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Pharmacognostic Studies of *Capparis cartilaginea* Decne Leaves

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ABSTRACT

Yemeni medicinal plants have very effective medicinal value, but only very few have been studied. Such plants include *Capparis cartilaginea* Decne, Capparaceae. The whole leaves and their powder of this plant were selected to evaluate the pharmacognostic characteristics; macroscopic and microscopic examinations, physicochemical and fluorescence determinations were carried out. The mature leaf is simple, entire, leathery and evergreen. It is petiolated, oval-shaped, broad and fleshy, often ending in a hooked, astriate and thick cuticle, anomocytic stomata, mesophyll is diffused with no clearly distinct palisade and spongy mesophylls have been found. The physicochemical studies showed ash value 6.10 %, acid insoluble ash value 1.50 %, water soluble ash 4.10, alcohol extractive value 13.11 %, water extractive value 36.40% and moisture content 9.5 %. Fluorescent study exhibited characteristic colour data. The obtained results are useful for the standardization of studied plant.

INTRODUCTION

The use of herbs as medicine is the oldest form of healthcare known to humanity and has been used in all cultures throughout history [1]. In recent years, there has been growing interest in alternative therapies and the therapeutic use of natural products, especially those derived from plants [2, 3]. Since crude plant drugs form the basis for the manufacture of many medical preparations, accurate determination of drug identity forms an essential part of its study. It becomes extremely important to make an effort towards standardization of the plant material as medicine. The process of standardization can be achieved by stepwise pharmacognostic studies [4]. The pharmacognostic parameters are necessary for confirmation of the identity and determination of quality and purity of a crude drug [5]. These studies help in identification and authentication of the plant *Capparis cartilaginea* Decne (Capparaceae) leaves, which are used to treat itching, shortness of breath, head cold and for tumors [6], and for wounds, boil [7]. The leaves are reported to possess antimicrobial [8, 9] and antioxidant activities [10]. Qualitative chemical tests revealed the presence of carbohydrates, saponins, polyphenols (flavonoids and tannins), triterpenes, sterols, amino acid and protein in the leaves [10]. However, available literature revealed that no pharmacognostic study has been carried out on the plant, hence the present investigation was undertaken. The object of present study is to evaluate various pharmacognostical parameters such as macroscopic, microscopy, physicochemical and fluorescence studies of the plant.

MATERIALS AND METHODS

Collection and identification of plant material

The sample leaves of *Capparis cartilaginea* Decne were collected from Zarah, Abyan, Yemen in September 2015 and authentication has been done by a taxonomist, professor Abdul Nasser Algifri, of the faculty of Biology, university of Aden. The sample leaves dried in the shaded area and then manually grinded and stored at room temperature for further analysis.

Macroscopic characteristics

Macroscopic studies of the plant were carried-out as per the produce given in WHO guidelines [11]. The plant was macroscopically examined for shape, size, surface characteristics, texture, color, consistency, odour and taste.

Microscopic characteristics of leaves and petiole

Free hand sections of leaves and petiole of leaves of *Capparis cartilaginea* Decne were studies as per the produce given in WHO guidelines [11]. Photomicrographs were taken with Leica USA model 2000ATC (ocular: CPL W10X; objective: 4X, 10X and 40X). Various identifying characters, such as type of trichomes, type of stomata and epidermal cells were recorded, and then photomicrography was done [12]. Photographs were taken with the help of digital camera (Sony 10 MP).

Microscopic characteristics of leaves powder

The powders of the dry leaves passed through 100 # sieve were cleared in chloral hydrate solution, mounted in 10% aqueous glycerin, covered with a cover-slip and examined to outline the diagnostic features of the leaves in the powdered conditions [11]. Photographs were taken with the help of digital camera (Sony 10 MP).

Physicochemical analysis

Physico-chemical analysis; loss on drying, total ash, acid insoluble ash, water-soluble ash, water soluble extractive and ethanol soluble extractive were determined for the quality and purity of the crude drugs according to the official methods described [11].

Fluorescence analysis of powdered drug

A fine powder of studied leaves was placed on a grease free clean microscopic slide and added 1-2 drops of the freshly prepared reagent solutions mixed properly and waited for 1-2 minutes. Then the slide was viewed in daylight and inside the UV viewer chamber short (254 nm) and long (365 nm) ultraviolet radiations. The colors observed by application of different reagents in different radiations were recorded [13].

RESULTS AND DISCUSSION

Macroscopic studies

The matured leaf of *Capparis cartilaginea* Decne is simple, entire, leathery and evergreen. The arrangement of the leaves on the stem is opposite and decussates. The leaf is petiolated (the petiole measures between 5– 10 cm), oval-shaped, broad and fleshy, often ending in a hooked. The leaf has entire margin, apex and base are rounded. The leaf measures between 7

– 10cm in width and 7 – 11 cm in length (Figure 1). The macroscopic and microscopic description of medicinal plants is the first step towards establishing the identity and degree of purity of such materials and should be carried out before any tests are undertaken [11].

Figure 1: Upper Surface of Leaf of *Capparis cartilaginea* Decne

Microscopic studies

Microscopic studies of leaves

The blade, in surface view of both upper and lower surfaces, has epidermal cells exhibiting an oval, hexagonal and polygonal shape (Figures 2, 3). A striate and reticular cuticle appears to be prominent on both the upper and lower sides of leaf lamina (Figure 4). The epidermis of both surfaces has anomocytic (irregular-celled) type of stomata. Each stoma consists of two kidney-shaped cells called guard cells (Figures 5). Rarely stoma consists of three kidney-shaped guard cells (Figure 6). Guard cells may have triangular or oval shape (Figure 7). Trichomes are absents on both surfaces of epidermis. Cluster crystals of calcium oxalate were found abundantly on both upper and lower surface (Figure 8).

Transverse section of leaf: Transverse section passing through midrib shows single layered upper and layer epidermis. Midrib showed one large vascular bundle. The mesophyll is diffused with no clearly distinct palisade and spongy mesophylls. However, palisade mesophyll-like layer showed smaller cells than the spongy mesophyll-like layer. The spongy mesophyll-like layer had few air spaces (Figure 9). No distinct palisade layer evident.

A striate and thick cuticle, anomocytic stomata, mesophyll is diffused with no clearly distinct palisade and spongy mesophylls have been found in Capparis members [14], as seen in the studied species.

Petiole: In surface view the epidermal cells showed with oval, hexagonal and polygonal shape with reticular cuticle. Anomocytic stomata are present (Figure 10).

Transverse section of Petiole: The general structure of the transverse section of the petiole appeared circular. The outermost layer is formed of one layer of epidermis covered with cuticle, followed by collenchyma and parenchyma cells**.** The vascular bundles are arranged in a circle. The phloem region is followed by the xylem. The pith is a wide region of thickened parenchyma cells. Cluster crystals of calcium oxalate were found in the pith and in parenchyma. Sclerenchyma cells were observed in parenchyma (Figure 11).

Powder microscopy

The powders of the dry leaves passed through 100 # sieve were cleared in chloral hydrate solution, mounted in 10% aqueous glycerin, covered with a cover-slip and examined to outline the diagnostic features of the leaves in the powdered conditions. The powder microscopy study showed the presence of epidermis in surface view with oval, hexagonal and polygonal shape coated with a striate and thick cuticle (figure 12 a). Cluster crystals of calcium oxalate, fragment of midrib and lateral vein (figure 12 b, c), and petted vessels (figure 12 d).

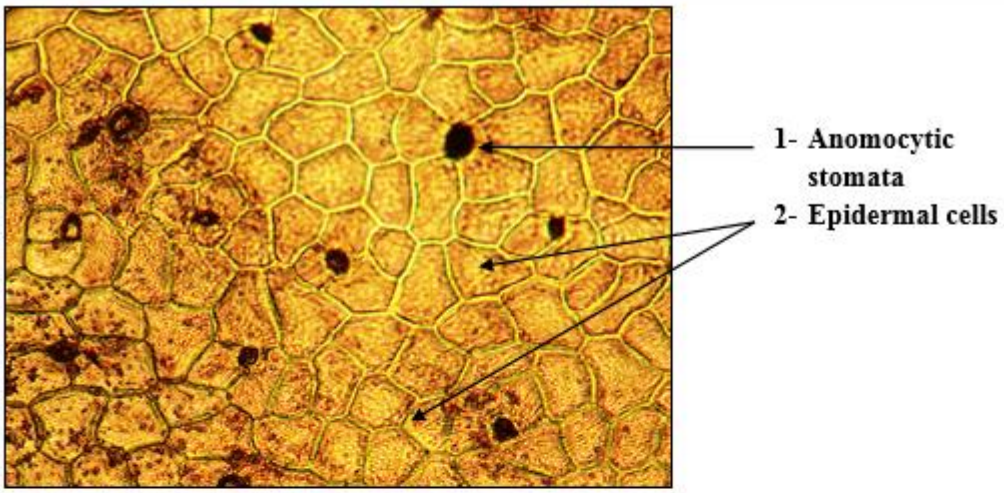

Figure 2: Surface view of upper epidermis of leaf (10x10)

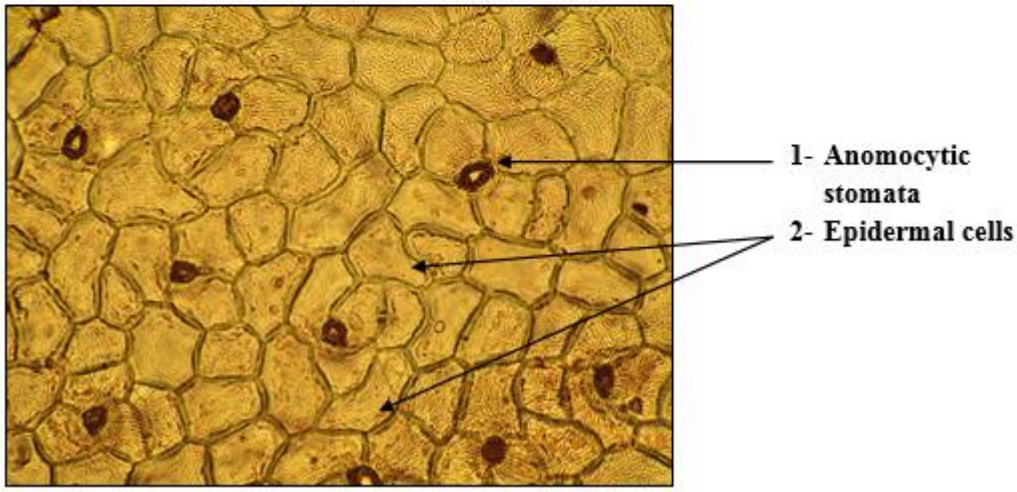

Figure 3: Surface view of lower epidermis of leaf (10x10)

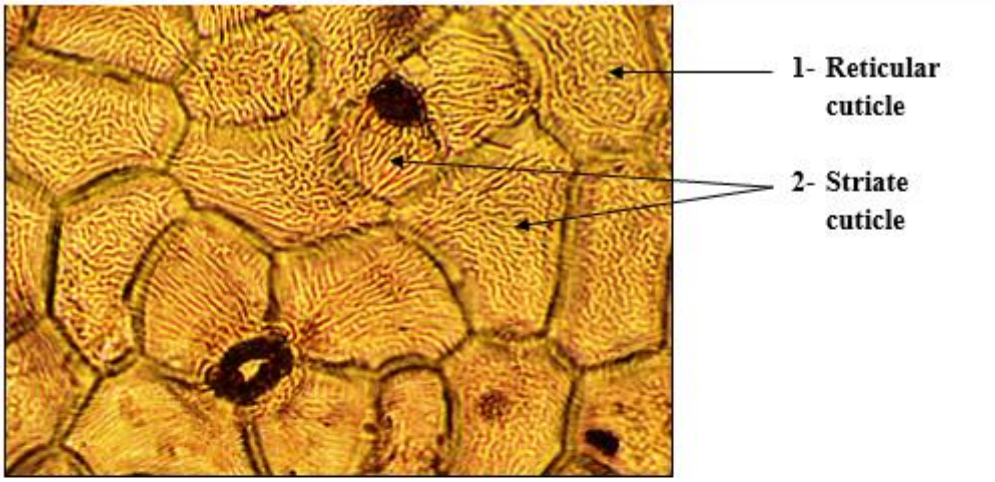

Figure 4: Stoma on of lower epidermis of leaf (10x40)

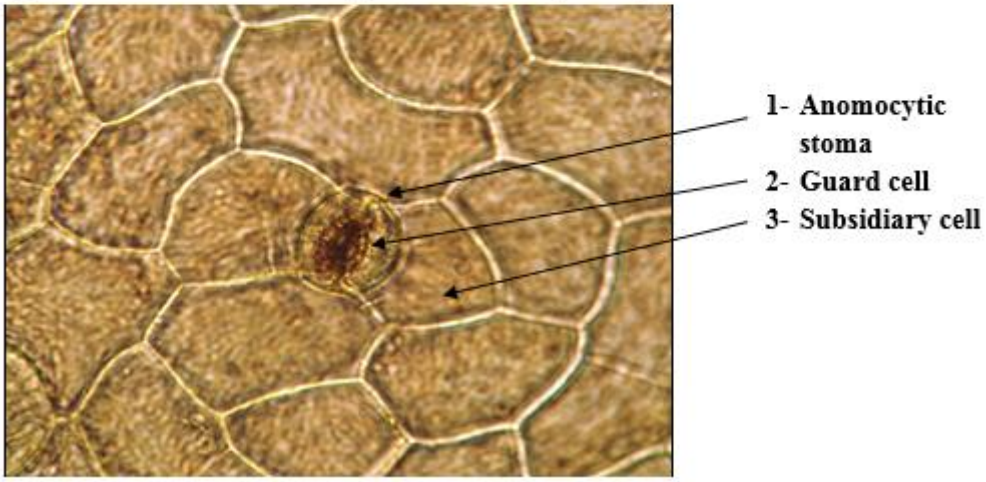

Figure 5: Stoma on epidermis of leaf (10x40)

Figure 6: Surface view of upper epidermis of leaf (10x40)

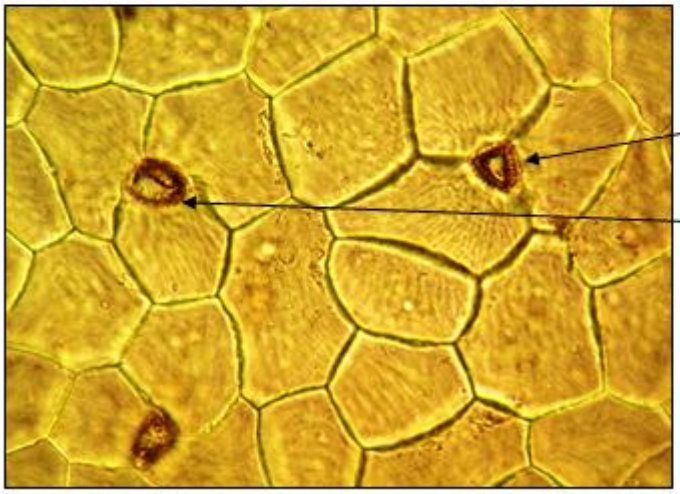

Figure 7: Stomata on upper epidermis of leaf (10x40)

Figure 8: Surface view of lower epidermis of leaf (10x40)

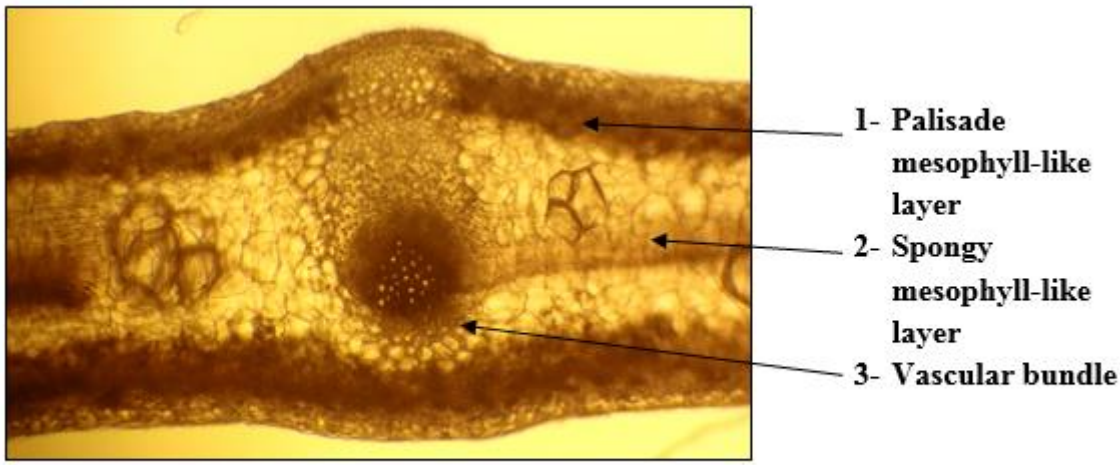

Figure 9: Transverse section of leaf (10x10)

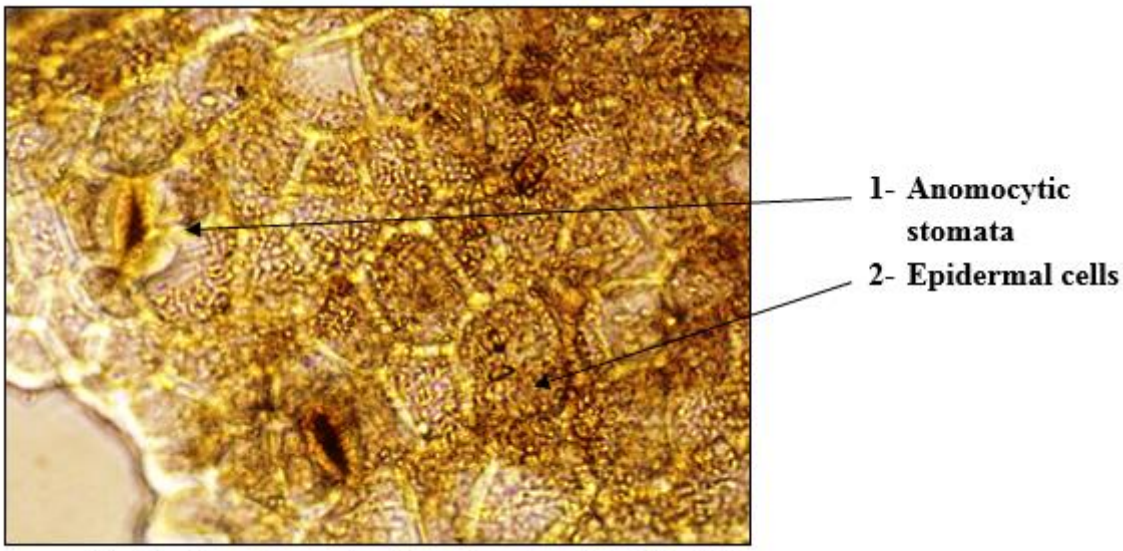

Figure 10: Surface view of epidermis of petiole (10x40)

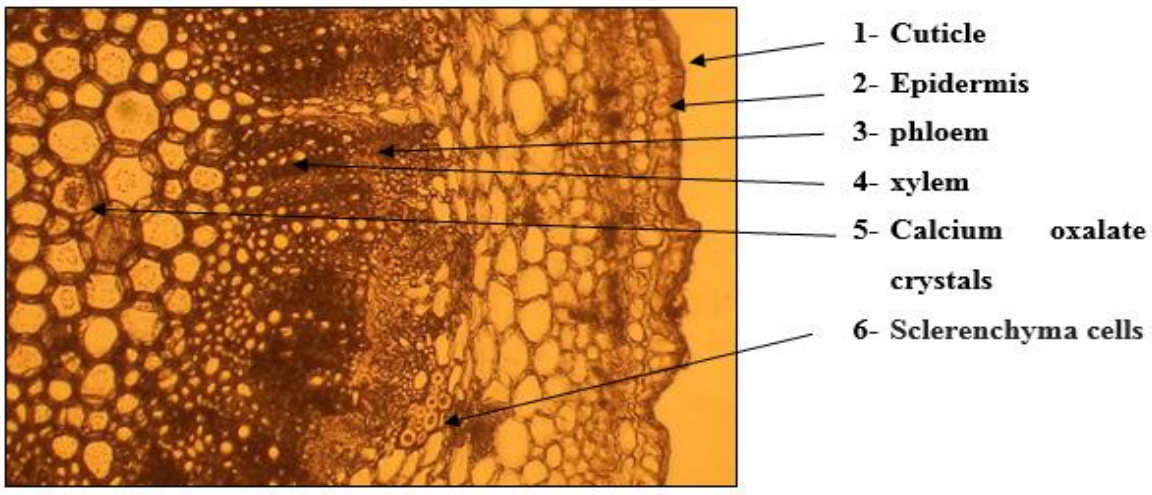

Figure 11: Surface view of epidermis of petiole (10x40)

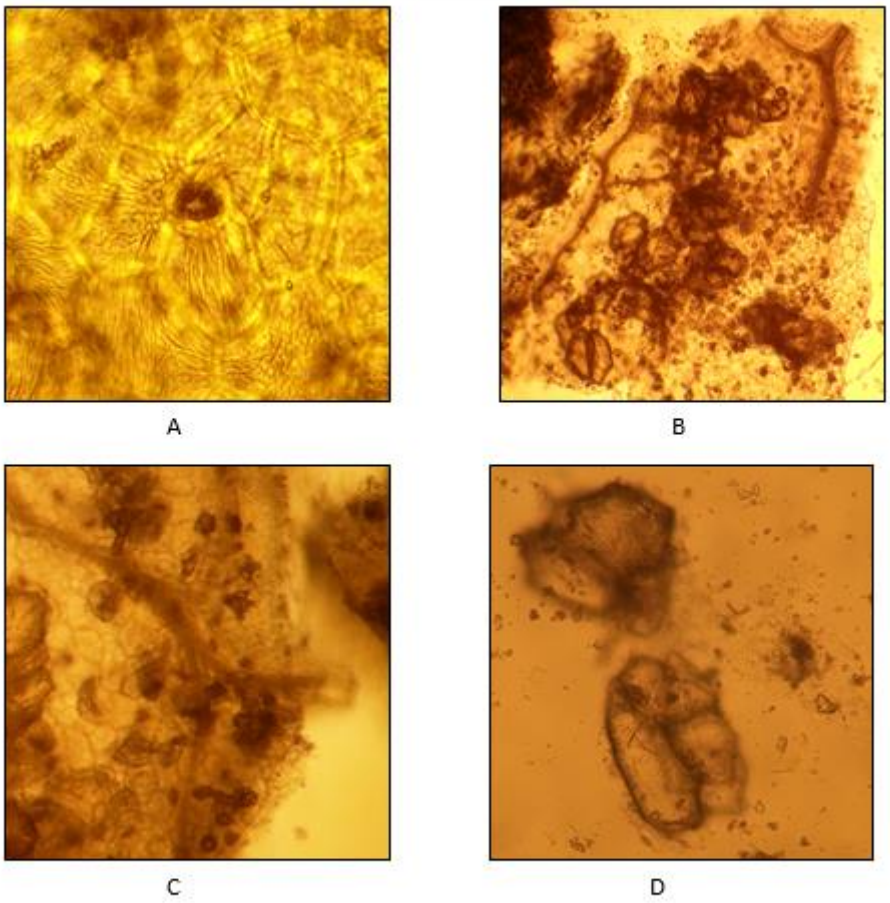

Figure 12: Powder microscopy: (a) Anomocytic stomata with triate and thick cuticle, (b) Fragment of midrib and lateral vein, (c) Fragment of epidermis and calcium oxalate crystals, (d) Pitted vessels.

Physicochemical analysis

The determination of Physico-chemical parameters is important in determination of adulterants and improper handling of powder. The total ash is particularly important in the evaluation of purity of drugs, i.e. the presence or absence of foreign inorganic matter such as metallic salts and/or silica [15, 16]. The extractive values are useful to evaluate the chemical constituents present in the crude drug and also help in estimation of specific constituents soluble in a particular solvent. High alcohol soluble and water soluble extractive values reveal the presence of polar substance like phenols, tannins and glycosides, as reported by [17, 18]. When moisture content of dry powder not very high, hence it would discourage bacteria fungi or yeast growth [5]. The results obtained from the physicochemical parameters are described in Table 1.

Fluorescence analysis of powdered drug

Fluorescence characteristic of any powdered drug is very distinctive and helpful in distinguishing features for the determination of the drug content [19]. The fluorescence analysis of the powder drug was also done and results are given in Table 2. The powder was treated with various reagents and the mixture was observed in daylight and under UV light to see the type of fluorescence.

Table 1: Physico-chemical parameter of leaves of *Capparis cartilaginea* Decne

Sr. No.	Parameters	Results (%w/w)
1	**Ash values**:	
	1) Total ash	6.10±0.05
	2) Acid insoluble ash	1.50±0.02
	3) Water-soluble ash	5.10±0.03
2	**Extractive value:**	
	1) Water soluble	36 ±0.06
	2) Ethanol soluble	13 ±0.05
3	**Moisture content** Loss on drying at 110ºC	9.5 ±0.03

Table 2: Fluorescence analysis of powdered *Capparis cartilaginea* Decne leaves.

S. No.	Treatments	Observations in		
		Daylight	**Short UV**	**Long UV**
1.	Powder as such	Yellowish Green	Dark brown	Dark brown
2.	Powder + 1N NaOH (aqueous)	Yellow	Dark yellow	Yellow
3.	Powder + 1N NaOH (alcoholic)	Yellowish brown	Dark brown	Dark brown
4.	Powder + 1N H_2SO_4	Light brown	Deep brown	Light green
5.	Powder + 50% N HNO_3	Orange	Brownish green	Dark green
6.	Powder + conc.HNO_3	Orange	Yellowish green	Light green
7.	Powder + dil. HNO_3 10%	Light green	Light green	green
8.	Powder + 1N HCl	Brownish green	Dark green	Dark green
9.	Powder + Ammonia	Brownish green	Green	Light green
10.	Powder + Acetic acid	Brown	Dark brown	Brown
11.	Powder + 5% Iodine	Reddish-brown	Dark brown	Dark brown
12.	Powder + 5% $FeCl_3$	Green	Florescent green	Green
13.	Powder + Methanol	Green	Dark brown	Brown
14.	Powder + water	Light green	Brown	Brown

CONCLUSION

Capparis cartilaginea Decne is used in folk medicine of Yemen for various illness. No pharmacognostic studies have been reported for this plant but biological evaluation is rare. The leaves were selected for the study with the aims: to establish pharmacognostic parameters in order to establish its identity and purity criteria.

The matured leaf is simple, entire, leathery and evergreen. It is petiolated (the petiole measures between 5 – 10 cm), an oval-shaped, broad and fleshy, often ending in a hooked. Result of microscopic examination of leaves showed that the epidermal cells with oval, hexagonal and polygonal shape, anomocytic stomata, no clearly distinct palisade and spongy mesophylls, cuticle appears to be prominent on both the upper and lower sides of leaf lamina and cluster crystals of calcium oxalate.

The physico-chemical constants like ash value (16.10 %), acid insoluble ash value (1.50 %), water soluble ash (5.10), alcohol extractive value (13.11 %) and water extractive value (36.40 %) and moisture content (9.5 ±0.03). Pharmacognostical evaluation is important to provide the standards and to avoid adulteration of drugs. Macroscopical and microscopical characters of the plant used for the identification of the drug. The fluorescence analysis helps in distinguishing the drug in powder form. However, a more extensive study is necessary such a pharmacological and toxicological analysis.

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