*' pollen grains are quite different from each other. *C. sanguinea* pollen size is considerably bigger than *C. mas*. Aperture type is tricolporate is the same for both of them while pollen shape is spheroidal for *C. mas* unlike the subprolate shape of *C. sanguinea*. Both of them have same pollen ornamentation as granulate.

Thus, it is possible to distinguish the pollen of *C. mas* and *C. sanguinea* on the species level by using with this detailed pollen morphology study.

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