



Pharmacognostic and phytochemical screening of *Crinum Asiaticum* and *Pedaliium Murex*

P. Vishnu Priya^{1*}, Dr. A. Srinivasa Rao²

¹ Assistant Professor, Department of Biotechnology, Joginpally B.R Pharmacy College, Yenkapally (V), Moinabad (M), Hyderabad-500 075, Telangana, India

² Principal, Professor, Department of Pharmacology, Bhaskar Pharmacy College, Yenkapally (V), Moinabad (M), Hyderabad-500 075, Telangana, India

ABSTRACT

Pharmacognostic studies mainly helps in plant identification and it lays the standards parameters which not only help but also prevents adulterations. Pharmacognostic studies will help in authentication of the medicinal plants and ensures in reproducible quality of plant products which lead to efficacy and safety of plant material. So, an attempt was made to study pharmacognostic parameters and phytochemical screening of *Crinum asiaticum* and *Pedaliium murex*. The pharmacognostic parameters mainly include macroscopical, powder microscopy, quantitative leaf microscopy (stomatal number, stomatal index, vein – islet number and vein termination number and palisade ratio), physiochemical parameters (total ash, water soluble ash, acid insoluble ash, water soluble extractive value, acetone soluble extractive value and alcohol soluble extractive value), fluorescent analysis and phytochemical screening of ethanol, acetone and aqueous extracts of *Crinum asiaticum* and *Pedaliium murex*.

Keywords: *Crinum asiaticum*, *Pedaliium murex*, ethanol, acetone and aqueous

INTRODUCTION

Now-a-days the drugs obtained from natural origin have renewed interest because they are mainly considered to be safe and are free from undesirable side effects. The main factor which emphasizes regarding synthetic drugs is the incidences of undesirable side effects which are harmful not only to human beings but also to environment. The advantage of plant based drugs is their readily availability, less or no side effects and economical. The disadvantage is that they are many sources of adulteration. The demand for plant based drug increases based on its effectiveness. In order to meet the growing demand of plant based drug, it is usually adulterated with low grade plant materials. The pharmacognostic study of medicinal plants has been widely felt very important in recent times. Pharmacognostic studies mainly helps in plant identification and it lays the standards parameters which not only help but also prevents adulterations.

Crinum asiaticum is a plant species widely planted in many warmer regions as an ornamental. It is a bulb forming perennial flowering plant. It is native of China, India, South Korea, Assam and Bangladesh belonging to family Amaryllidaceae. *Pedaliium murex* is an annual plant species widely planted in many warmer regions as an ornamental. It is a bulb forming perennial flowering plant. It is distributed in India, Srilanka and Tropical Africa belonging to family Pedaliaceae.

RESEARCH METHODS

Plant identification

The *Crinum asiaticum* and *Pedaliium murex* plants was identified and authenticated by plant taxonomist Dr. K. Madhava Chetty, Assistant Professor, Department of Botany, Sri Venkateshwara University, Tirupathi, Andhra Pradesh, India. The plants were collected at Kapalitheertham forest, Andhra Pradesh, India.

Collection of the leaves

After identification and confirmation of the plants by plant taxonomist Dr. K. Madhava Chetty, the leaves were separated from the *Crinum asiaticum* and *Petalium murex* plants. The plant material was shade dried, without exposing the material to direct sunlight. After drying the leaves were powdered separately in mixer. The powder was then passed through sieves no: 40. This fine leaf powders were stored separately in a cool and dry place until its use.

Preparation of leaf extracts

For the preparation of various leaf extracts 250 g of plant materials were separately extracted using ethanol, acetone and aqueous solvents using soxhlet extractor at a temperature not exceeding 45°C. The extracts were concentrated and dried by using rotary evaporator and was stored in a refrigerator at 4°C until use.

Instruments

Electron microscope, camera lucida, stage micrometer, eye piece micrometer and UV spectroscopy.

PHARMACOGNOSTIC STUDIES

Macroscopical characters¹

It is the simplest means to establish the identity and to ensure quality of a particular drug. The macroscopic characters of medicinal plants are observed by naked eye or through magnifying lens. The following macroscopic characters for the fresh leaves were noted: size, shape, colour, surfaces, venation, presence or absence of petiole, apex, margin, base, odour and taste are evaluated. The results are listed in table: 1 & 2

Powder microscopic studies²

Dried powders of *Crinum asiaticum* and *Petalium murex* were observed under microscope. Powder study is similar to microscopic study except here dried powder is taken instead of section of the plant material. All the reagents used are also same like above. The results are furnished in fig: 3 to 10

Quantitative leaf microscopy^{3,4}

It includes palisade ratio, stomata number, stomata index, vein-islet number and vein termination number in epidermal strips. The results are presented in table: 3 & 4

Stomatal number

It is the average number of stomata per square mm of the epidermis of the leaf.

Stomatal index

It is defined as the percentage number of stomata as compared to all the epidermal cells (including stomata) in a unit area of leaf.

It is calculated by using this formula:

$$S. I = \frac{S}{E + S} \times 100$$

Where,

S. I = Stomatal Index,

S = No. of stomata per unit area,

E = No. of epidermal cells in the same unit area

Palisade ratio

It is the average number of palisade cells beneath each epidermal cell.

Vein-islet number

It is determined by counting the number of vein-islets in an area of 1 square mm of the central part of the leaf between the midrib and the margin.

Vein termination number

Vein termination number is defined as the number of vein terminations per square mm of the leaf surface, midway between mid-rib of the leaf and its margin.

Physico-chemical analysis^{5,6}

The parameters which are studied are moisture content, loss on drying, total ash, acid-insoluble ash, alcohol and water-soluble extractive values, petroleum ether soluble extractive value, ethyl acetate soluble extractive value, acetone soluble extractive value, etc. The results are furnished in table: 5 & 6.

Fluorescence analysis⁹

A small quantity of dry plant powder is placed on grease free clean microscopic slide and 1-2 drops of freshly prepared reagent solution is added, mixed by gentle tilting the slide and wait for few minutes. Then the slide is placed inside the UV chamber and observe the colour in visible light, short (254 nm) and long (366 nm) ultra violet radiations. The colour observed by application of different reagents in different radiations is recorded. Generally the colour change is noted in reagents like Powder + 5% NaOH, Powder + 5% KOH, Powder + 5% FeCl₃, Powder + Conc. H₂SO₄, Powder + Conc. HCL, Powder + Conc. HNO₃, Powder + iodine, Powder + KMnO₄, Powder + dil. NH₃, Powder + Na₂CO₃, etc. The results are listed in table: 7 & 8

Preliminary phytochemical analysis^{7, 8}

The various extracts of *Crinum asiaticum* and *Pedaliium murex* are tested for various phytoconstituents by standard procedures. They are generally tested for the presence of alkaloids, flavonoids, tannins, phenols, cardiac glycosides, triterpenes, steroids and saponins. The results are furnished in table: 9 & 10.



Figure 1: *Crinum asiaticum* plant.



Figure 2: *Pedaliium murex* plant.

RESULTS AND DISCUSSION

Crinum asiaticum

Crinum asiaticum is a ornamental, perennial herb widely grown in warmer region. Leaves are simple, linear in shape, with entire margin, parallel venation and arranged in a rosette. Leaves are typically 2-3 feet long and 5 inches wide.

Table 1: Morphological characteristic of *Crinum asiaticum* leaves.

Characters	Leaves
Colour	Green
Odour	Faint
Taste	Characteristic

Pedaliium Murex

Pedaliium murex is a ornamental plant species widely grown in warmer region. It is perennial plant, bulb forming with showy flowers. All parts of the plant are poisonous.

Table: 2 Morphological characterization of *Pedaliium murex*.

Characters	Leaves
Colour	Green
Odour	Characteristic odour
Taste	Taste less

Powder microscopic characters of *Crinum asiaticum* and *Pedaliium murex*

Powder of *Crinum asiaticum* is usually green in colour, contains acicular type of calcium oxalate crystals, diacytic stomata, stomata containing epidermal cells, starch grains are also present. Powder characteristics of *Crinum asiaticum* are shown in fig: 3, 4, 5 & 6.

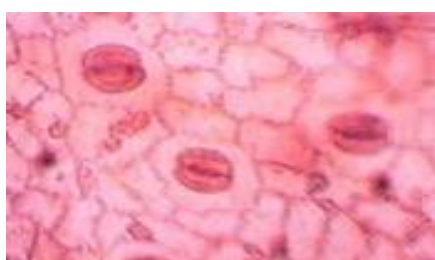


Figure 3: Diacytic stomata.



Figure 4: Acicular type of calcium oxalate crystals.

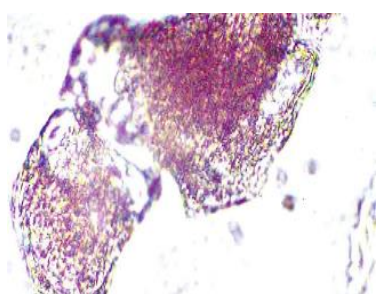


Figure 5: Parenchyma cells.

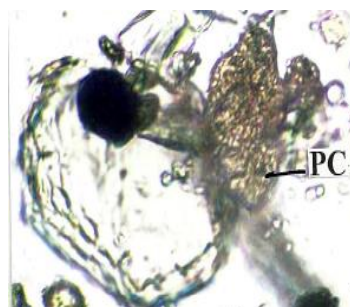


Figure 6: Starch grains and granular parenchyma cells.

Powder of *Pedaliium murex* is usually green in colour, contain calcium oxalate crystals, anisocytic stomata, stomata containing epidermal cells, parenchyma cells with granular contents and starch grains are also present. Powder characteristics of *Pedaliium murex* are shown in fig: 7, 8, 9 & 10.

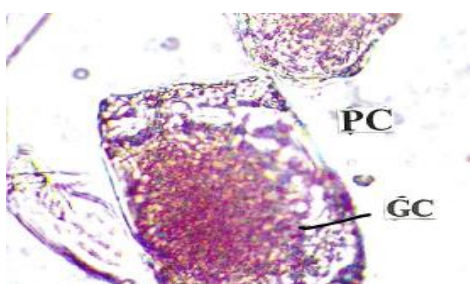


Figure 7: Parenchyma cells with granular contents.

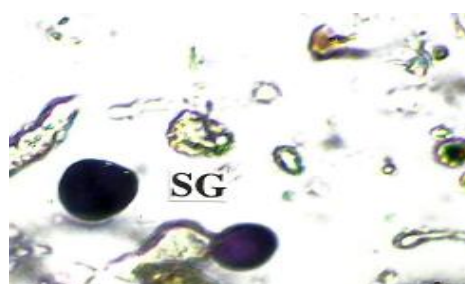


Figure 8: Starch grains.



Figure 9: Calcium oxalate crystals.

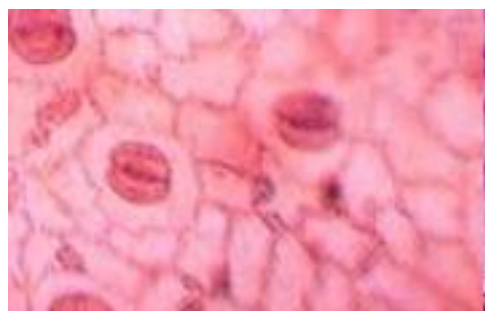


Figure 10: Anisocytic stomata.

Results of quantitative microscopic evaluation

Table 3: Determination of leaf constants of *Crinum asiaticum*.

Leaf constants	<i>Crinum asiaticum</i>
Stomatal number/sq mm (Upper epidermis)	4.5
Stomatal number/sq mm (Lower epidermis)	5.2
Stomatal index (Upper epidermis)	9.05
Stomatal index (Lower epidermis)	8.2
Palisade ratio	4.3
Vein islet number/sq mm	2.1
Vein termination number/sq mm	4.3

Table 4 Determination of leaf constants of *Pedaliium murex*.

Leaf constants	<i>Pedaliium murex</i>
Stomatal number/sqmm (Upper epidermis)	5.3
Stomatal number/sqmm (Lower epidermis)	5.9
Stomatal index (Upper epidermis)	7.79
Stomatal index (Lower epidermis)	7.02
Palisade ratio	5.2
Vein islet number/sq mm	2.5
Vein termination number/sq mm	4.2

Table 5: Physicochemical parameters of *Crinum asiaticum* leaves.

Physicochemical properties	<i>Crinum asiaticum</i> values
Total ash (% w/w)	23
Acid insoluble ash (% w/w)	8.2

Water soluble ash (% w/w)	17.2
Water soluble extractive value (% w/w)	30.1
Acetone soluble extractive value (% w/w)	21.7
Alcohol soluble extractive values (% w/w)	20.6
L.O.D	18
p ^H	5.8

Table 6: Physicochemical parameters of *Pedaliium murex* leaves.

Physicochemical properties	<i>Pedaliium murex</i> values
Total ash (% w/w)	24.4
Acid insoluble ash (% w/w)	10.2
Water soluble ash (% w/w)	17.24
Water soluble extractive value (% w/v)	32.5
Acetone soluble extractive value (% w/v)	20.6
Alcohol soluble extractive value (% w/v)	22.8
L.O.D	8
p ^H	5

Table 7: Fluorescence analysis *Crinum asiaticum* leaf powder.

Treatment	Normal light	Under UV at 254 nm	Under UV at 366 nm
Dry powder	Light Green	Green	Green
Powder + 5% NaOH	Yellowish green	Greenish brown	Dark brown
Powder + 5% KOH	Light yellow	Light greenish brown	Light brown
Powder + 5% FeCl ₃	Yellowish green	Dark brown	Dark black
Powder + Conc. HCL	Dark brown	Black	Black
Powder + Conc. H ₂ SO ₄	Reddish brown	Dark black	Dark black
Powder + Conc. HNO ₃	Orange	Yellow	Dark yellow
Powder + dil. NH ₃	Light orange	Dark brown	Yellow
Powder + Na ₂ CO ₃	Yellowish brown	Yellowish brown	Yellow
Powder + NH ₄ OH	Dark brown	Dark orange	Yellow
Powder +1% KMnO ₄	Dark brown	Black	Black
Powder + AgNO ₃	Yellow	Light yellow	Light yellow
Powder + iodine	Greenish brown	Dark black	Dark brown

Table 8: Fluorescence analysis *Pedaliium murex* leaf powder.

Treatment	Normal light	Under UV at 254 nm	Under UV at 366 nm
Dry powder	Green	Brown	Brown
Powder + 5% NaOH	Light yellow	Greenish brown	Dark brown
Powder + 5% KOH	Yellow	Greenish brown	Brown
Powder + 5% FeCl ₃	Yellowish green	Dark brown	Dark black
Powder + Conc. HCL	Dark brown	Brown	Black
Powder + Conc. H ₂ SO ₄	Reddish brown	Dark black	Dark black
Powder + Conc. HNO ₃	Yellow	Yellow	Dark yellow
Powder + dil. NH ₃	Orange	Light brown	Light brown
Powder + Na ₂ CO ₃	Yellowish brown	Yellowish brown	Yellow
Powder + NH ₄ OH	Brown	Dark orange	Dark Yellow
Powder +1% KMnO ₄	Yellowish brown	Black	Black
Powder + AgNO ₃	Yellow	Yellow	Light orange
Powder + iodine	Greenish brown	Dark black	Dark black

Table 9: Phytochemical analysis of various extracts of *Crinum asiaticum*.

S.No	Constituents	Ethanol	Acetone	Aqueous
1	Carbohydrates	+	+	+
2	Alkaloids	+	+	+
3	Cardiac glycosides	-	-	-
4	Anthroquinone glycosides	-	-	-
5	Proteins	-	-	-
6	Flavonoids	+	+	+
7	Saponins	-	-	+
8	Tannins	-	-	-
9	Sterols	+	+	-

Present (+) Absent (-)

Table 10: Phytochemical analysis of various extracts of *Pedaliium murex*.

S.No	Constituents	Ethanol	Acetone	Aqueous
1	Carbohydrates	+	+	+
2	Alkaloids	+	+	+

3	Cardiac glycosides	-	-	-
4	Anthroquinone glycosides	-	-	-
5	Proteins	-	-	-
6	Flavonoids	+	+	+
7	Saponins	-	-	-
8	Tannins	-	-	-
9	Sterols	+	+	-

Present (+) Absent (-)

CONCLUSION

The pharmacognostic studies are mainly helpful for identification and assessing the purity of *Crinum asiaticum* and *Pedaliium murex*. So, plants are evaluation for macroscopic, powder microscopic, quantitative powder microscopy, physicochemical analysis, florescence analysis and phytochemical analysis using standard procedure. The results of *Crinum asiaticum* and *Pedaliium murex* are helping in establishing quality control standards and purity assurance of pharmacognostic studies will also be helpful for assessing the purity of raw material. Briefly, the results described here can be considered as important characteristics to identify and authenticate *Crinum asiaticum* and *Pedaliium murex*.

ACKNOWLEDGEMENT

The authors are grateful to Sri. J. Bhaskar Rao, Chairman, JB Educational Society, Moinabad, for providing research environment as well as necessary support to carry out this work.

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