# A Morphological and Anatomical Study on Endemic Alkanna haussknechtii Bornm. (Boraginaceae), Critically Endangered in Turkey

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**Abstract:** Alkanna haussknechtii Bornm., which occurs only in the vicinity of Amasya, is critically endangered. The morphological and anatomical properties of the species, an endemic plant of the Irano-Turanian phytogeographic region, were investigated. The root is perennial. The leaves are ecvifacial and have stomata cells that are anisocytic and anomocytic. The stomata index is 18.048 on the upper epidermis and 15.428 on the lower epidermis.

Key Words: Alkanna haussknechtii, critically endangered, endemic, morphology, anatomy.

# Türkiye'de Vahim Durumda Olan Endemik *Alkanna haussknechtii* Bornm. (*Boraginaceae*) Üzerine Morfolojik ve Anatomik Bir Çalışma

**Özet:** Sadece Amasya ve çevresinde yayılış gösteren *Alkanna haussknechtii* Bornm. yok olma tehlikesi altındadır. İran-Turan floristik bölgesinin bir endemik bitkisi olan türün morfolojik ve anatomik özellikleri incelenmiştir. Kök çok yıllıktır. Yapraklar ekvifasiyal ve stoma hücreleri anizositik ve anomositiktir. Stoma indeksi üst epidermisde 18.048, alt epidermisde 15.428 dir.

Anahtar Sözcükler: Alkanna haussknechtii, çok tehlikede olan, endemik, morfoloji, anatomi.

#### Introduction

The genus *Alkanna* Tausch is a member of the family *Boraginaceae* and is found throughout temperate and subtropical areas of the world, with a major centre of distribution in the Mediterranean region (Heywood, 1978). Thirty-four species are found in Turkey: 76% of which are endemic to Turkey (Davis, 1978; Özhatay, 1994; Güner et al., 2000).

Various local names are used for *Alkanna* species. The names "Havaciva", "Eğnik", "Kızılenik", "Tüylüboya", and "Yerineği" are used for *Alkanna tinctoria* L.. *Alkanna orientalis* (L.) Boiss. is known by the names "Tosbağaotu" "Kanburuyan" and "Kurbağaotu" (Baytop, 1994). The name "Boyaotu" is used for *Alkanna cappodocica* Boiss. &

Bal. and the names "Kökboya" and "Karakök" are used for *Alkanna pseudotinctoria* Hausskn. ex Hub.-Mor. in the vicinity of Aksaray (Ertuğ, 1999).

Some *Alkanna* species are used as potherbs and for dye. *A. tinctoria* is used to stain wood and marble and to colour medicines, wines and cosmetics (Heywood, 1978). A red dye is obtained from the roots of *A. orientalis* and *A. lehmanii* Tineo (Blamey & Grey-Wilson, 1993; Baytop, 1994; Viney, 1994). The economic importance of this species is unclear. Nevertheless, it is presumed to be used in the medical and dying industries, like the other members of the family *Boraginaceae* (Baytop, 1984).

*A. haussknechtii* Bornm. is an taxon endemic to Turkey and Irano-Turanian phytogeographic region

elements. This species was first collected by Bornmüller (1889) around Kirazlidere on Mount Lokman (Amasya) and was described as a new species by the same author in 1894. After 100 years A. haussknechtii species were collected by Karaer and Cansaran (1998) on Mount Karaman and around Yuvacık village in Amasya. There are few specimens in these areas. In these areas where this species has been collected and seen if is under severe threat and the risk of extinction is high because of construction in Amasya and the annual cleaning of vineyards. A. haussknechtii grows on steppe, forest clearings, grassy plains and vineyards. Its altitude range varies between 400 and 600 m (Davis, 1978). A. haussknechtii has been previously reported in the CR category (Critically endangered) (Ekim et al., 2000; IUCN, 2001).

Studies on its morphology are limited. The pollen morphology of some *Alkanna* species were studied by Doğan and Sümbül (1997). The anatomical structure of *A. haussknechtii* has not been studied. Therefore, the purpose of this paper is to investigate the morphologic and anatomic properties of *A. haussknechtii*.

## Materials and Methods

Plant samples were collected from around Amasya. These localities are listed below. Specimens were preserved in the herbarium at Ondokuz Mayıs University, Amasya Education Faculty. The plant numbers for *A. haussknechtii* in 2000 and 2001 are given in Table 1.

A5 Amasya: Amasya, Karaman Mountain, roadside and open areas, 500 m, 18.05. 2000, Akçin 1100.

A5 Amasya: Amasya, Education Faculty district, open areas, 400 m, 19.05.2000, Akçin 1101.

Table 1. Plant numbers for A. haussknechtii in 2000 and 2001.

Locality	The plant numbers (100 m <sup>2</sup> )	
	2000	2001
Amasya, high School	6	3
Amasya, Education Faculty district	5	0
Amasya, Direkli, Yassıçal village	10	4
Amasya, Karaman Mountain	5	1
Amasya, centre district (Şehycui)	6	2
Amasya, Yukarı Kale village	7	4

A5 Amasya: Amasya, centre (Şeyhcui district), cultivation areas (vineyard field), 400 m, 22.05.2000, Akçin 1102.

A5 Amasya: Amasya, high school, roadside and scrub areas, 450 m, 26.05.2000, Akçin 1103.

A5 Amasya: Amasya, Yukarı Kale village, cultivation areas (vineyard field), 600 m, 26.05.2000, Akçin 1104.

A5 Amasya: Amasya, Direkli, Yassıçal village, steppe areas, 300 m, 04.06.2000, Akçin 1105.

The taxonomical description of the plants was carried out according to Davis (1978). Fresh samples were used for morphological measurements. Samples were fixed in 70% alcohol for anatomical studies. The paraffin method was used for preparing cross – sections of root, stem, and leaf (Algan, 1981). The length and width of the stomata were measured with an ocular micrometer using surface sections from the upper and lower parts of the leaf epidermis. The stomatal index was calculated according to the method described by Meidner & Mansfield (1968).

#### Results

#### I- Morphological Properties (Figure 1)

Perennial. Root 4-7 cm long, dark brown. Stem 7-20 cm tall with few branches, glandular and eglandular hairy. Leaves glandular and eglandular hairy. Basal leaves 1.5-3 x 0.2-0.4 cm, linear to oblong–lanceolate. Cauline leaves 2.2-2.8 x 0.3-0.5 cm. Bracts as long as calyx, narrowly or broadly lanceolate. Calyx 4-8 mm in flower, 6-10 mm in fruit, lobes linear-lanceolate. Corolla 7.5-10 mm, glabrous outside, limb sulphur-yellow 5-lobed. Anthers 2-3.5 mm, filaments 0.5-1 mm. Nutlets 4-6 mm in diameter, tuberculate; beak strongly deflexed.

# **II-Anatomical Properties**

# Root (Figure 2)

Periderm is multilayered on the other surface of root. Elements of phellem are brown. Cortex is 15-20 - layered and parenchymatic. Parenchymatic cells are 8-20 x 15-50  $\mu$ . Cambium cells are 2-3 - layered and distinguishable. Primary pith rays are 2-4 - layered but sometimes 6-7 - layered. Secondary rays are 1-2 - layered. In the pith a primary xylem tissue is present. Secondary xylem has hard sclerenchymatic cells and trachea. Root is red in cross - section (Table 2).



Figure 1. a: general appearance of A. haussknechtii, b: corolla and stamens, c: fruit, d: seed.

Stem (Figures 3,6)

Cuticle layer is thin. Epidermal cells are oval, single layered and with thin walls. There are glandular and eglandular hairs on the epidermis. They are 1-2 cellular. Underneath the epidermis there is collenchyma with 3-4 - layered cells. Cortex parenchyma consists of 4-5 layers of usually oval cells. There is a sclerenchymatic sheath on the phloem part. Cambium is distinguishable and 1-2 -

layered. Xylem tissue is large. Xylem cells are sclerenchymatic. Pith cells are large, cylindrical and thin walled (Table 2).

### Leaf (Figures 4 - 6)

Leaf is isobilateral. Leaves are covered by a thick cuticular layer on both upper and lower surfaces. The upper and lower epidermis consists of a single row of cells in which the width and length are almost equal.



Figure 2. Root, pd: peridermis, p: parenchyma, c: cambium, ph: phloem, sx: secondary xylem, t: trachea, px: primary xylem.

		Broad (µ)		Length (µ)	
		Min	Max	Min	Max
	Periderm cell	25	55	10	20
	Parenchyma cell	17.5	30	10	17.5
ROOT	Cambium cell	20	22.5	7.5	15
	Secondary ray	5	7.5	7.5	17.5
	Primary ray	7.5	17.5	10	28
	Diameter of trachea	15	45	-	-
STEM	Epidermis cell	10	15	10	15
	Collenchyma cell	10	20	10	20
	Parenchyma cell	15	60	20	40
	Diameter of trachea	20	30	10	30
	Diameter of pith cell	30	100	-	-
LEAF	Cuticle	1.5	2.5	-	-
	Upper epidermis cell	20	38	17	25
	Lower epidermis cell	15	30	20	40
	Palisade paren. cell	12.5	17	34	42
	Spongy paren. cell	18	45	20	22.5

Table 2. Anatomical me	asurements of A	A. haussknechtii.
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Figure 3. Stem, e: epidermis, cl: collenchyma, p: parenchyma, s: sclerenchyma, c: cambium, ph: phloem, t: trachea, pr: pith region.



Figure 4. Leaf, cu: cuticle, ue: upper epidermis, pp: palisade parenchyma, sp: spongy parenchyma, v: vascular bundle, cl: collenchyma, le: lower epidermis.

There are many glandular and eglandular hairs on the epidermis. Most of these are eglandular. They are unicellular or multicellular. Palisade parenchyma cells are

1 or rarely 2 – layered and long and cylindrical on both lower and upper epidermis. Palisade cells are rich in chloroplasts. Spongy parenchyma cells are 1-3 - layered.



Figure 5. Surface-section of leaf, a: upper epidermis, b: lower epidermis, st: stoma, ec: epidermis cell, h: hair.



Figure 6. Hair types of stem and leaf a,c: eglandular hair of stem, e: glandular hair of stem, a-d: eglandular hair of leaf, e-f: glandular hair of leaf.

There is a large vascular bundle on the median region of the leaf. Vascular bundles are surrounded by a parenchymatic bundle sheath. Collenchyma cells are present under the lower epidermis. Collenchyma is 1-2 -

layered. The type of vascular bundle is collateral. Stomata type is anomocytic and anisocytic. Stomata cells are present in both the upper and lower epidermis. The number of stomata is  $74 \pm 4$  on the upper epidermis and  $54 \pm 3$  on the lower epidermis of the leaf. The stomata index is 18.048 on the upper epidermis and 15.428 on the lower epidermis (Tables 2,3).

#### Discussion

Alkanna species are used as potherbs and for red dye. A. haussknechtii is an endemic taxon and this species was only known from the type. No information on A. haussknechtii was found in the literature except for some morphological properties (Davis, 1978; Karaer & Cansaran, 1998). Morphological characters such as shape of leaf, corolla and nutlet, structure of bract and calyx and properties of hairs at the stem, leaf and corolla were used as taxonomic characters in determining the species.

Although our results generally agree with the description in the Flora of Turkey, a few differences were also determined. Davis (1978) reported that basal and

Table 3. Stomata features on the upper and lower epidermis of A. haussknechtii.

	Upper surface of leaf	Lower surface of leaf
Number of stomata (1 mm <sup>2</sup> )	74 ± 4	54 ± 3
Number of epidermis cells (1 mm <sup>2</sup> )	336 ± 5	$296 \pm 4$
Stomata index	18.048	15.428
Stomata length (µ)	20-25	20-25
Stomata width (µ)	7-10	5-10

cauline leaves were 1-3 x 0.2-0.8 cm, the calyx was 4-6 mm in flower and 7-8 mm in fruit, and the nutlet was 2-2.5 mm. In our study, basal leaves were  $1.5-3 \times 0.2-0.4$  cm, cauline leaves were  $2.2-2.8 \times 0.3-0.5$  cm, the calyx was 4-8 mm in flower and 6-10 mm in fruit, the corolla was 7.5-10 mm and the nutlet was 4-6 mm. Bract, filament and anther sizes and hair properties are reported here for the first time.

Metcalfe & Chalk (1979) gave information about the general anatomical characteristics of the family Boraginaceae. There was no information about the anatomical structure of this species. The root has primary and secondary pith rays. Primary pith rays are 2-4 layered but sometimes they are 6-7 - layered. Secondary rays are 1-2 - layered. In the pith a primary xylem tissue is present. There is a thin cuticle layer on the stem. Epidermal cells are oval, single - layered with thin walls. There are glandular and eglandular hairs on the epidermis. They are 1-2 cellular. The leaf was isobilateral. It was reported that there was an anomocytic stomata in the family Boraginaceae (Özörgücü, 1991). In addition, Metcalfe & Chalk (1979) pointed out that there were both anomocytic and anisocytic stomata in this family. In this study, it was determined that this species had anomocytic and anisocytic stomata. The numbers of stomata on the upper and lower epidermis were  $74 \pm 4$ and  $54 \pm 3$  respectively. The number of epidermis cells was  $336 \pm 5$  on the upper epidermis and  $296 \pm 4$  on the lower epidermis. The stomata index was 18.048 on the upper surface and 15.428 on the lower surface of the leaf (Table 2).

It was determined that spongy paranchyma increased in the mesophyll in xerophytic plants (Esau, 1977; Fahn, 1982; Yentür, 1995). We found the same characteristics in our research.

This species was only known from the type (Davis, 1978). However Karaer & Cansaran collected *A. haussknechtii* in the vicinity of Amasya (1998). There are few specimens in these areas (Table 1). The species is strongly affected by a number of factors (pollution, extreme drought, annual cleaning of vineyards etc.) because it occurs in cultivated areas. The investigation results show that, the seeds of the species need special environmental conditions to sprout. Unfortunately, seeds have not sprouted for some 2 years due to extreme temperatures and cold in the vicinity of Amasya. Therefore this species has not been studied in terms of chrosome numbers and morphology. *A. haussknechtii* will prove to be an asset to the Turkish economy in the future.

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