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A Study on the Standardization Parameters of a Halophytic Plant (*Cressa cretica* L.)

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Abstract: Now-a-days, herbal medication is proving to be better and safer mode of therapy of various diseases, without exhibiting any adverse effects as those with the allopathic medication. *Cressa cretica* L. belongs to the plant family Convolvulaceae. It is commonly called as Rudravanti. It is an erect, small, dwarf shrub, commonly grown in coastal areas. *Cressa cretica* is distributed throughout India, Timor and Australia. The present study provides updated information on its pharmacognostic, phytochemical analysis and pharmacological properties. Transverse sections of Cressa cretica root shows an upper epidermis followed by cortex containing cortical fibers and pith in center. It can be used medicinally as antibilious, antiviral, antitubercular, antidiabetic, expectorant, antiasthamatic, aphrodisiac, antibacterial. The presence of cortical fibers in stem, starch grains, aerenchyma in root are some of the distinguishing characteristics observed in *Cressa cretica*. It consists of flavonoids, quercetin, scopoletin, umbelliferone, stigmasterol, creticane, cressatetracosanoate, etc. It proved that *Cressa cretica* plant is one of the medicinally important plants.

Key words: *Cressa cretica* L. · Favonoids

INTRODUCTION

Cressa cretica L. belongs to the Convolvulaceae family [1]. It is an erect, small, dwarf shrub, usually grows in sandy and muddy saline habitats alongwith the species *Suaeda maritima*, *Salicornia europaea*, *Salsola soda*, *Limonium vulgare* subsp. *Serotinum* and *Crypsis aculeate* [2]. It is known as Rudanti in indigenous system of medicine in India and Molleich or Nadewa in Arabic [1].

Prior to any research on herbal medication, it is very crucial to estimate and analyze the standardization parameters of any medicinal plant. By standardization of the herbal drug; we will be able to utilize this lesser known drug in further research studies to estimate various pharmacological activities. *Cressa cretica* is a potentially active plant and is used as Anti-inflammatory, bronchodilator, antidiabetic, antiviral, antitubercular, aphrodisiac, stomachic, etc.

Geographical Source: *Cressa cretica* is a remarkable salt tolerant plant, common in coastal areas usually occurring in mono-specific stands along the landward edge of marshes. It is distributed throughout India, Timor and Australia [2].

Taxonomical Classification: The plant belongs to Kingdom - Plantae; Phyllum - Angiosperms; Class - Magnoliatae; Subclass - Asteridae; Order - Polemoniales; Family - Convolvulaceae; Genus - *Cressa* and Species - *Cretica.*

Synonyms: Sanskrit - Rudanti; Hindi -Rudravanti; Oriya - Dahna; Bengali - Rudravanti; Tamil - Uppusanaga; Telugu-Uppugaddi, Uppusenaga; Kannada-Mullumaddugida; Konkani- Chaval; Malayalam-Azhukanni and in Marathi - Lona, Rudravanti.

Medicinal Uses: *Cressa cretica* has been reported to possess antibilious, antitubercular, antiviral, antibacterial, antidiabetic, expectorant, anthelmintic, stomachic, aphrodisiac, enriches the blood, constipation, leprosy, asthma, urinary discharges [1]. It also exhibited cytotoxic, anti-inflammatory activity [3]. The dry leaves of *Cressa cretica* when crushed with sugar can be used as an emetic in Sudan [4].

Macroscopic Characters: *C. cretica* L. is an erect, small, dwarf shrub Roots are horizontal, geminate, with lateral branches leading upward to produce above-ground

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parts. It is a perennial sub shrub or herb, usually much-branched. Stems are at first erect and then become decumbent, apparently short-lived, gray appressed pilose to sericeous. Leaves on main branches are often larger than those on branchlets. The blade is 1-12 mm long, lanceolate, ovate or elliptic- to scale-like, sessile, or shortly petiolate. Flowers are solitary, white or pink, axillary, 5-8 mm long, sessile or on short peduncles, bracteates, in spicate to head-like clusters at tips of branchlets, bracteoles unequal in length. Sepals ovate to obovate imbricate. Corolla salver form, the limb 5-lobed, the lobes mostly ovate, imbricate, spreading to reflexed. Stamens exserted; filaments filiform; styles exserted. Ovary 2-locular, 4-ovulate; styles 2, distinct to the base; stigmas capitate. Fruit is capsular, ovoid, unilocular, 2-4-valved and usually one seeded. Seeds are 3-4 mm long, glabrous and smooth and shining to reticulate, dark brown [3].

Chemical Constituents: *Cressa cretica* consists of five flavonoids *viz.,* Quercetin, kaempferol-3-O-B -D-glucoside, quercetin-3-O-p-Dg lucosid, kaempferol-3-O-a-L-rhamnosyl+-(61) -O-p-D-glucoside and quercetin-3-O-o-L-rhamno--(+16)- B-D-glucoside (rutin) [6]. Cressatetracosanoate was isolated as colorless compound from hexane-petroleum ether (1:3) eluants. Cressanonacontanoic acid was obtained as a white amorphous powder from petroleum ether: benzene (3:1) eluants. Cressatetratriacontanoic acid was obtained as a colorless compound from petroleum ether:benzene (1:1) eluants. Cressatriacontanone was obtained as colorless crystalline mass from petroleum ether and benzene (1:3) eluants. Cressanaphthacenone was isolated as colorless crystals from benzene eluants [10]. It also contains terpenic compounds, syringaresinol-β-d-deglucoside, triacontanoic acid, stigmasterol, ursolic acids, β-amyrin and edible fixed oil. It also contains quercetin, n-octacosanol, scopoletin and umbelliferone [2].

MATERIALS AND METHODS

The shade dried whole plant of *Cressa cretica* L. was collected from the sandy shores along the mangrove creeks near Devanampattinam Beach, Cuddalore district in Tamilnadu and identified (specimen number- 74052) by taxonomist Dr. K. Ravikumar FRLHT Bangalore. A voucher specimen was preserved in the Department of Pharmacognosy MIET, Meerut for further reference. For Pharmacognostical studies plant material was stored in a solution containing 5ml formaldehyde, 5ml acetic acid and 90 ml 70% alcohol and rest of plant material was further size reduced and stored until use, in an air tight container. Free hand sections were taken for microscopical evaluation and studies were conducted as per standard methods. Iodine, Potassium iodide (IKI) & Aniline Blue in lactophenol was used for differential staining along with Phloroglucinol & HCl. Concentrated nitric acid (50%) with a pinch of potassium chlorate was used as the macerating fluid. Powdered drug was used for powder microscopy, moisture content, ash values, swelling index, foaming index, foreign organic matter, extractive values were carried out and fluorescence studies were carried out by treating 0.5 gm of powdered drug with different reagent and observation in color was made in visible light, U.V light of short (254nm) and long wavelength (365nm) under U.V chamber. Photomicrographs were obtained by compound binocular microscope OLYMPUS BX41and photomicrography was done using Olympus C7070 camera [3]

RESULTS AND DISCUSSION

Powder Characteristics

Microscopical Examination of Powder: Powder of the crude material was pale green colored, coarse and free flowing, it was unpleasant

- Some isolated epidermal cells in surface view, irregular outline,
- Stomata of paracytic type and anomocytic type;
- Parenchyma cells with crystals;
- Spirally thickened vessel members and tracheids; members short with horizontal end wall and simple perforation plate, length
- Round to ellipsoidal chloroplasts in parenchyma cells;
- Long, narrow, pitted fibres with pointed ends;
- Glandular as well as nonglandular trichomes and
- Columnar palisade cells [8].

Microscopical study to the leaf of *Cressa cretica* reveals that it is obilateral nature with palisade cells on both the upper and lower surfaces and covering trichomes on the upper surface of the leaf along with paracytic and anomocytic stomata which are found evenly dispersed in the mesophyll of the leaf.

Pharmacognostic Evaluation of the Plant: Air dried materials was used for quantitative determination of physicochemical values. Ash value, Loss on drying, extractive values, foaming index, foreign organic matter were also performed.

Fig. 1: A complete plant of Cressa cretica L.

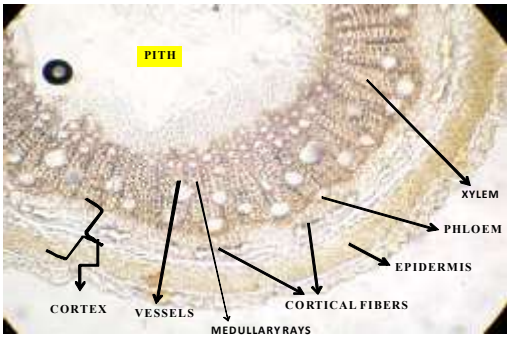

Fig. 2: T.S. of Root

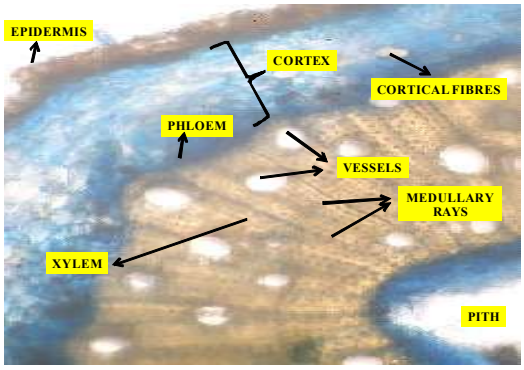

Fig. 3: T.S. of Root

Determination of Ash Value

Total Ash Value: Two grams of dried and powdered plant material was taken in the pre-weighed clean sintered silica crucibles. Then, they were incinerated by gradual increasing of the temperature (400-500 °C) in the muffle

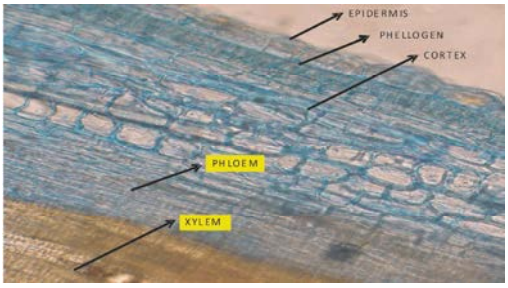

Fig. 4: RLS of Stem

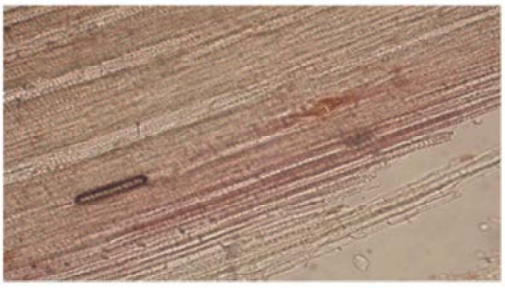

Fig. 5: RLS of Stem

Fig. 6: Vessels with annular thickenings and wide lumen.

Fig. 7: Polygonal parenchymatous cells of the epidermal cells

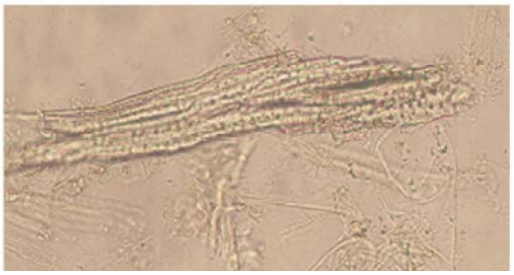

Fig. 8: Trachiedial fibres with simple pits.

Table 1: Ash values of *Cressa cretica* L.

S. No.	Parameters	Values
1	Total ash value	18.00%
2	Water - insoluble ash value	7.00%
3	Water - soluble ash value	11.00%
4	Acid- insoluble ash value	17.00%

Table 2: Extractive values of *Cressa cretica* L. [9].

S.No.	Solvent	Values
1	Water	5.8%
2	Ethanol	1.0%
3	Chloroform	0.8%
4	Chloroform - water	4.0%
5	Petroleum - ether	0.8%

furnace till white ash obtained until constant weight of ash obtained. The crucible was cooled to room temperature in a desecrator and weighed the ash and calculated the % of total ash with reference to air dried sample of the crude drug using following formula:-

$$Total\ Value\,(\%) = \frac{Z - X}{Y} \times 100$$

Where, Z= Weight of the crucible; X = Weight of the crucible with ash; Y = Weight of the powder taken (g). [13].

Determination of Extractive Values and Organoleptic Studies of Crude Extract: For the determination of Organoleptic characters such as color, nature, taste and yield of the extracts, 100g of dried and powdered plant material was successively extracted in the soxhlet extractor using petroleum ether, chloroform, ethanol (99%, v/v) and distilled water solvents in the increasing order of polarity for 24h. The Resulting liquid extracts were evaporated to dryness under reduced pressure. The yield of the extracts was calculated using the ollowing formula described by [11].

$$\text{Ectractive value}(\%) = \frac{\text{Weight of the Residue} \times 100}{\text{Weight of the plant material taken}}$$

Water Soluble Extractive Value: 5 gm coarse and air dried drug material was macerated with 100ml water in a stoppered flask for 24 hrs with frequent shaking for first 6 hrs. The extract was filtered rapidly through filter paper taking precaution to prevent excessive loss of solvent. The residue was evaporated in a flat bottom shallow dish, dried at 105°C weighed and kept in a desiccator. Average extractive value in Percentage w/w (on dry weight basis) was calculated with reference to air dried drug [12].

Chloroform Soluble Extractive Value: Accurately weighed 5 gm coarse and air dried powdered drug was macerated with 100ml chloroform in a stoppered flask for 24 hrs with frequent shaking for 6 hrs. It was then filtered rapidly through filter paper taking precautions to prevent excessive loss of chloroform. The volume was made up to 100ml with chloroform. The residue was evaporated in a flat bottom shallow dish, dried at 105°C, weighed and kept in desiccators. Average extractive value in percentage w/w (on dry basis) was calculated with reference to air dried drug.

Petroleum Ether Soluble Extractive Value: A 5 gm coarse and air dried drug material was macerated with 100ml petroleum ether in a stoppered flask for 24 hrs with frequent shaking for first 6 hrs. The extract was then filtered rapidly through filter paper taking precaution to prevent excessive loss of solvent. The residue was evaporated in a flat bottom shallow dish, dried at 105 °C weighed and kept in a desiccators. Average extractive value in Percentage w/w (on dry weight basis) was calculated with reference to air dried drug [11].

Acid Insoluble Ash Value: The total ash content of the plant material obtained was boiled for 15min, after adding 25ml of 25 % (v/v) HCl in to a 100 ml beaker and was allowed to cool. It was then filtered through a Whatman filter paper No. 44 (ash less) and wash the residue twice with hot water. The insoluble ash thus retained on filter paper along with paper was ignited in a preweighed sintered crucible (1000 °C). Then the crucible along with the residue was weighed and calculated the acid insoluble ash content using the following formula:-

$$\text{Acidinsolubleash Value}(\%) = \frac{a}{Y} \times 100$$

were,
a = weight of the residue;
Y= Weight of powder taken (g) [11].

Water Soluble Ash Value: The total ash value was determined using 2 g of the air-dried powdered sample. The total ash was boiled for 5 minutes with 25 ml of distilled water; the insoluble matter was collected on an ash less filter paper, washed with hot distilled water

Table 3: Fluorescence studies of *Cressa cretica* L. [7, 8].

S. No.	Material	Visible	Short (254 nm)	Long (365 nm)
1	1 N NaOH in Water	Brown	Green	Dark Brown
2	1 N HCl	Light Brown	Dull green	Dark brown
3	50 % H_2SO_4 (1M)	Grey	Green	Dark green
4	Chloroform extract	Dark brown	Dark green	Dark brown
5	Petroleum ether extract	Dark brown	Dark green	Brown
6	Picric acid	Light brown	Green	Dark brown
7	$FeCl_3$	Dark brown	Dark green	Brownish-black
8	Ammonia	Light brown	Dull green	Dark green
9	Iodine	Grey	Green	Dark brown
10	Methanol	Brown	Green	Dark grey

and ignited for 15 minutes at emperature ot xceeding 450°C. The weight of the insoluble matter was subtracted from the weight of the total ash; the difference in weight represents the water-soluble ash. The percentage of the water-soluble ash was calculated with reference to the air-dried powdered plant sample. It was calculated by using following formula [13].

$$\text{Waterinsolubleash Value}(\%)=\frac{a}{Y}\times 100$$

Where,
a = Weight of the residue;
Y= Weight of powder taken (g)

Water soluble ash Values (%) =
Total ash value - Water insoluble ash value

Fluorescent Studies of Powder Drugs: A lot of herbs show fluorescence when the cut surface or powder is exposed to UV light and this can be useful in their identification. The fluorescence character of the plant powders (40 mesh) was studied both in daylight and UV light (254 nm and 366 nm) and after treatment with different reagents like sodium hydroxide, hydrochloric acid, nitric acid and ferric chloride etc [14].

Loss on Drying: The loss on drying of the plant = 14 %.
Phytochemical Screening: The entire plant was collected and dried in shade and reduced to coarse powder. The powdered material was extracted with petroleum ether, chloroform, ethanol and water in Soxhlet apparatus. The extract was filtered hot and solvent removed by distillation under reduced pressure. The percentage yield

Chemical Tests:

S.No	Tests	Water	Ethanol	Petroleum Ether	Chloroform
.	Flavonoids				
1.	Shinoda Test	+	+	-	-
2.	Lead acetate solution	+	+	+	+
3.	Sodium hydroxide solution	-	+	+	+
4.	Ammonia Test	-	+	-	-
	Alkaloids				
1.	Dragendroff Test	+	+	+	+
2.	Wagner Test	+	+	+	+
3.	Mayer's Test	-	+	-	+
	Tannins				
1.	5%$FeCl_3$ solution	+	-	-	-
2.	Lead acetate solution	+	+	+	+
3.	Acetic acid	-	-	-	-
4.	Dil. HNO_3	+	-	-	+
	Proteins				
1.	Millon's Test	+	+	-	-
	Glycosides				
1.	Keller - Killiani Test	+	-	-	+
2.	Legal's Test	+	+	+	+
3.	Baljet's Test	+	+	+	+
	Saponins				
1.	Foam Test	_	_	_	_

was calculated and the extract was further subjected to phytochemical tests for Alkaloids, Glycosides, Flavonoids and Tannins

Foaming Index: No foam is present.

Foreign Organic Matter: No foreign organic matter is present.

CONCLUSIONS

Cressa cretica L. is an erect, small, dwarf shrub of Convolvulaceae family. It is commonly found in coastal areas. Medicinal plants have been traditionally used for the treatment of various diseases. *Cressa cretica* possesses antibacterial, antifungal, antitussive, testicular functions and antifertility activities. It is composed of flavonoids, heavy metals, lead, copper, zinc, thus, being one of the important medicinal plants. The estimation of the morphological characters, microscopical study, ash value, extractive value, loss on drying, fluorescence studies have been performed for the standardization and quality control of the medicinal plant.

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