A study on the anatomical characteristics of *Vitex agnus-castus* (*Verbenaceae*)

Yunus Dogan[1], Gungor Ay[2] & Ekaterina Kozuharova[3]

[1] Department of Biology, Faculty of Education, Dokuz Eylul University, 35160, Buca–Izmir, Turkey, e-mail: yunus.dogan@deu.edu.tr

[2] Celal Bayar University, Faculty of Science & Art, Department of Biology, Muradiye–Manisa, Turkey

[3] Department of Pharmacognosy and Botany, Faculty of Pharmacy, Medical University, Sofia, Bulgaria

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Abstract. *Vitex agnus-castus* is naturally distributed in many parts of Anatolia and Trace Turkey. In this study, the aim was to investigate the anatomical characteristics of this economically important plant species. As a result of the study, it was discovered that the stem is quadrangular and there are non-glandular hairs on the epidermis of the stem. Starch was also identified in the pith cells. In the leaves of the plant, a single layer hypodermis was clearly seen just beneath the epidermis. Stomata are of anomocytic type. Oxalate crystals were identified in the parenchymatous cell.

Key words: anatomy, medicinal and useful plant, Turkey, *Vitex agnus-castus*

Introduction

Vitex agnus-castus L. is a perennial, deciduous, grey-felted shrub or rarely a small tree, with a 1–6 m height, with a strong aromatic odor (Davis 1982). The species generally grows in humid habitats like stream banks and valleys, in littoral habitats, mostly on sandy soils, parched alluvial soils and rocky areas near the sea, sometimes on limestone slopes, in sunny and hot places and in ditches. It is a Mediterranean element and grows along the Mediterranean and Aegean coasts, penetrating inwards up to approximately 250 km and between the altitudes from sea level to 750 m, particularly in areas in which the Mediterranean climate is dominant (Dogan & Mert 1998). However, Boissier (1879) has reported its existence up to 1200 m.

There are more than 250 species of *Vitex* genus belonging to the *Verbenaceae* family in the world (Dutta 1970). Their distributions vary from the Mediterranean area, South Europe, West Asia, Crimea, Azerbaijan, Georgia, Armenia, Iran, Africa boreals, Central Asia to India, and in sheltered positions in New York, USA (Boissier 1879; Post 1933; Bailey 1947; Hegi 1966; Davis 1982; Saden-Krehula & Kustrak 1991). In Turkey, only two of the species, *V. agnus-castus* and *V. pseudonegundo* (Hausskn. ex Bornm.) Hand.-Mazz., are distributed.

The species *V. agnus-castus* is used for medicinal purposes (Baytop 1984; Saden-Krehula & Kustrak 1991; Brown 1994; Christie & Walker 1997; Lauritzen & al. 1997; Veal 1998; Klepser & Nisly 1999). Its economical use is based on the flexibility of thin and long twigs and this species is used in making small and big baskets in Western Anatolia (Baytop 1984). In order to preserve woolen fabrics from moths, its leaves and fruits are ground and sprinkled over the fabrics (Baytop 1984).

A well known "agnus castus honey" is obtained from this plant in the apiculture throughout Western Anatolia and its leaves are used for dying fabrics in handicrafts (carpet and rugs). In most cities of Western Anatolia the species studied is used as an ornamental plant in parks and gardens (Bailey 1947; Orçun 1975).

Although there is a rise in the investigation of the anatomical characteristics of the plant species in Turkey (Gönüz & Özörgücü 1999; Uysal 1999; Kaya 2003; Orcan & Binzet 2003; Uysal & al. 2003; Ataşlar 2004; Ergen Akçin 2004; Ergen Akçin & al. 2004; Ocak & al. 2004), most of the studies are on the weedy forms, not on the wooden forms, like *V. agnus-castus*.

Since there was no study found on the anatomical characteristics of *V. agnus-castus*, and due to its medicinal and economical importance mentioned above, in this study, the aim was to identify the anatomical characteristics of *V. agnus-castus*.

Material and methods

Sample materials of *V. agnus-castus* investigated in this study were collected from the localities determined from five different cities of Western Anatolia from different altitudes. These are listed below according to the Grid system used by Davis (1982): **1.** A1 Canakkale–Ayvacik; **2.** B1 Balikesir–Savastepe; **3.** B1 Izmir–Beydag; **4.** C2 Denizli–Honaz; **5.** B2 Mugla–Bodrum.

The collected materials were identified taxonomically according to Davis (1982). Then, they were put into a 70 % alcohol-water solution to store for later investigation. Their root, stem and leaf anatomical characteristics were investigated in cross–sections taken in laboratory conditions. The cross-sections were dyed by sartur reactive during microscopic investigation, and then their coloured pictures were taken by a microphotography apparatus.

Results and discussion

Anatomical studies made on *V. agnus-castus* showed similar characteristics of other species of the *Verbenaceae* to which this species belongs (Metcalfe & Chalk 1957).

In the cross-section of the secondary root, it was seen that, derived from the cork, the periderm tissue is formed, which replaces the epidermis in the outer layer (Fig. 1). The next tissue identified inwards was

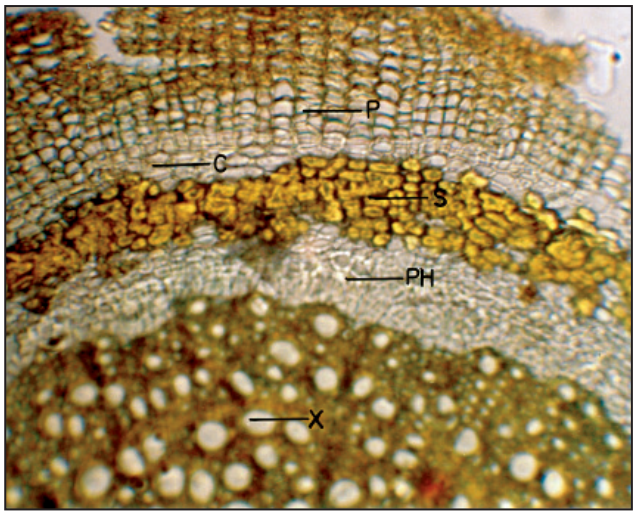

Fig. 1. Root of *V. agnus-castus* (cross section): **P** – peridermis; **C** – cortex; **S** – sclerenchyma**;** **PH** – phloem; **X** – xylem (×160).

the cortex. Sclerenchyma was seen as a ring in the secondary roots, while as a group, formed from a group of cells, in the mature ones. In the middle, xylem tissue occupies a large area, just below the phloem tissue. The observations concerning the root showed a parallelism with the results of Metcalfe & Chalk (1957).

The stem, as a typical characteristic of the *Verbenaceae*, is quadrangular (Watson & Dallwitz 1992). Non-glandular hairs take place on the epidermis. In the outer layer of the young stem, the epidermis covered by a cuticle was observed. In the mature stem, the periderm replaces the epidermis, and the cork cells are thin-walled. In the peripheral part of the stem, collenchymatous tissue is seen as a layer. The sclerenchymatous tissue is found as a continuous ring in the young stem, while it is found as cell groups in the mature stem (Fig. 2). Just below the cortex, phloem tissue is seen. Beneath the phloem tissue, cambium tissue from 3 to 5 cell layers is found. Under the cambium tissue, xylem tissue takes place. Wood parenchyma is similar, simple and elongated. Rays become much broader in the mature wood. The pith of the plant stem is formed from parenchymatous cells.

On the other hand, the existence of the starch was determined in the pith cells (Fig. 3). Richter & Dallwitz (2000) studied the *Vitex* species used as commercial timbers. Since *V. agnus-castus* is not used as a timber, they did not include the species in their study. They reported that they saw tylosis in the vessels of the species that they studied, and they concluded that this was the characteristics for the entire *Vitex* genus. However, in this study, the same structure is not seen in *V. agnus-castus*. They also concluded that there is

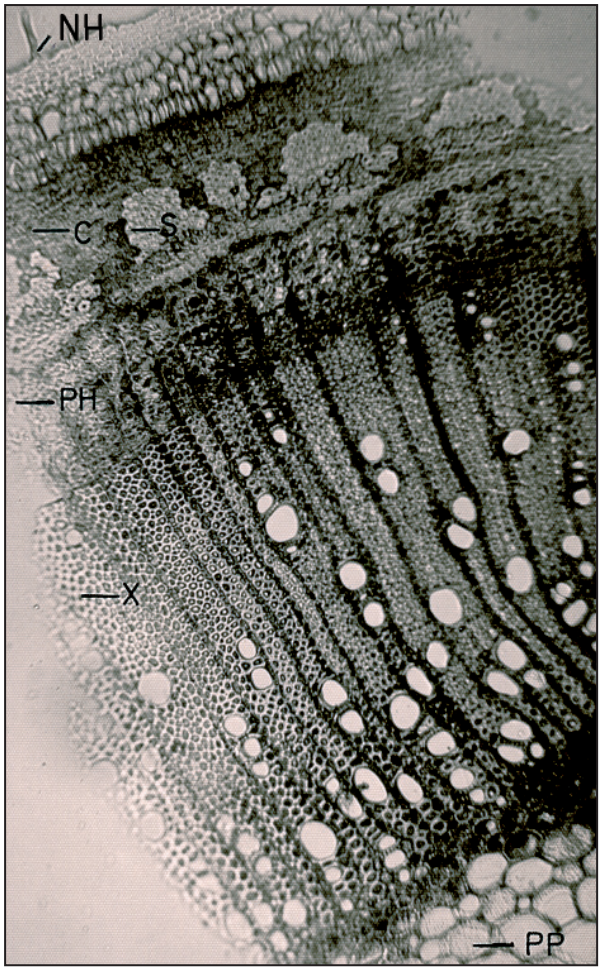

Fig. 2. Stem of *V. agnus-castus* (cross section): **NH** – non-glandular hair; **C** –cortex; **S** – sclerenchyma; **PH** – phloem; **X** – xylem; **PP** – parenchymatous pith (×100).

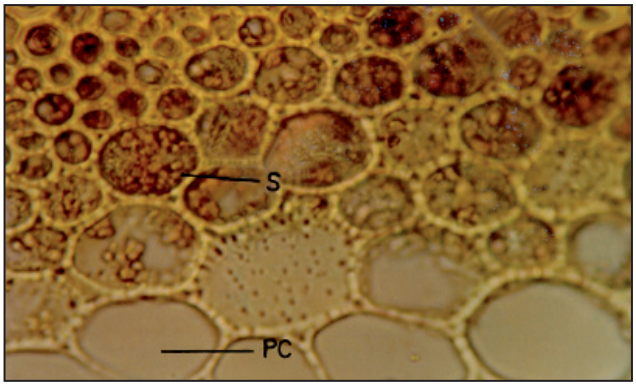

Fig. 3. Starch in the pith of stem (cross section): **S** – starch; **PC** – parenchymatous pith (×640).

no stored structure in *Vitex* genus, but it was found as a result of our study that *V. agnus-castus* has starch deposition in the pith of the stem. The anatomical structure in the cross section of the stem is in accord with the general characteristics of the members of the *Verbenaceae* (Metcalfe & Chalk 1957).

In the leaves, the outer surface of the upper epidermis is covered by a cuticle with non-glandular hairs on its surface. These hairs are short and conical (Fig. 4). These results show a parallelism with the record of Metcalfe & Chalk (1957). However, two-celled, non-glandular longer hairs in the lower epider-

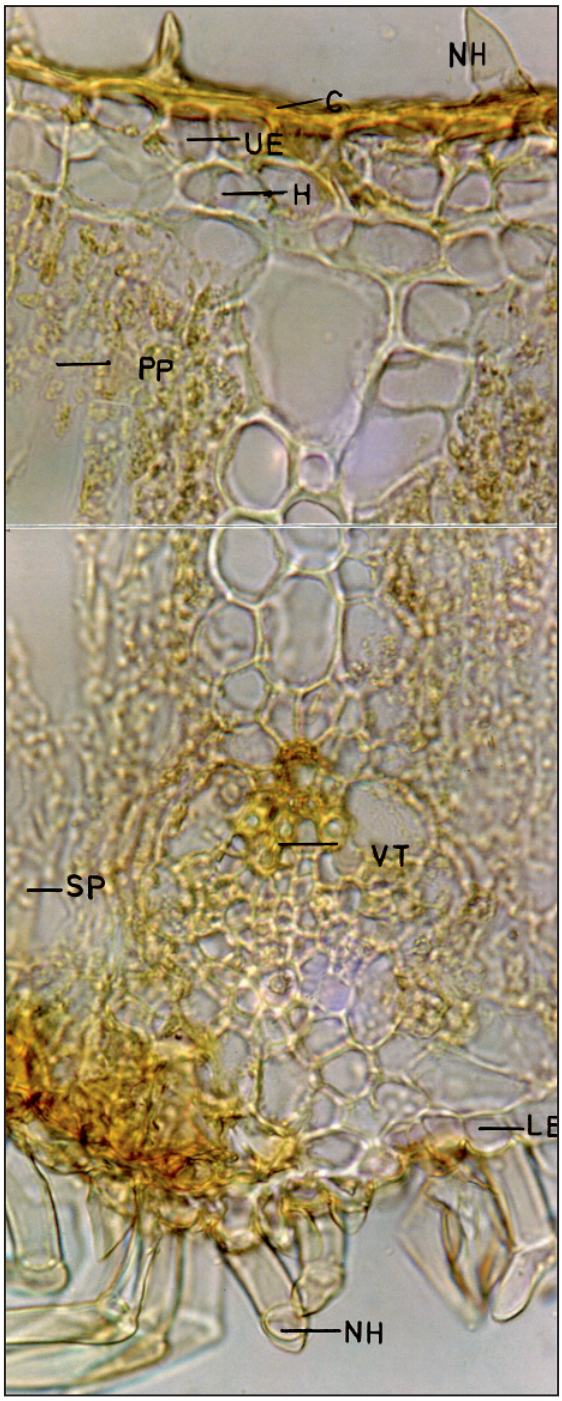

Fig. 4. Vascular tissue in leaf (cross section): **NH** – non-glandular hair; **C** – cuticle; **UE** – upper epidermis; **H** – hypodermis; **PP** – palisade parenchyma; **SP** – spongy parenchyma; **VT** – vascular tissue; **LE** – lower epidermis (×640).

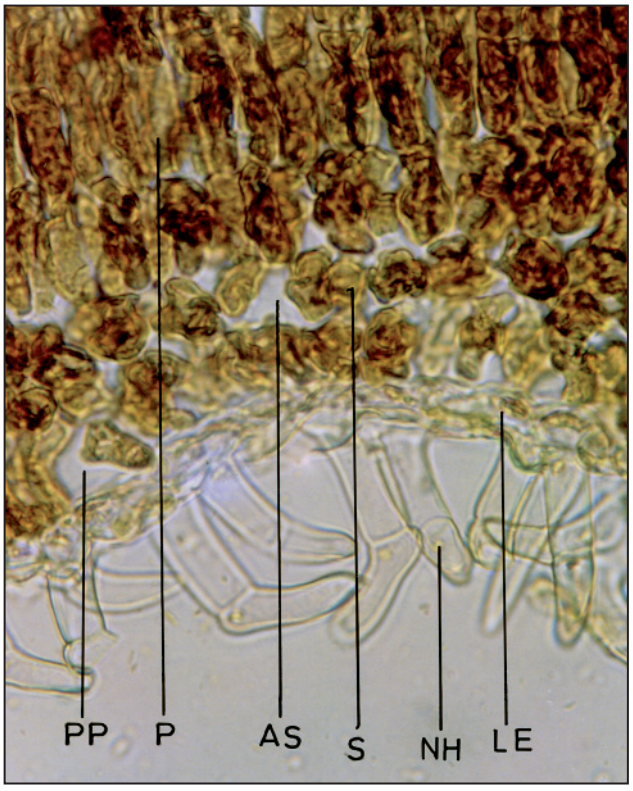

Fig. 5. Leaf of *V. agnus-castus* (cross section): **PP** – palisade parenchyma; **SP** – spongy parenchyma; **AS** – air space; **LE** – lower epidermis; **NH** – non-glandular hair (×640).

mis (Fig. 5) were not mentioned by Metcalfe & Chalk (1957). The stomata were found only in the lower surface of the leaf, on the same level with the epidermis (Fig. 6). It was observed that leaves of *V. agnus-castus* do not have subsidiary cells, and the epidermis cells surround the stomata (anomocytic type). Beneath the upper epidermis, a single layer hypodermis is found. The next layer is the palisade parenchyma. Its cells are arranged very tightly in three to four rows and are rich with chlorophyll. The intercellular spaces are rare.

In the records of Metcalfe & Chalk (1957), we do not find any information about the thickness of the palisade parenchyma. Just beneath the palisade parenchyma, the spongy parenchyma is found. Its cells contain less chlorophyll in comparison to the palisade parenchyma cells and their air spaces are larger.

Oxalate sands were found in the transverse section of the leaves (Fig. 7). Described as prismatic cyristals, the existence of oxalate sands in the anatomic sections of the leaf is also reported by Metcalfe & Chalk (1957) and Özörgücü & al. (1991). Similarly, Richter & Dallwitz (2000) reported that some *Vitex* species have prismatic crystals, while others do not. In *V. agnus-castus*, they were seen only in the cross sections of the leaf.

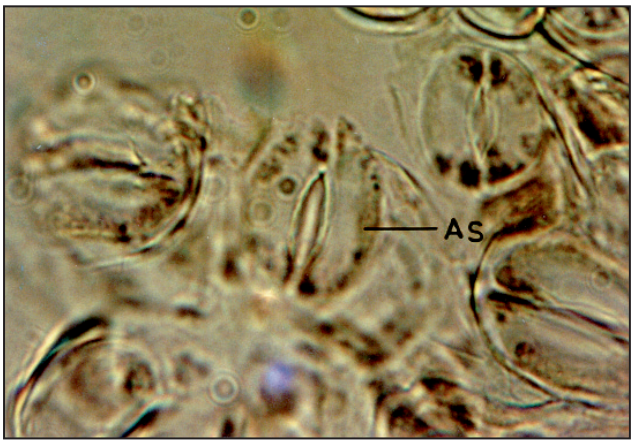

Fig. 6. Lower epidermis with stomata of a leaf (transverse section): **AS** – anomocytic stoma (×1600).

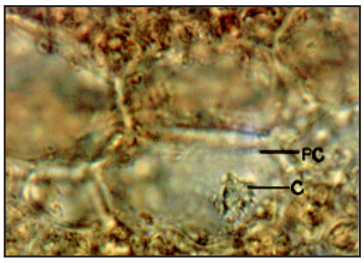

Fig. 7. A crystal in the leaf of *V. agnus-castus* (transverse section): **PC** – parenchymatous cell; **C** – crystal (×1600).

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