# An anatomical and ecological study of some Crocus L. taxa (Iridaceae) from the west part of Turkey 

Fatih Satil*, Selami Selvi<br>Department of Biology, Science and Arts Faculty, Balıkesir University, 10145 Balıkesir, Turkey

A comparative anatomical and ecological study was made of nine Crocus taxa (C. gargaricus subsp. gargaricus, C. chrysanthus, C. biflorus subsp. nubigena, C. flavus subsp. dissectus, C. olivieri subsp. istanbulensis, C. candidus, C. pallasii subsp. pallasii, C. cancellatus subsp. mazziaricus, C. pulchellus) from the west part of Turkey. Leaf anatomy has been differentiated among the taxa. The lower surfaces of the arms and the sides of keels have projections only in C. biflorus subsp. nubigena. Anticlinal walls of epidermis in stomatal regions are generally sinuous, but sometimes straight. There are four large vascular bundles in the margin of the arms and on the corner of the keels. Sclerenchymatous tissue was observed in phloem and xylem poles of larger bundles in C. flavus subsp. dissectus, C. olivieri subsp. istanbulensis, C. candidus, C. pallasii subsp. pallasii, while it was observed only in the phloem pole in C. gargaricus subsp. gargaricus, C. chrysanthus, C. biflorus subsp. nubigena, C. cancellatus subsp. mazziaricus, C. pulchellus. All taxa grow on clayey-loamy and clayey soils, with $\mathrm{pH} 6.09-7.18$, with $3.1-5.7 \%$ of $\mathrm{CaCO}_{3}, 3.1-5.7 \%$ of organic matter, $2.5-67.7 \%$ of phosphorus, and $52.7-198.7 \%$ of potassium.

Key words: Anatomy, Crocus, ecology, leaf, Turkey.

## Introduction

There are 80 species of Crocus worldwide (Mabberly 1997), distributed mainly in the Mediterranean region. There are 63 taxa in Turkey. Thirty-one of these are endemic to Turkey (Mathew 1984, 2000, Davis et al. 1988). Systematic studies have been presented by Davis et al. (1988), Kerndorff and Pasche (1994, 1996a, 1996b, 1997), Mathew (1984, 1995, 2000), and PASCHE (1994a, 1994b), but there have been a few studies on the morphology and anatomy of Crocus species in Turkey (Özdemir et al. 2004, Özyurt 1978, Akan and Eker 2004, Akan et al. 2006).

The leaves of most Crocus species have a unique and distinctive shape in cross section. In the present study, detailed descriptions of the anatomical characteristics of leaves in nine Crocus species were studied in Turkey (Fig. 1), in order to provide more detailed descriptions. Five of these species are endemic to Turkey (Tab. 1).

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Fig. 1. Collection sites in Turkey for Crocus samples (1-C. gargaricus subsp. gargaricus, 2-C. chrysanthus, 3-C. biflorus subsp. nubigena, 4-C. flavus subsp. dissectus, 5-C. olivieri subsp. istanbulensis, 6-C. candidus, 7-C. pallasii subsp. pallasii, 8-C. cancellatus subsp. mazziaricus, 9-C. pulchellus).

Tab. 1. Collection data of studied Crocus taxa from Turkey.

|  | Collection data and collector number |
| :---: | :---: |
| Croccus gargaricus subsp. gargaricus (E) C. chrysanthus | B1 Balıkesir: Edremit, Kazdağ, Kartalçimen, 1700 m, 07.04.2005, FS1382. <br> B1 Balıkesir: Dinkçiler, 140 m, 15.02.2005, SV1011. <br> B2 Balıkesir: Sindırg1, Ulus mountain, $1400 \mathrm{~m}, 19.04 .2005$, SV1040. <br> C2 Muğla: Göktepe, $1600 \mathrm{~m}, 29.03 .2005$, FS1392. |
| C. biflorus subsp. nubigena (E) | B1 Balıkesir: Edremit, Kazdağ, 1400 m, 15.02.2005, FS1384. <br> B2 Balıkesir: Sindırgı, Kocabey village, $700 \mathrm{~m}, 01.02 .2005$, SV1008. <br> C2 Muğla: Göktepe, $1800 \mathrm{~m}, 29.03 .2005$, FS1402. |
| C. flavus subsp. dissectus (E) | B1 Balıkesir: Çağış village, $140 \mathrm{~m}, 20.02 .2005$, SV1014. <br> B2 Balıkesir: Dursunbey, Esentepe, 800 m, 24.03.2005, SV1032. <br> A1 Balıkesir: Manyas, Akçaova village, 140 m, 15.03.2004, FS1420. <br> A2 Balıkesir: Bandırma, Dedeoba village, 145 m, 17.03.2004, FS1415. <br> C2 Denizli: Kazıkbeli, 1130 m, 28.03.2005, FS1403. |
| C. olivieri subsp. istanbulensis (E) | B1 Balıkesir: Değirmen boğazı, $180 \mathrm{~m}, 04.03 .2005$, SV1019 |
| C. candidus (E) | B1 Çanakkale: Ezine to Truva, 200 m, 27.02.2005, FS1385. <br> B1 Balıkesir: Edremit, Ortaoba village, 350 m, 28.02.2005, SV1034. |
| C. pallasii subsp. pallasii | B1 Balıkesir: Savaştepe, Kozören village, 500 m, 14.12.2004, SV1006. C1 Aydın: 4. km from Selçuk to Kusadası, 40 m, 22.11.2005, FS1404 |
| C. cancellatus subsp. mazziaricus | B1 Balıkesir: Savastepe, Kozören village, 500 m, 14.12.2005, SV1007. C1 Muğla: 18. km Milas to Bodrum, 23.11.2005, FS14012 |
| C. pulchellus | B1 Balıkesir: Savaştepe, Kozören village, 500 m, 06.12.2004, SV1005 B1 Balıkesir: Edremit, Kazdağ, 1400 m, 14.11.2005, FS1410 <br> A2 Bursa: Uludağ, $1800 \mathrm{~m}, 15.11 .2005$, FS1411 |

## Materials and methods

Material used in this study was collected from different localities in Turkey (Fig. 1). Voucher specimens (Tab. 1) were deposited in the Balıkesir University, Department of Biology.

Fresh plants were fixed in $70 \%$ alcohol. Developed leaves of species from fully flowered plants were used in the anatomical study. Five samples were taken from each specimen for transverse section. Transverse sections of leaves were made manually. Tissues were stained with Floroglusin +HCl and embedded in glycerine jelly (BАчтор 1972). An Olympus BX50 phase contrast microscope with drawing tube was used in anatomical studies.

The physical and chemical analyses of the soil samples were taken from the sites presented in Fig. 1. Texture, pH , calcium carbonate, total soluble salts and organic matter content were determined (Öztürk et al. 1997). K\% and $\mathrm{P} \%$ were determined by flame photometer and the ammonium molybdate-tin chloride method, respectively (KAÇAR 1996).

## Results

## Leaf anatomy

Leaves of Crocus taxa have a unique morphology in the cross-section with a central square or rectangular »keel«, and two lateral »arms« (Fig. 2A). There are projections in the lower surfaces of the arms and the sides of keels only in C. biflorus subsp. nubigena. This structure is absent in other taxa (Figs 3C, 4C).

Epidermis cells are either rectangular or elliptical. Anticlinal walls of epidermis in stomatal regions are generally sinuous (in C. gargaricus subsp. gargaricus, C. biflorus subsp. nubigena, C. flavus subsp. dissectus, C. olivieri subsp. istanbulensis, and C. candidus) but sometimes straight (in C. chrysanthus, C. pallasii subsp. pallasii, and C. cancellatus subsp. mazziaricus) and straight-sinuous in C. pulchellus (Fig. 2D). The upper epidermis cells are longer and more orbicular than the lower epidermis cells (Fig. 2C).

The cuticle of the upper epidermis is thicker than in the lower epidermis. Micropapillae are inconspicuous on the cuticle of C. gargaricus subsp. gargaricus (Tab. 2), but there are a conspicuous micropapillae on the cuticles of other 8 taxa (Figs. 2B, 4).

Hair like papillae are generally present on prominent ridges over the largest vascular bundles, or at the keel corner of C. gargaricus subsp. gargaricus, C. flavus subsp. dissectus, C. olivieri subsp. istanbulensis, C. candidus, C. pallasii subsp. pallasii, C. cancellatus subsp. mazziaricus, and C. pulchellus (Fig. 3). Hair-like papillae are absent in C. chrysanthus and C. biflorus subsp. nubigena.

Stomata of all taxa are anomocytic and occur only on the lower surfaces. The stomata were mainly confined to the grooves in the recurved undersurface of the arms (Fig. 2B, 2D).

Mesophyll on the arms is differentiated into palisade and spongy parenchyma (Figs. 2B, 4). Palisade tissue is 2-layered (in C. chrysanthus, C. olivieri subsp. istanbulensis, C. candidus, C. pallasii subsp. pallasii, C. cancellatus subsp. mazziaricus, and C. pulchellus) or 1-2-layered (in C. gargaricus subsp. gargaricus, C. biflorus subsp. nubigena, and C. flavus subsp. dissectus). Spongy tissue is 2-3-layered in C. chrysanthus and C. flavus subsp. dissectus; 3-4-layered in C. biflorus subsp. nubigena, C. candidus, C. pallasii
subsp. pallasii, C. pulchellus, and 4-5-layered in C. gargaricus subsp. gargaricus; 3-5--layered in C. olivieri subsp. istanbulensis and C. cancellatus subsp. mazziaricus (Tab. 2). Spongy cells are round or oval. The central part of the keel consists of large thin walled parenchymatic cells.



C


D


Fig. 2. A-General drawing of transverse sections in Crocus leaves from Turkey, B-Cross-section of leaf (in C. olivieri subsp. istanbulensis), C-Surface view of upper epidermis (in C. olivieri subsp. istanbulensis), D-Surface view of lower epidermis in regions with stomata (a-C. chrysanthus, b-C. biflorus subsp. nubigena). k-kell, as-air space, vb-vascular bundle, mp--micropapillae, ue-upper epidermis, pp-palisade parenchyma, sp-spongy parenchyma, le--lower epidermis, st-stomata, sc-sclerenchyma, xy-xylem, ph-phloem. Scale bars: $\mathrm{A}=500$ $\mu \mathrm{m}, \mathrm{B}=50 \mu \mathrm{~m}, \mathrm{C}, \mathrm{D}=100 \mu \mathrm{~m}$.


Fig. 3. General drawing of transverse sections in Crocus leaves from Turkey. A) C. gargaricus subsp. gargaricus, B) C. chrysanthus, C) C. biflorus subsp. nubigena, D) C. flavus subsp. dissectus, E) C. olivieri subsp. istanbulensis, F) C. candidus, G) C. pallasii subsp. pallasii, H) C. cancellatus subsp. mazziaricus, I) C. pulchellus. Scale bars: $400 \mu \mathrm{~m}$.

Vascular bundles are usually around the lower surface of keel and extend along the arms. There are large and small vascular bundles in the mesophyll. Four large bundles are always present in keel corners and arm margins. Small vascular bundles extend in between arm margins and keel. Vascular bundles are collateral. There are sclerenchymatous cells as caps only at the phloem poles of larger bundles and smaller bundles in C. gargaricus subsp. gargaricus, C. chrysanthus, C. biflorus subsp. nubigena, C. cancellatus subsp. mazziaricus, and C. pulchellus. There are also sclerenchymatous cells in phloem poles and xylem poles of larger bundles in C. flavus subsp. dissectus, C. olivieri subsp. istanbulensis, C. candidus, and C. pallasii subsp. pallasii (Figs. 2B, 4).

Larger bundles extend to the epidermis as girds in C. chrysanthus, C. cancellatus subsp. mazziaricus, and C. pulchellus, but do not do so in other taxa (Figs. 3, 4B, H, I).

## Ecological characteristics

The investigated Crocus taxa grow on exposed limestone rocks, stony slopes, sparse woods, scrub, grassy places, alpine turf, and damp pastures, between sea level and 3000 m .
Tab. 2. The comparison of leaf anatomical characteristics.

|  | Micropapillae <br> in cuticle | Hair like <br> papillae | Anticlinal walls <br> of epidermis in <br> stomatal regions | Projecting in <br> arms and side <br> of keels | Palisade <br> layer | Spongy <br> layer | Larger bundles <br> to epidermis |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Croccus gargaricus subsp. gargaricus | inconspicuous | Present | sinuous | - | $1-2$ | $4-5$ | not extending |
| C. chrysanthus | conspicuous | - | straight | - | 2 | $2-3$ | extending |
| C. biflorus subsp. nubigena | conspicuous | - | sinuous | projecting | $1-2$ | $3-4$ | not extending |
| C. flavus subsp. dissectus | conspicuous | Present | sinuous | - | $1-2$ | $2-3$ | not extending |
| C. olivieri subsp. istanbulensis | conspicuous | Present | sinuous | - | 2 | $3-5$ | not extending |
| C. candidus | conspicuous | Present | sinuous | - | 2 | $3-4$ | not extending |
| C. pallasii subsp. pallasii | conspicuous | Present | straight | - | 2 | $3-4$ | not extending |
| C. cancellatus subsp. mazziaricus | conspicuous | Present | straight | - | 2 | $3-5$ | extending |
| C. pulchellus | conspicuous | Present | straight-sinuous | - | 2 | $3-4$ | extending |



Fig. 4. Transverse sections of Crocus leaves. A) C. gargaricus subsp. gargaricus, B) C. chrysanthus, C) C. biflorus subsp. nubigena, D) C. flavus subsp. dissectus, E) C. olivieri subsp. istanbulensis, F) C. candidus, G) C. pallasii subsp. pallasii, H) C. cancellatus subsp. mazziaricus, I) C. pulchellus. Scale bars: 100 mm .

Tab. 3. Soil characteristics of Crocus taxa localities.

|  | Texture | Total <br> soluble <br> salt (\%) | pH | $\mathrm{CaCO}_{3}$ <br> $\%$ | Org. <br> Matter <br> $(\%)$ | P <br> $\%$ | K <br> $\%$ |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Croccus gargaricus <br> subsp. gargaricus | clayey | 0.103 | 6.98 | 4.0 | 5.3 | 5.4 | 120.4 |
| C. chrysanthus | clayey-loamy | 0.041 | 6.25 | 0.8 | 3.1 | 2.5 | 52.7 |
| C. biflorus subsp. <br> nubigena | clayey-loamy | 0.093 | 6.74 | 2.4 | 4.9 | 7.6 | 147.5 |
| C. flavus subsp. <br> dissectus | clayey | 0.080 | 6.92 | 4.0 | 5.5 | 3.6 | 82.8 |
| C. olivieri subsp. <br> istanbulensis | clayey-loamy | 0.051 | 6.67 | 1.6 | 5.7 | 47.0 | 144.5 |
| C. candidus | clayey-loamy | 0.046 | 6.34 | 0.4 | 4.3 | 18.3 | 82.8 |
| C. pallasii subsp. <br> pallasii <br> C. cancellatus subsp. <br> mazziaricus | clayey-loamy | 0.066 | 7.18 | 20.8 | 4.0 | 2.9 | 78.3 |
| c. pulchellus | clayey-loamy | 0.077 | 7.03 | 2.4 | 4.7 | 67.7 | 198.7 |

Crocus shares a habitat with maquis and phrygana like Pinus sp. Quercus sp., Muscari sp. Scilla sp. Romulea sp. and Galanthus sp.

The Crocus taxa investigated generally grow on clayey-loamy and clayey soils (Tab. 3). C. pulchellus, C. candidus, C. biflorus subsp. nubigena, C. olivieri subsp. istanbulensis, C. cancellatus subsp. mazziaricus and C. chrysanthus prefer clayey-loamy soils. C. flavus subsp. dissectus, C. pallasii subsp. pallasii and C. gargaricus subsp. gargaricus can grow well on clayey soils.

The pH values were from 6.09 to 7.18 . Total salts were $0.041-0.112 \%$, and calcium carbonate values were $0.4-20.8 \%$. The organic matter values were $3.1-5.7 \%$. The phosphorus and potassium values were $2.5-67.7 \%$ and $52.7-198.7 \%$, respectively (Tab. 3).

## Discussion

Leaves of Crocus species have unique and distinctive shapes in cross section. They comprise a central square or rectangular »keel«, and two lateral»arms«, with their margins generally recurved towards the keel. The central part of the keel consists of large thin walled parenchyma cells which usually break down to form air space.

The arms in all taxa are symmetrical. The abaxial side of arms and adaxial of keels have protrusions only in C. biflorus subsp. nubigena. Similar results have been observed for $C$. danfordiae (Özdemir et al. 2004) and C. cancellatus (Rudall and Mathew 1990).

Hair-like papillae are absent only in C. chrysanthus and C. biflorus subsp. nubigena. There are inconspicuous micropapillae only on the cuticle of C. gargaricus subsp. gargaricus, but it is conspicuous in other taxa.

Anticlinal walls of the epidermis in stomatal regions for all taxa are generally sinuous, or sometimes straight. The same features have been observed on the leaf of Crocus candidus, C. flavus and C. benadicus (Rudall and Mathew 1990).

The mesophyll of all taxa consists of 1-2-layered palisade and 2-5-layered spongy cells. Vascular bundles are collateral. There are four large bundles in the margins of the arms and on the corners of keels. Small bundles extend between the large bundles.

Larger bundles extend to epidermis as girds in C. chrysanthus, C. cancellatus subsp. mazziaricus, and C. pulchellus but not in C. gargaricus subsp. gargaricus, C. biflorus subsp. nubigena, C. flavus subsp. dissectus, C. olivieri subsp. istanbulensis, C. candidus, C. pallasii subsp. pallasii.

The investigated Crocus taxa generally grow on clayey-loamy and clayey soils. The pH of the soil varies between $6.09-7.18$. Crocus taxa generally prefer neutral and slightly acidic soils. All taxa in this study generally grow on calcareous soils. Soils where Crocus taxa occurs are rich in potassium and organic matter. Soils where C. pulchellus, C. pallasii subsp. pallasii, C. candidus, C. olivieri subsp. istanbulensis occur are rich in phosporus, but soils where C. gargaricus subsp. gargaricus, C. chrysanthus, C. flavus subsp. dissectus, and C. cancellatus subsp. mazziaricus occur are poor in phosphorus. Phosphorus values are medium in soils where C. biflorus subsp. nubigena is found.

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[^0]:    * Corresponding author: fsatil@balikesir.edu.tr

