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Pharmacognostical Investigation of *Ionidium suffruticosum* Ging. –A Seasonal Multipotent Medicinal Herb.

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ABSTRACT

Ionidium suffruticosum Ging. is a seasonal multipotent medicinal herb belongs to the family Violaceae. The plant claims highest medicinal values and used traditionally to treat diabetes, jaundice, male sterility, urinary tract infections, gonorrhea, bowel complaints, urinary problems and various ailments. No detailed pharmacognistical works are available, hence, in the present study, morphological, microscopical and preliminary phytochemical investigations of the herb were undertaken. All the parameters were studied according to the WHO & Pharmacopoeal guidelines. Anatomical studies showed that secondary growth occurs in roots and basal stocks of stem indicating the seasonal and perennial nature of the herb. Powder microscopy revealed that phloem fibres, xylem vessels with spiral, annular, scalariform lignifications and calcium oxalate crystals were abundant in dried material of the plant. The qualitative chemical tests of the petroleum ether, chloroform, methanol and aqueous extracts of plant material revealed the presence of alkaloids, steroids, triterpenoids, flavonoids & carbohydrates.

Key words: Pharmacognosy, Ionidium suffruticosum, Phytochemical screening, Violaceae.

INTRODUCTION

Ionidium suffruticsoum, Ging. Syn. *Hybanthus enneaspermus* (L). F. Muell. belongs to the family Violaceae, is a rare ethnomedicinal ephemeral herb¹ or under shrub (Resembling a shrub, especially in having basal woody stems and branches (Latin fruticosus means bushy stem). Plants are distributed in the tropical and subtropical regions of the world. It is an herb, found in the warmer parts of India. The plant is popularly called as Spade Flower (English), Purusharathna (Kannada) and Ratan purush (Hindi). Spade Flower is a perennial herb or small shrub of 40 to 60 cm height with a long slender tap root. The

leaves are sub sessile, linear, lanceolate, margin serrate, apex acute and stipules acuminate, 1-4 mm long. Pink-purple spade-shaped flowers solitary. Sepals 3-4 mm long. Lower petal broad spade-shaped, pink-purple with deep purple veins. Upper petals linear-oblong, 3-4 mm long; lateral pair 4.5-5 mm long. The fruits are capsules 4-9 mm long; ribbed seeds 5-12, pitted between ribs. Flowering is from June to November in India. Plants are found along river banks, open grasslands, sandy places and rocky regions. In nature (*in vivo*) the plants are seasonal and appear for few months. The roots and few basal stem stocks retaining in the soil and are regenerating during rainy season and soon after the rainy season the aerial part dries up and the plants disappear.

The plant is considered to have highest medicinal value and widely used by traditional healers to treat several diseases like diabetes, ² malaria (antiplasmodial activities ³), male sterility in Ivory Coast ⁴, urinary tract infections & water retention and is used as tonic ⁵. The tender leaf stalks are used as demulcent; the roots are antigonorrhoeic, diuretic, bowel complaints and urinary problems¹. Despite its multipotentiality as a medicine there are some limitations in the propagation of this species. Propagation of the species by seeds or any other conventional methods is not reliable because the plants are seasonal and available for short duration in nature. Moreover, the seeds are viable for short period and loose their viability within few weeks. The species are under threat due to their exploitation from their natural habitat by traditional healers, over grazing by animals, seasonal habitat and their short seed dormancy. At the same time, no data are available concerning its biological activities ^{6,7}. The present communication reports the macroscopic, microscopic, dry powder analysis in different solvents/reagents facilitates the correct identification of the dried plants or powdered drug which detects and prevents the adulteration(s), if any. The challenge ahead of this is to authenticate the therapeutic efficacy and safety of the plant, using standard methods.

MATERIALS AND METHODS

The studies were executed in the fresh specimens of *Ionidium suffruticsoum* collected from their natural habitat, local area of Hubli and were authenticated by one of the authors, Dr. M. Jayaraj. A voucher specimen has been deposited in the P. G. Department of Botany, Karnatak University, Dharwad for future reference. The external morphology of plant parts and other structural peculiarities were studies in the macroscopic observation. Anatomical characters of root, stem, leaf and stomatal characters were included in the microscopic investigations. The dried material of the plant was stored under normal environmental conditions. The Physico-chemical parameters such as extractive values, ash values, loss on drying were performed as per the official standard procedures. ^{12, 13} Microscopical investigations were made by microtome sections and powder microscopy was performed according to the prescribed procedure. ^{14,15}

The fresh parts of the plant are subjected to morphological characterization. Fresh hand cut sections of the root, stem and leaf were taken from the fresh material as per method described by Trease and Evans ⁸. T. S. of root, stem and leaf were stained with saffranin and Hematoxylin. The microphotographs were taken by trinocular microscope with digital Olympus Camera for detailed studies.

The plant material collected from their natural habitat was cleaned, shade dried at room temperature, coarsely powdered and stored in an air tight glass container. 100g of coarse powder was successively extracted with petroleum ether (40-60), chloroform and methanol in Soxhlet extractor for 18 hours. The extracts were filtered and concentrated

using rotary flash evaporator and residues were dried in desiccators over sodium sulfite below 60°C. Freshly prepared extract was subjected to phytochemical evaluation for the detection of various constituents using conventional protocol ⁹.

Physico-chemical parameters were determined as per Indian Herbal Pharmacopoeia and were analysed for moisture content, total ash, water sulphated ash, petroleum ether soluble extract, water soluble ash, alcohol soluble extract and water soluble extract ¹⁰. Preliminary phytochemical investigations of whole plant extract was carried out to know the medicinally important chemical constituents. The methanol, petroleum ether, chloroform and aqueous extracts of the plant were subjected to the different chemical tests to find out the nature of chemical constituents ¹¹.

RESULTS AND DISCUSSION

Pharmacognistical investigations:

The detailed and systematic pharmacognistical evaluation would give valuable information for the future studies. The detailed morphology of *Ionidium suffruticosum* Ging. was carried out to support proper identification of drug.

Stomata and stomatal index: The stomata are predominantly paracytic (Rubiaceous or parallel celled) and some are anisocytic (Cruciferous or unequal celled) (Fig. 1 D). The epidermal cells are larger than subsidiary cells. Stomatal index, the percentage of stomata found in unit area of leaf exhibited marked variation in the adaxial and abaxial surface of the leaf. Abaxial surface has an increased stomatal frequency than adaxial surface. The values are represented in the Table 1.

Table 1. Stomatal index on adaxial and abaxial leaf surfaces of *Ionidium* suffruticsoum.

Trials	Adaxial surface of		Abaxial surface of		
_	leaf		leaf		
	Margin	Middle	Margin	Middle	
1.	167	143	153	218	
2.	186	156	163	234	
3.	179	148	158	212	
4.	168	145	172	225	
5.	176	151	176	234	
6.	183	153	160	229	
Average	176.5	149.34	163.67	225.34	
	162.92		194	1.50	

Organoleptic, anatomical and powder microscopic study

The powder of *I. suffruticosum* is dull brown. It has no characteristic odour or taste. Microscopic examination (Fig. 2. A-J) of the powdered plant material indicated the presence of xylem vessels which are lignified, bordered pitted, spiral, annular and scalariform thickenings of varying length were associated with fibres in bundles. Apexes of the fibres were bluntly pointed. Thick walled xylem fibres and phloem fibres were present in large number. There are unbranched unicellular trichomes or hairs of specific

form and structures and function were noted. Powder microscopy showed the presence of testa of the seeds, thick and thin walled cork cells, cortex cells containing calcium oxalate crystals and medullary rays crossing across the cortex.

The T. S of fresh part of the root (Fig.1 A) showed the presence of bark, secondary cortex, annular rings, secondary medullary rays and compressed xylem at centre indicate the roots had secondary growth and perennial in nature The T. S. of fresh stem (Fig.1 B) revealed the presence of thick cuticle, compact layer of epidermis, trichomes, distinguished cortex, endodermis, semilunar sclerenchyma patches, a cambium tissue separating phloem outside and wider secondary xylem and primary narrow xylem cells towards inside. Abundant pith (parenchyma) encloses the centre. The T.S. of fresh leaf (Fig.1 C) showed the presence of the upper and lower epidermis enclosing the compact mesophyll. A distinct separating layer between the palisade and spongy parenchyma is a peculiar character.

Physicochemical and Preliminary Phytochemical Studies

Physical constant values like extract values, ash values and moisture content are tabulated in Table 2. Preliminary phytochemical tests of plant part extracts were conducted separately and detected for different medicinally important chemical constituents. The alcoholic extracts of the different organs of the plant i.e., roots, leaves, flowers, fruits and seeds were subjected to the different following chemical tests and found out the nature of constituents present in the plant organs (Table 3). Alcoholic extracts showed that roots, leaves, fruits and seeds contain steroids. Triterpenoids and flavanoids are present only in flower. Alkaloids are present in roots, leaves and flowers except fruits and seeds. Saponins, Proteins Glycosides, amino acids and Tannins are absent in plant. Carbohydrates are present in roots, leaves, fruits and seeds except flowers. Only the seeds contain fatty acids (Table 4).

Table 2. A. Extractive values B. Moisture content and C. Ash values of *Ionidium* suffruticosum (whole plant).

Sl.	Parameter	Determined
No.		value (%)
A	Extractive values	
1	Alcohol soluble extractive value	5.60
2	Water soluble extractive value	11.88
3	Ether soluble extractive value	8.0
В	Moisture Content	8.74
\mathbf{C}	Ash Values	
1	Total ash	18.50
2	Water soluble ash	2.97
3	Sulfated ash	23.50

Table 3. Qualitative chemical analysis of various extracts of *I. suffruticosum* Ging.

Sl.	Tests	PE	CHCl ₃	MeOH	AQ
No.					
1.	Alkaloids	-	+	+	-
2.	Glycosides	-	-	-	-
3.	Steroids	+	-	+	+
4.	Triterpenoids	-	+	-	-
5.	Saponins	-	-	-	-
6.	Flavanoids	-	+	+	-
7.	Tannins	-	-	-	-
8.	Carbohydrates	+	-	+	+
9.	Amino acids	-	-	-	-
10.	Fatty acids	+	-	-	-

Note: PE=Petroleum Ether, CHCl₃= Chloroform, MeOH = Methanol, AQ=Aqueous; + indicate presence – indicates absence

Table 4. Qualitative chemical analysis of alcoholic extracts of different parts of *I. suffruticosum*, Ging.

S1.	Tests	Roots	Leaves	Flower	Fruits	Seeds
No.						
1.	Alkaloids	+	+	+	-	-
2.	Glycosides	-	-	-	-	-
3.	Steroids	+	+	-	+	+
4.	Triterpenoids	-	-	+	-	-
5.	Saponins	-	-	-	-	-
6.	Flavanoids	-	-	+	-	-
7.	Tannins	-	-	-	-	-
8.	Carbohydrates	+	+	-	+	+
9.	Amino acids	-	-	-	-	-
10.	Fatty acids	-	-	-	-	+

Note: + = Present - = Absent.

Conclusions

Pharmacognistical analysis of *I. suffruticosum* showed many important features useful for the identification of the drug plant. Macroscopic characters like secondary growth in root and basal stems depicting the name suffruticosum bearing bushy nature although it is a herb. Presence of single row of sub epidermal collenchyma and chlorenchymatous cortex are well indicating the herbaceous character of the plant. Massive sclerenchymatous patches, abundant xylem vessels and phloem fibres are unique to plant. Hypostomatic leaves, flat upper surface and semicircular lower surface of the midrib were the important diagnostic feature of the drug plant. Predominantly paracytic and some anisocytic type of stomata are useful identification marks of the plant. High stomatal frequency establishes the xerophytic and ephemeral character of *I. suffruticosum*. Morphological and anatomical studies revealed that the plant is seasonal and perennial herb available for short period to traditional healers and for other preparation. Due its high

medicinal values the plant is in continuous exploitation. The presence of alkaloids, steroids, Triterpenoids, Flavanoids and carbohydrates (Table No.3&4) suggests that *Ionidium suffruticosum* has valuable and medicinally important chemical ingredients in its different organs. It is present need to conserve the plant for medicinal usage. Tissue culture techniques may be more useful in the conservation point of view.

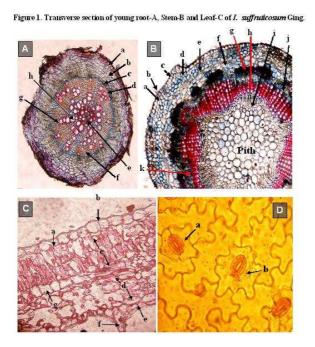


Figure 1. Transverse section of young root-A, Stem-B and Leaf-C of *I. suffruticosum* Ging.

Legend: Fig. 1. A – T.S. of young root a-epidermis, b-cortex, c-endodermis, d-pericycle, e-medulla filled with xylem tissue, f-phloem, g-protoxylem, h-metaxylem. **B**- T.S. of stem, a-epidermis, b-stem hair, c-cuticle, d-hypodermis, e-cortex, f-endodermis, g- sclerenchyma patches, h-secondary xylem, i-primary xylem, j-secondary phloem, k-vascular cambium. **C** – T.S. of leaf, a-upper epidermis, b-cuticle, c-palisade parenchyma, d- spongy parenchyma, e- lower epidermis, f- leaf hair, g-separating layer between palisade and spongy parenchyma. **D**- Surface view of epidermis showing a-paracytic stomata and b-anisocytic stomata.

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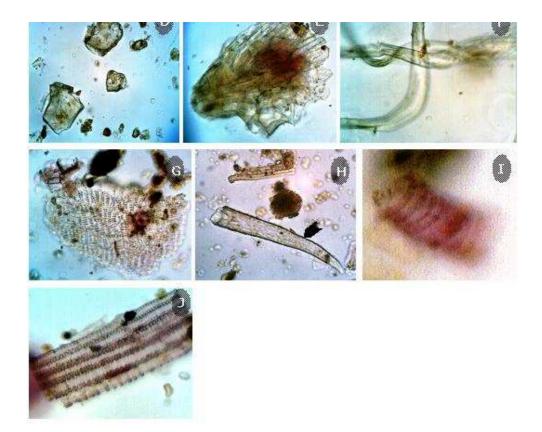


Figure 2: Microphotographs of powder microscopy.

Legend: Fig.2 A-phloem fibres **B**-Part of lamina with upper palisade and lower spongy parenchyma **C**-cortex of stem **D**-crystal forms of calcium oxalate **E**-testa **F**-medullary rays **G**-medullary along with sieve tissue **H**-covering trichomes **I**- spiral thickenings of vessel **J**-annular, spiral, reticulate & bordered pitted vessels.

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