## MORPHOLOGY AND ANATOMY OF THREE SUBSP. OF CROCUS SPECIOSUS BIEB.

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## Abstract

Moprhology and anatomy of *Crocus speciosus* Bieb. subsp. *speciosus*, *C. speciosus* Bieb. subsp. *ilgazensis*, *C. speciosus* subsp. *xantholaimos* were done. Two of them (subsp. *ilgazensis* and *xantholaimos*) are endemic to small areas of Turkey. The subsp. *xantholaimos* has flowers with tube stained yellow. The subsp. *ilgazensis* has a corm splitting into vertical fibres. These properties are characteristic for the two subspecies investigated. Cross-sections of root and aerial stem of three subspecies were examined and characterized. A key to the identification of the three taxa, based solely on anatomical features is provided here.

#### Introduction

The genus *Crocus* is represented by about 80 species in the world, and in Turkey there are 37 species (Güner *et al.* 2000). The original saffron is being obtained from *C. sativus* L. since ancient times. In addition to this species a large number of *Crocus* species were brought into cultivation (Brigton *et al.* 1980). The three subsp. of *C. speciosus* investigated during this study are autumn-flowering species (Fig. 1). Autumnal *Crocus* species have been popular for about 150 years and have several cultivars. The corms of the investigated *Crocus* subspecies, that flowered during autumn are eaten raw or cooked in ash after gathering from underground during spring in Turkey. People in some regions of Anatolia have some traditional celebrations by making "çi dem pilavı" (Crocus pilaf). The leaves of these plants are also used in making a local cheese called "otlu peynir" (herbed cheese).

Recently, some researchers have reported that the extract of *Crocus* spp. has antitumor, antimutagenic and cytotoxic activities and inhibits nucleic acid synthesis in human malignant cells (Nair *et al.* 1991, Abdullaev *et al.* 2003, Loscutov *et al.* 2000, Fatehi *et al.* 2003). In the present study morphology and anatomy of the three subspecies of *C. speciosus* has been carried out.

## **Materials and Methods**

Plant samples were collected from natural populations between 1997 and 2002 (Table 1), and were preserved in the Herbarium of Ondokuz Mayıs University. Taxonomic description of the plant was made according to Mathew (1982) and Davis (1984). Anatomical works were carried out on fresh samples preserved in 70% alcohol. Paraffin embedding method was used for preparing cross sections of the tissues (Algan 1981).

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Subsp.	Locality	Collection date	Herbarium no.
C. speciosus subsp. speciosus	Samsun - Kocada, 1310 m	28.09.1997	
		24.11.1998	Özdemir 030
		25.09.1999	
	Trabzon - Zigana da 1, 2010 m	25.10.1997	Özdemir 031
	Trabzon - Karada, 2000 m	25.10.1997	Özdemir 032
C. speciosus subsp. ilgazensis	Sinop - Dranaz da 1, 1350 m	20.09.1997	Özdemir 033
		11.10.1997	
	Sinop - sfendiyar da 1,1455 m	20.09.1997	Özdemir 034
		24.10.1998	
C. speciosus subsp. xantholaimos	Amasya - Akda ,1800 m	27.09.1997	Özdemir 035
		18.10.1998	
	Çankırı - Ilgaz da 1, 1850 m	18.10.1997	Özdemir 036

Table 1. Information on three investigated subspecies of Crocus speciosus L.

## **Results and Discussion**

## Morphological features

*C. speciosus* subsp. *speciosus* Mathew: Flowers solitary, lilac coloured and veined with dark lilac. Perianth segments lanceolate, 1-5.5 cm in length. Perianth tube 6-19 cm in length, throat white. A small part of the tube remains underground. Style longer than stamens, deep orange, divided into many slender branches. Leaves ca. 2-3 in number, hysteranthous, about 2-4 mm in width, ca. 4-18 mm in length, green with a distinct median white stripe. Corm 7-22 mm in diameter, tunics membranous or subcoriaceous, splitting into horizontal rings at the base. Capsule elipsoid, 0.8-1 cm in length. Seeds subglobose, 1.5-2 mm in diameter and reddish-brown in color (Fig. 1).

*C. speciosus* subsp. *ilgazensis* Mathew: Flowers 1 - 2 in number, lilac-blue with 2 - 4.5 cm long, segments lanceolate. Perianth tube 2 - 11 cm in length, throat white. A small part of the tube remains underground. Style shorter than stamens, coloured and is divided into 6 - 8 branches. Leaves 5 - 6 in number, hysteranthous and about 8 - 11 cm in length, 0.1 - 0.2 cm in width, dark-green with a distinct median white stripe. Corm-tunics membranous, splitting into vertical fibres without distinct horizontal rings at the base. Capsule 0.8 - 1 cm in length. Seeds 1.0 -1.5 mm in diameter, dark reddish-brown in color (Fig. 2).

*C. speciosus* subsp. *xantholaimos* Mathew: Flowers solitary, lilac or dark lilac with 1.5 - 8.0 cm long segments, oblong in shape. Perianth tube 5-13 cm in length, throat deep yellow. Style shorter than stamens, many-branched. Leaves 3-4-5 in number, hysteranthous,  $4 - 20 \times 0.1 - 0.2$  cm, dark-green coloured with a distinct median white stripe. Corm-tunics membranous and splitting into horizontal rings at the base. Capsule 0.6 - 1.0 cm in length. Seeds 0.5 - 1.5 mm in diameter (Fig. 3).

## Anatomical features

#### C. speciosus subsp. speciosus Mathew

*Root:* Epidermis single layered, prismatic and thin-walled. Cortex 5-6-layered, cells ovoidal, 13-38  $\mu$ m. Endodermis single layered with casparian strip. Pericycle thin-walled. Metaxylem two in number on the median part of the vascular cylinder. Protosxylems are 8, reaching the pericycle (Fig. 4).

Stem: Stem is a corm. Epidermis single layered and isomorphic,  $8-10 \times 13-30 \,\mu\text{m}$  in size. Six metaxylems are at the middle of aerial stem. Small protoxylems are (11-14) are located in the peripheral part of the aerial stem (Fig. 5).



- Fig. 1. Habit sketch and morphological parts of *C. speciosus* subsp. *speciosus*. (a) flowered plant, (b) leafy plant, (c) corm, (d) perianth, (e) style, (f) stamen, (g&h) fruit, (i) ovary, (j) seed (Bars show 1 cm in a-f and 5 mm in g-i and 2 mm in j).
- Fig. 2. Habit sketch and morphological parts of *C.speciosus* subsp. *ilgazensis*. (a) flowered plant, (b) leafy plant, (c) corm, (d) perianth, (e) style, (f) stamen, (g&h) fruit, (i) ovary, (j) seed (Bars show 1cm in a-f and 5 mm in g,h,j and 2 mm in i).
- Fig. 3. Habit sketch and morphological parts of *C.speciosus* subsp. *xantholaimos*. (a) flowered plant, (b) leafy plant, (c) corm, (d) perianth, (e) style, (f) stamen, (g&h) fruit, (1) ovary, (j) seed (Bars show 1 cm in a-f,i and 5 mm in g,h and 2 mm in j).

#### C. speciosus subsp. ilgazensis Mathew

Root: Epidermis single-layered, cells irregularly different in size. Cortex 5-7-layered, parenchymatous, cells ovoidal, 15-30 µm in diameter. Endodermal cells single-layered. The wall thickenings of the endodermal cells are three sided. Pericycle single layered. Metaxylem single on the median part of the vascular cylinder, protoxylems are 4 in number (Fig. 6).

Stem: Epidermis single-layered, and its cells are nearly of the same size. Cortex cells 15-30  $\mu$ m in diameter, many of which posses starch grains. Vascular bundles present in the peripheral and central part of the stem, 6 big vascular bundles are located in the central part of the aerial stem. The small vascular bundles are 8-10 in number and are located in the periphery (Fig. 7).

#### C. speciosus subsp. xantholaimos Mathew

Root: Epidermis single-layered, thin-walled and its cells are nearly of the same size. Cortex 3-6-layered, 10-30 µm in diamater. Endodermal cells with casparian strip. There are 2-4 metaxylem on the median part of the vascular cylinder (Fig. 8).

Stem: Epidermis single-layered and consists of cubical cells. Cortex cells 15-45 µm in diameter. Six big vascular bundles are located in the central part of the aerial stem and 4-7 small vascular bundles in the peripheral part (Fig. 9).

The morphological characters of three subspecies of C. speciosus were investigated with a view to evaluate their taxonomic value. The structure of corm tunic, the style, the color of perianth tube and perianth segments were considered to be useful for the purpose. Corm tunic of subsp. speciosus and xantholaimos are split into rings at the base, while subsp. ilgazensis has a corm tunic splitting into vertical fibres. Perianth tube of subsp. xantholaimos is colored yellow. Subsp. ilgazensis has a style divided into 6-8 expanded branches, while subsp. speciosus and xantholaimos have a style divided into many expanded branches.

In anatomical studies, it has been found that casparian strips appear in the walls of endodermal cells of subsp. speciosus and xantholaimos. Endodermal thickening is three-sided and towards the pericycle of root. The researchers have observed the same characteristics were also met with in the root of Crocus aerius Herb., Gladiolus atroviolaceus Boiss. and Crocus danfordiae Maw. (Özyurt 1978, Özdemir et al. 2004). The wall thickenings of the endodermal cells of subsp. *ilgazensis* were three-sided and towards the cortex. This type of endodermal cells are common in the roots of monocotyledones (Fahn 1982). The investigated three subsp. have vascular bundles in both central and peripheral part of the aerial stem. This feature has also been observed in the cross-section of the aerial stem of Crocus fleischeri and C. donfordiae (Özdemir et al. 2004). But the vascular bundles in C. aerius and C. pulchellus are located only in the central part of the aerial stem (Özyurt 1978, Özdemir and Akyol 2004). The bundles are arranged in two circles at the stem of subsp. speciosus and subsp. xantholaimos, while subsp. ilgazensis has also a single vascular bundle at the center of the aerial stem. Number of small vascular bundles in stem are 11-14 at subsp. speciosus, while that is 4-7 in subsp. xantholaimos. Evaluating the findings of the three subspecies, obtained from the present study, a key, based solely on anatomical features below has been provided here to identify the three infraspecific taxa.

1 Endodermal thickening three-sided and towards the pericycle of root; metaxylem 2-4; aerial stem vascular bundles arranged so as to form two circles

subsp. speciosus

2. Number of small vascular bundle in stem within corm 11-14

2. Number of small vascular bundle in stem within corm 4-7 subsp. xantholaimos

1 Endodermal thickening three-sided and towards the cortex of root; metaxylem single; aerial stem vascular bundles arranged so as to form three circles subsp. ilgazensis



Fig. 4. Cross-section of the root of *C. speciosus* subsp. *speciosus*. e. epidermis, k. cortex, en. endodermis, p. pericycle, f. phloem, m. metaxylem. (Camera lucida drawing on left and photmicrograph on right).

- Fig. 5. Cross-section of the root of *C.speciosus* subsp.*ilgazensis*. a. Camera lucida drawing, b. Photomicrograph). e. epidermis k. cortex en. endodermis p. pericycle f. phloem m. metaxylem.
- Fig. 6. Cross-section of the root of *C. speciosus* subsp.*xantholaimos* (photomicrographs). a. Camera lucida drawing, b. Photmicrograph. (e. epidermis k. cortex en. endodermis p. pericycle f. phloem m. metaxylem).



- Fig. 7. a. Cross-section of the aerial stem of *C. speciosus* subsp. *speciosus*. b. enlargement of the shown area of a. (e. epidermis, i. vascular bundle, k. cortex).
- Fig. 8. a. Cross-section of the aerial stem of *C. speciosus* subsp. *ilgazensis* (Camera lucida drawing). b. enlargement of the shown area of a. (e. epidermis, i. vascular bundle, k. cortex).
- Fig.9. a. Cross-section of the aerial stem of *C. speciosus* subsp. *xantholaimos* (photomicrographs). b. enlargement of the shown area of a. (e. epidermis, i. vascular bundle, k. cortex).

Leaves of the autumn-flowering *Crocus* species remain dormant for a considerable time before emerging (Mathew and Brighton 1977). The investigated taxa were autumn-flowering. In the cross-sections of the corms collected in autumn, which do not bear any leaf, leaves remaining dormant were seen (Figs. 5,7,9). The leaves of subsp. *speciosus* and *ilgazensis* have a square central keel while the leaves of subsp. *xantholaimos* have a slightly triangular central keel. Rudall (1994) has also pointed out that the leaves of most *Crocus* species have a unique and distinctive shape comprising a square or rectangular central keel in cross-section.

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