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## Pharmacognostic Studies on the Leaf of *Anisotes trisulcus* (Forssk.) Nees

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**Abstract:** Anatomical Studies on the leaf of *Anisotes trisulcus* (Forssk.) Nees have been carried out. Macro and microscopical characters are illustrated. Fluorescence characteristics and ash and extractive values are given. Many diagnostic elements were found to be useful in the identification of the plant.

**Key words:** Leaf *Anisotes trisulcus*, pharmacognostic

### Introduction

*Anisotes trisulcus* (Forssk.) Nees, family Acanthaceae is indigenous to Yemen and widely distributed in southern mountainous region of the Kingdom of Saudi Arabia, particularly in rocky Wadi of Jabal Abu Hassan, between Abha and Najran (Collenette, 1985). The plant is distasteful to animals, and is popular amongst the natives by the local names "Madh", "Moze" and "Moddaid" (Al-Hubaishi and Multer-Hohenstein, 1984). *Anisotes trisulcus* is a traditional herbal medicine in the Arabian peninsula and found its way in the folk medicine of Saudi Arabia as an antidiabetic. The antidiabetic effect of the plant was confirmed. The traditional Pharmacopoeia of Yemen described this plant as a herbal medicine for the treatment of all hepatic syndromes, including jaundice and hepatitis, gallstone and other hepatic disorders (Fleurentin and Pelt, 1982; Fleurentin *et al.*, 1983). The pharmacology of various extracts of *Anisotes trisulcus* (Forssk.) Nees were studied, the hepatoprotective effects were evaluated and the therapeutic values of the plant were confirmed (Fleurentin *et al.*, 1986). A literature survey revealed that the genus *Anisotes* is rich in alkaloids (Arndt *et al.*, 1967). In pharmacognostic field no work is on record except that Metcalfe and Chalk (1965) have given a brief and generalized description of the genus. As part of our study programme (pharmacognostic and phytochemical) on the plant *Anisotes trisulcus* (Forssk.) Nees, the pharmacognostic studies on the leaves have been carried out. In the present work the diagnostic characters of the leaves are described with the illustrations.

### Materials and Methods

Samples of *Anisotes trisulcus* (Forssk.) Nees, were collected from plants growing in different localities of southern region of Saudi Arabia (Fayfa mountain). Histological studies were carried out on 1) fresh leaves, 2) air dried leaves and 3) leaves preserved in formalin-acetic alcohol (F.A.A.) mixture. Free hand sections were taken, stained and mounted in the usual way. Representative diagrams were sketched with the help of a projection microscope (Lampenhau 250, W. Germany). Microchemical tests for the cell contents and cell wall structures were performed according to the methods described by Trease and Evans (1983) and Johansen (1940). Methods for determining the quantitative values of the leaf were those adopted by Wallis (1953). The extractive and ash values were made as described in the British Pharmacopoeia (1980). For fluorescence analysis the

powder of the leaf was examined under UV light according to the method described by Chase and Pratt (1949). The measurements of various cells in the transverse section and in maceratin are reported in Table 1. The values in Table 1, 2, 3 and 4 are written in a range where the first numerical figure is the minimum value, the middle numerical figure is the average value and the last numerical figure is the maximum value.

### Results and Discussion

**Morphology:** *Anisotes trisulcus* (Forssk.) Nees is a stiffy erect shrub which grows to a height of 1-3.5 m. The plant bears bright orange-red tubular flowers (Fig. 1). Branches bear opposite and decussate leaves at the nodes. The leaves are simple, petiolate and stipulate. Petioles are 3-20 mm long and leave permanent scar after falling. The lamina is ovate to elliptic and measures 2-10 cm long and 0.5-6 cm wide. The apex of the leaf is obtuse or sub-acute, the margin is entire and the base is somewhat truncate or cuneate. The lamina is glabrescent, yellowish green and glandular dotted on both sides. The venation is reticulate with 6-9 pairs of lateral veins. The midrib of the leaf projects on both sides, being more prominent on the lower side. The leaf has no characteristic odour and has bitter taste.

**Histology:** The leaf is dorsiventral and transcurrent. The transverse section of the leaf exhibits the following characters (Fig. 2, 3).

**Epidermis:** The upper epidermis consists of single layer of polygonal cells with fairly thick cuticle. The upper epidermis contains numerous trichomes and stomata. Stomata are caryophyllaceous and are abundant in the upper epidermis. Trichomes are glandular and non-glandular. The glandular trichomes are short stalked with radiating glandular head. The non-glandular trichomes are either unicellular covering or multicellular covering trichomes. The unicellular covering trichomes are characterized by thick walls and narrow lumina. The latter being enlarged towards the base of each trichome. The multicellular covering trichomes are either bicellular or tricellular and have acute apex. The lower epidermis resembles the upper epidermis. The stomatal index and stomatal number of the lower epidermis are less than that of the upper epidermis.

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Table1: Measurements of different cells and tissues of the leaf of *Anisotes trisulcus* (Forssk.) Nees

Cell specification	Measurements (mean)	
	Leaf	Petiole
Upper epidermis	16-28-40 × 13-25-35	9-13-17 × 0-12-17
Lower epidermis	13-25-35 × 9-20-30	-
Palisade cells	33-80-1 25 × 9-12-16	-
Spongy parenchyma	13-22-30 9 (diameter)	-
Collenchyma	13-30-46 (diameter)	9-25-40 (diameter)
Cortical parenchyma Collenchymatous-pericycle	16-31-46 (diameter)	9-35-60 (diameter)
Phloem parenchyma	4-10-18 (diameter)	6-13-20 (diameter)
Xylem vessel	3-5-8 (diameter)	2-6-9 (diameter)
Xylem fibre	9-18-26 (diameter)	6-16-26 (diameter)
Xylem parenchyma	150-375-600 × 3-17-30	115-445-780 × 3-18-33
Tracheidal fibre	6-8-10 (diameter)	6-12-19 (diameter)
Covering trichome (unicellular)	165-180-198 × 7-16	66-116-165 × 13-19-25
Covering trichome (multicellular)	19-43-66 × 9-13-1 7	26-40-53 × 9-14-20
Glandular trichome Stomata:	48-107-165 × 9-13-17	66-140 200 × 13-19-26
a) Upper epidermis	33-32-36 (diameter)	26-30-33 (diameter)
b) Lower epidermis	16-20-25 × 9-13-16	16-8-20 × 10 -12-13
	16-18-20 × 10 - 12-13	-

Table 2: Quantitative values of the leaf of *Anisotes trisulcus* (Forssk.) Nees

Determination of	Range
Palisade-ratio	8-10-12
Vein-islet number	18-23-28
Vein-terminal number	5-6-12
Stomata! number	190-270-352
(Upper epidermis) Stomatal number	80-130-180
(Lower epidermis) Stomatal index	14-17-20
(Upper epidermis) Stomatal index	6.5-10.7-15.0

Table 3: Ash values of the leaf of *Anisotes trisulcus* (Forssk.) Nees

Determination of Ash	Percentage of Ash
Total ash	18.6-19.2-19.6
Acid insoluble ash	0.2-0.3-0.4
Water soluble ash	3.8-4.4-4.8

Table 4: Extractive values of the leaf of *Anisotes trisulcus* (Forssk.) Nees

Name of the Extract	Percentage of the Extract
Pet. Ether Extract	1.0-1.5-2.0
Chloroform Extract	2.5-2.7-3.0
Alcohol Extract	13.5-14.0-15.0
Water Extract	9.0-9.5-10.0

Table 5: Fluorescence Characteristics of the leaf of *Anisotes trisulcus* (Forssk.) Nees

Powder observed	Colour observed	
	Day light	UV light
Dry powder	Green	Yellow
Powder treated with NaOH in Methanol	Yellowish-green	Yellow
Powder treated with AlCl <sub>3</sub> in Methanol	Dark yellowish-green	Dark greenish-yellow
Powder treated with Antimony solution trichloride	Dark green	Black
Powder treated with 5% ferric Solution chloride	Dark bluish-green	Black Powder
Treated with HNO <sub>3</sub> (1:1)	Yellowish brown	Dark brown
Powder treated with HCl (1:1)	Gray	Black
Powder treated with H <sub>2</sub> SO <sub>4</sub>	Dark grayish green	Black

**Mesophyll:** The mesophyll is clearly differentiated into palisade and spongy mesophyll. Under each upper epidermis the mesophyll contains a single layer (rarely two layers) of palisade which is composed of compactly arranged cylindrical cells. The spongy mesophyll consists of 3.8 layers of spongy parenchyma cells. Numerous cystolith of calcium carbonate are scattered in the spongy

June 20, 2000 parenchyma and in palisade as well. The cystoliths are characteristic, solitary, either rounded or elongated with blunt extremities or elongated with one end pointed.

### Midrib

**Epidermis:** The upper and lower epidermises of the midrib are similar to that of the lamina except that the cells are smaller. Glandular trichomes are abundant.

**Cortex:** The cortex contains two prominent zones of colenchyma, one adjacent to each epidermis. Each zone is made up of 2-6 layers of thick walled collenchymatous cells. The remainder of the cortex consists of rounded parenchyma cells. The cortical parenchyma cells are isodiametric and contain acicular fibers and cystolith of calcium carbonate. The cystoliths are either rounded or elongated with blunt extremities or elongated with one end pointed. The acicular fibers are in bundles and resemble large raphides.

**Meristele:** The meristele is crescent shaped with radiating xylem, an arch of phloem and batches of collenchymatous pericycle. The xylem is lignified and composed of xylem vessels, xylem fibres, tracheidal fibres and xylem parenchyma. The vessels exhibit spiral and pitted thickening. These fibres are very few and are slightly lignified. The phloem is composed of 5-15 layers of compactly arranged thin walled cells. The pericycle consists of batches of 1-3 layers of collenchymatous cells. *C. petiole* (Fig. 3). The transverse section of the petiole is crescent shaped in outline and it exhibits the following characters.

**Epidermis:** The epidermis is composed of single layer of polygonal cells with smooth and thick cuticle. The epidermis contains glandular and non-glandular trichomes and caryophyllaceous stomata. The trichomes of the lamina of the leaf and of petiole resemble each other.

**Cortex:** Adjacent to epidermis the cortex contains 2-6 layers of thick walled rounded collenchyma cells. The rest of the cortex is composed of rounded parenchyma cells. Some of the parenchyma cells contain cystolith and others contain acicular fibres. The cystolith and acicular fibres are

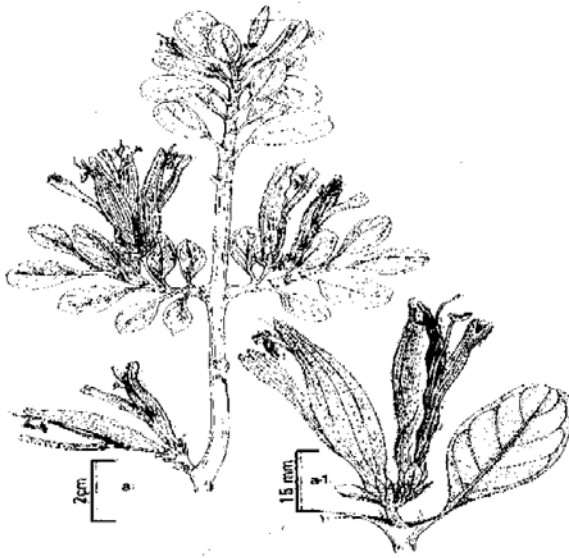


Fig. 1: *Anisotes trisulcus* (Forssk.) Nees

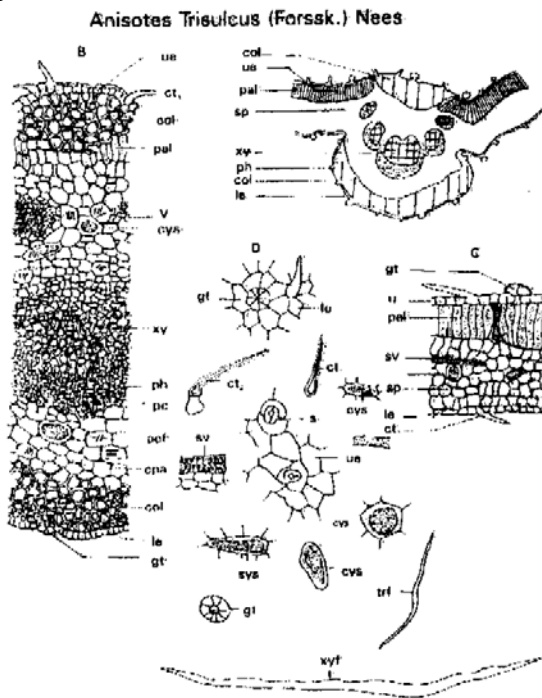


Fig. 2: *Anisotes trisulcus*. (Forssk.). Nees-THE LEAF

A: T.S. of the leaf through midrib-ground plan (x 50)  
 B: T.S. of the midrib-portion enlarged (x 170)  
 C: T.S. of the lamina a portion, enlarged (x 70)  
 D: Powder characters of the lamina and midrib  
 acf: Acicular fibres col = collenchyma, cpa = cortical parenchyma, ct<sub>1</sub> = unicellular covering trichome, ct<sub>2</sub> = multicellular covering trichome, cys = cystolith, gt = glandular trichome, le = lower epidermis, pal = palisade cells, pc = pericycle, ph = phloem, s = stoma, sp = spongy parenchyma, sv = spiral vessel, ue = upper epidermis v = vascular bundle and xy = xylem

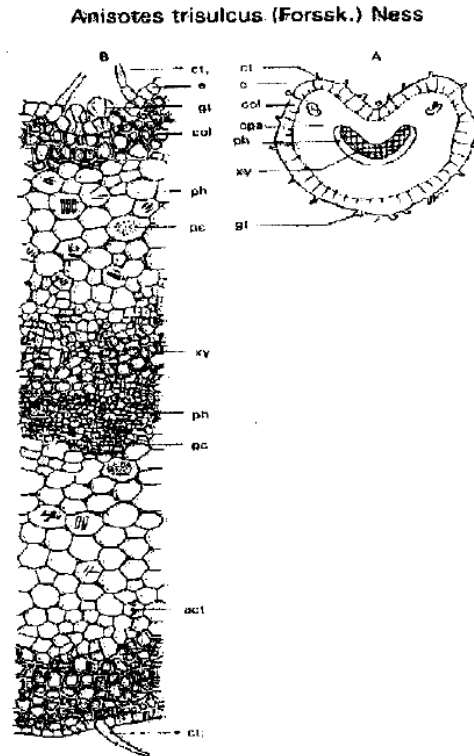


Fig. 3: *Anisotes trisulcu* (Forssk.) Nees-THE LEAF

A: T.S. of petiole-ground: plant (x 50)  
 B: T.S. of petiole - a portion enlarged (x 170)  
 acf: Acicular fibres, col = collenchyma, cpa = cortical parenchyma, ct = covering trichome, ct<sub>1</sub> = unicellular covering trichome, ct<sub>2</sub> = multicellular covering trichome, sys = cystolith, e = epidermis, gt = glandular trichome, ph = pericycle, ph = phloem and xy = xylem

similar to those found in midrib and lamina of the leaf.

**Vascular bundles:** The petiole contains a large crescent shaped vascular bundle in the center of the cortex and two smaller vasculae bundles on either side. The vascular bundles consist of radiating xylem, an arch phloem and batches of collenchymatous pericycle. The xylem is made up of xylem vessels, xylem fibres, tracheidal fibres and xylem parenchyma. The vessels are lignified and exhibit spiral and pitted thickening. The fibres are few and are lignified. The phloem consists, of compactly arranged thin walled cells. The pericycle is made up of bathces of collenchymatous parenchyma cells.

**Quantitative values:** The quantitative values of the leaf palisade-ratio, vein-islet and vein-terminal numbers, stomatal number and stornatal index have been determined in different samples of the leaf at different regions and the consolidated results-are reported in the Table 2. It is observed that the vein-islet and vein-terminal numbers are higher in the young leaf.

**Constant**

**Ash value:** The percentage of total ash, water soluble ash and acid insoluble ash for the leaf are tabulated (Table 3).

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**Extractive value:** The percentage of the extracts of the leaf in pet. Ether, chloroform, alcohol and water are reported in Table 4.

**Fluorescence analysis:** The fluorescence characters of the leaf powder under ultra-violet light are recorded in Table 5.

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