Vol 13 | Issue 2| 2022 | 38-47.

e - ISSN 2249-7544
Print ISSN 2229-7464

INTERNATIONAL JOURNAL
OF
PHYTOPHARMACY RESEARCH
www.phytopharmacyresearch.com

PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATION ON AERIAL PARTS OF CYPERUS IRIA.L

Malavika T M[1]*, Sarath lal P.S[2], Ajith Babu T K[2], Poornima M[2], Dias Dileep E[2]

[1]Department of Pharmacognosy, Nirmala College of Health Science, Chalakudy, Thrissur
[2]Department of Pharmacognosy, Malik Deenar College of Pharmacy, Seethangoli, Bela post, Kasaragod.

ABSTRACT
Cyperus iria is the plant that comes under the category of sedges. Cyperus species were traditionally used to treat various clinical conditions. It is one of the most widespread, problematic and economically damaging agronomic weeds, growing widely in various tropical and subtropical regions of the world. It is a weed plant commonly occurred in rice fields known as rice flatsedge. The present study highlights the pharmacognostic and phytochemical studies on aerial parts of Cyperus iria. The pharmacognostic studies like moisture content, ash values, extractive values, histology and powder analysis were carried out. Successive solvent extraction and phytochemical screening was carried out. The extracts showed the presence of alkaloids, phenols, steroids, glycosides, flavonoids, terpenoids, carbohydrates and saponins.

Keywords: Population, Resistance, Histology, Pharmacognostic and Subtropical.

INTRODUCTION

Traditional system of medicine continues to be widely practiced on many accounts. Population rises, inadequate supply of drugs, prohibitive cost of treatments, side effects of several synthetic drugs and development of resistance to currently used drugs for infectious diseases have led to increased emphasis on the use of plant materials as a source of medicines for a wide variety of human ailments [1]

Traditional herbal products are heterogenous in nature. They impose a number of challenges to qualify control, quality assurance and regulatory process. Most herbal products on the market today have not been subjected to drug approval process to demonstrate their safety and effectiveness [2].

Phytochemicals are chemical compounds produced by plants, generally to help them resist fungi, bacteria and plant virus infections, and also consumption by insects and other animals. Some phytochemicals are poisons, in their low doses used as medicine [3]. The phytochemical studies include extraction and isolating compounds from the origin plant, followed by defining their structure and therapeutic potential in laboratory model systems, such as cell cultures, in vitro experiments, or in vivo studies using laboratory animals. Challenges in that field include isolating specific compounds and determining their structures, which are often complex, and identifying what specific phytochemical is primarily responsible for any given biological activity [4]. As the target compounds may be non-polar to polar and thermally labile, the suitability of the methods of extraction must be considered. Various methods, such as sonification, heating under reflux, soxhlet extraction and others are commonly used for the plant samples extraction. In addition, plant extracts are also prepared by maceration or percolation of fresh green plants or dried powdered plant material in water and/or organic solvent systems. The other modern extraction techniques include solid-phase micro-extraction, supercritical-fluid extraction, pressurized-liquid extraction, microwave-assisted extraction, solid-phase extraction, and surfactant-mediated techniques, which possess certain advantages [5].

PLANT PROFILE

Description

Cyperus iria. L is a tufted annual herb or occasionally perennial, with fibrous roots and height varies from 8 to 60 cm. Numerous, short, sharply 3 angled, smooth, yellowish red stem with 5-80 cm high and leaf is linear- lanceolate, basal, rough to touch in upper part, flaccid with gradually tapering point and 3-8 mm wide in figure 1 [6].

Synonym - Chlorocyperus iria Rikl, Cyperus microiria Steud

Distribution - China, Japan and Korea, Cambodia, India, Indonesia

Common Names - Rice flatsedge, in India - morphula

Corresponding Author: **Malavika T M** Email:- malavikatm1995@gmail.com

Taxonomic Representation

Domain : Eukaryota

Kingdom : Plantae
Phylum : Spermatophyte
Sub Phylum : Angiospermae
Class : Monocotyledonae
Order : Cyperales
Family : Cyperaceae
Genus : Cyperus
Species : *Cyperus Iria*

MATERIALS AND METHODS

Plant Collection and Authentication

The aerial parts of *Cyperus iria* was collected from Thrissur. The plant material was aunthentified by the botanist, Dr. Jose Kutty E J, HOD, Dept. of Botany, Government College, Kasaragod. The plant materials were dried under shade for few days, powdered with mechanical grinder and stored in an air tight container.

Pharmacognostic Studies

Determination of moisture content

Five grams of the powdered aerial parts of plant were placed in tared evaporating dish. Drying was carried out at 105˚C for five hours. The drying was continued with intermittent weighing at one-hour interval until difference between two successive weighing was not more than 0.25%. Constant weight was reached when the two-consecutive weighing after drying for 30minutes and cooling for 30 minutes in desiccator, showed not more than 0.01gm difference [7].

Loss on drying = Initial weight – Final weight

$$\% \text{ Loss on drying} = \frac{\text{Loss on drying}}{\text{Weight of powdered drug}} \times 100$$

Determination of ash value

The ash value is an important parameter for the evaluation of crude drugs, due to variation of values within wide limit. The ash value of any organic material is composed of inorganic material like metallic salt and silica [8].

The following three methods were developed;

- Total ash
- Acid insoluble ash
- Water soluble ash

Total ash

Two grams of ground air dried material were accurately weighed out in a crucible previously ignited for 30 minutes. The material was spread in an even layer and ignited at a temperature more than 450˚C until it was indicating the absence of carbon, cooled in the desiccator and weighed. Calculated the content of total ash per gram of air-dried material [9].

$$\% \text{ Total Ash} = \frac{\text{Weight of Ash}}{\text{Weight of Sample}} \times 100$$

Acid insoluble ash

25 ml of 2N HCl was added to the crucible containing the total ash, covered with a watch glass and boiled gently for 5 minutes. The watch glass was rinsed with5 ml of hot water and added in to the crucible. Collected the insoluble matter on an ash-less filter paper and washed with hot water until the filtrate was neutral. Filter paper containing the insoluble matter was transferred to the original crucible, dried on a hot plate and ignited to constant weight. The residue was allowed to cool in a desiccator for 30 minutes; then weighed, calculated the content of acid insoluble ash per gram of air-dried material [10].

$$\% \text{ Acid Insoluble Ash} = \frac{\text{Weight of Acid insoluble ash}}{\text{Weight of Sample}} \times 100$$

Water soluble ash

25 ml of water was added to crucible containing the total ash and boiled for 5 minutes. The insoluble matter was collected in sintered glass crucibles. Washed with hot water and ignited in a crucible for minutes at a temperature not exceeding 450˚C. The weight of those residue in mg was subtracted from the weight of total ash. The content of water-soluble ash was calculated per gram of air-dried material [11].

Weight of water-soluble ash = Weight of total ash – Weight of water insoluble ash

$$\% \text{ Water Soluble Ash} = \frac{\text{Weight of Water soluble ash}}{\text{Weight of Sample}} \times 100$$

Determination of extractive values

This method determines the number of active constituents in each amount of plant material when extracted with solvent. The extractive value is used as a means of evaluating crude drug which are not readily estimated by other means. For example, lowering from the prescribed values indicates the addition of exhausted or unwanted material with original rug or in correct processing of the drug.

$$\% \text{ Extractive Value} = \frac{\text{Weight of extract obtained}}{\text{Weight of Sample}} \times 100$$

Alcohol soluble extractive value

Macerated 5 grams of coarsely powdered air-dried aerial parts of plant with 100 ml ethanol in a stoppered flask for 24 hours, with occasional shaking during the 1st 6 hours and allowed to stand un disturbed for another 18 hours. Filtered rapidly, by taking precautions against loss of alcohols. The 25 ml of the filtrate was evaporated to dryness in a tared flat-bottomed shallow dish, dried at 105˚C and weighed. Calculated W/W ethanol soluble extractive with reference to air dried material [12].

Water soluble extractive value

Macerated 5 grams of coarsely powdered air-dried plant *Cyperus iria* with 100 ml water in a stoppered flask for 24 hours, with occasional shaking during the 1st 6 hours and allowed to stand undisturbed for another 18 hours. Filtered rapidly, then 25 ml of the filtrate was evaporated to dryness in a tared flat-bottomed shallow dish, dried at 105˚C and weighed. Calculated W/W ethanol soluble extractive with reference to air dried material.

Organoleptic evaluation of aerial parts of plant

Organoleptic evaluation can be done by means of organs of sense. This refers to the evaluation of drug by colour, odour, size, shape, taste and special features including touch, texture etc. for this purpose authentic specimen of the material under study and sample of Pharmacopoeial quality should be available to serve as a reference. However, the judgement based on the sensory characteristics like odour, taste, etc. may vary from person to person and time to time based on individual's nature. No preliminary treatment is necessary for evaluating the sample in this manner [11, 12].

Microscopic Evaluation

Histology of leaf

Transverse sections were prepared using pith. Thin transparent floating dissections were kept in watch glass along with water. Then stained with phlouroglucinol: concentrated HCl (1:1) and mounted in glycerine studied.

Histology of stem

Transverse sections were prepared using pith. Thin transparent floating dissections were kept in watch glass along with water. The sections were cleared by warming a few drops of alcoholic potassium hydroxide, wash the sections using water. Then stained with phlouroglucinol: concentrated HCl (1:1) and mounted in glycerine studied [12].

Powder Analysis

For powder analysis the plant was collected and washed thoroughly with water to remove the unwanted matter. This was further dried in the shade. After complete drying, the plant was powdered and passed through sieve no. 60. A small quantity of the powder was treated with phlouroglucinol and conc. HCl (1:1) solution for the detection of various microscopic characters proving the authenticity of the drug. Another sample was mounted in water to see whether it contained calcium oxalate and yet another sample in iodine solution to detect the presence of starch grains [10,11]

Quantitative microscopy

Eye piece was calibrated by using stage micrometer and calibration factor was found out. Small amount of powder was stained with phloroglucinol and concentrated HCl. The treated powder was mounted in dilute glycerin and slide was observed under low power. The length and width of stained fibre was measured by focusing them on the line of eye piece micrometer. The volume of 20 fibres was calculated and multiplied with calibration factor and average length and width of fibres were calculated [12, 13]

Successive Solvent Extraction

Successive solvent extraction of the dried powder of aerial parts of *Cyperus iria* was carried out by using solvents of increasing polarity viz. petroleum ether, chloroform, acetone, methanol and water. Around 18.6 g of dried powder was weighed, moistened with the respective solvent and packed in the soxhlet extractor and was then extracted with 500 ml each of the petroleum ether, chloroform, acetone, methanol and water. After each extraction, the same dried marc was used for the subsequent extraction. Each extract was then filtered, the solvent distilled off and finally the dried extract was obtained. The percentage yield of each extract was calculated. These extracts were subjected to preliminary phytochemical screening [10].

Preliminary Phytochemical Screening of Plant Extracts

Chemical tests for alkaloids

A small portion of dried alcoholic extract was shaken (acidified) with dilute hydrochloric acid and filtered. The acidified filtrate was tested with the following reagents, to detect the presence of alkaloids [11].

a) Mayer's test

The acidified extract (two ml) was treated with 1 ml of Mayer's reagent (potassium mercuric iodide), shaken and noted for the presence of a creamy precipitate.

b) Wagner's test

The acidified extract (two ml) was treated with a few ml of Wagner's reagent (solution of Iodine in potassium iodide) and observed for the presence of reddish-brown precipitate.

Chemical tests for glycosides

A small portion of the extract was hydrolyzed with dilute hydrochloric acid for few hours on a water bath and the hydrolysate was later subjected to following tests to detect the presence of glycosides.

a) Legal's Test

The residue (dry extract) left after evaporation was dissolved in a few milliliters of pyridine. Two milliliters of freshly prepared sodium nitro prusside solution was added to it and then made alkaline with sodium hydroxide solution. It was observed for the formation of pink red Colour.

b) Baljet's test

The few ml of the extract was treated with 1ml sodium picrate solution and a yellow to orange color reveals the presence of cardiac glycosides.

Chemical tests for phenolic compounds and tannins

Ferric chloride test

A small quantity of the extract diluted with water was treated with dilute ferric chloride solution (5%) and observed for the presence of blue color.

Chemical tests for flavanones and flavonoids

Aqueous sodium hydroxide test

Aqueous sodium hydroxide solution was added to the few ml of the extract and the presence of yellow colouration of the solution was noted. The filter paper was wetted with small quantity of alcoholic solution of the extract. That filter paper was exposed to ammonia vapors and noted the yellow color.

Chemical tests for carbohydrates

A small quantity of ethanolic extract was mixed with water or alcohol and filtered. The filtrate was subjected to the following tests to detect the presence of carbohydrates

Molisch's Test

The filtrate (two ml) was treated with a few drops of Molisch's reagent and two ml of concentrated sulphuric acid was added through the sides of the test tube without shaking. Observed for the presence of violet ring at the junction of two solutions.

Fehling's Test

The filtrate (one ml) treated with 1 ml each of Fehling's solution A and B and boiled on a water bath for half an hour, then observed for the presence of red residue at the bottom of test tube.

Chemical tests for proteins and amino acids

Million's Test

The extract (two ml) was treated with few drops of Million's reagent (1gm of mercury+ 9ml of fuming nitric acid) and observed for the presence of white precipitate, which on warming turn into a red colored solution.

Chemical test for terpenoids

Salkowski's Test

The extract (few ml) was dissolved in chloroform. An equal volume of concentrated sulphuric acid was added to it and noted for the appearance of red colour in the chloroform layer and greenish yellow fluorescence in the acid layer.

Chemical tests for sterols

A little amount of the alcoholic extract was refluxed with solution of alcoholic potassium hydroxide until the saponification was observed. The mixture was diluted and extracted with solvent ether. The ethereal extract was evaporated, and the residue subjected to Liebermann-Burchard's and Salkowski's tests.

Liebermann – Burchard's test

The residue was taken with dry chloroform (one ml) and then it was mixed with two ml of specially distilled acetic anhydride followed by a few drops of concentrated sulphuric acid through the sides of the test tube and observed for the development of a deep red colour in the lower portion and green colour in the upper portion which changes to blue and violet.

Salkowski's Test

The residue was dissolved in chloroform and equal volume of concentrated sulphuric acid was added to it and observed for the red colour in the lower layer.

Chemical tests for saponins

Foam or Froth Test

A small quantity of extract was diluted with 20 ml of distilled water in a graduated cylinder. The suspension was shaken for 15 minutes and waited to see if any froth was formed [13].

Thin Layer Chromatographic Analysis

TLC was introduced by Izmailov and Schriber in 1938. Separation of individual components on TLC plate is due to adsorption/partition chromatography or a combination of these two processes. TLC is a very sensitive technique and requires very little amount of substance for analysis. Selection of solvent for TLC is based upon its polarity. The developing period is relatively short, 15 to 60 seconds. The TLC plate can be heated to higher temperature and aggressive reagents can be sprayed on the plate for identification. The movement of chemical compounds relative to solvent front in a chromatographic system is constant [14]. The relative movement is called the retardation factor represented as Rf value, and it is calculated as follows.

$$Rf = \frac{\text{Distance travelled by the solute}}{\text{Distance travelled by the solvent front}}$$

Sample preparation

The plant extracts dissolved in suitable solvent, filtered.

TLC plates

For this study TLC plates were prepared with silica gel and activated in hot air oven for 30 minutes at the temperature of 105˚C.

Spotting of sample

The sample was spotted using a capillarytube on TLC plates 2 cm above from the bottom of the plate.

Selection of a mobile phase

The selection of a mobile phase and detecting agents depends upon nature of active principles in each extract.

Thin layer chromatography study was tried on the chloroform extract of *C.iria* using different mobile phases. The stationary phase used was silica gel G and the detection by iodine chamber method and vanilline sulphuric acid reagent [15].

RESULTS AND DISCUSSION

Pharmacognostic Studies

Determination of Moisture content of the aerial parts of plant

The moisture content of the aerial parts of plant was determined as described earlier. The average loss of moisture content of the aerial parts of plant was determined. The results are presented in table 1

Determination of Ash value of aerial parts of plant

The ash values of the aerial parts of plant were determined according to the procedure given in the earlier. The percentage content of acid insoluble ash was found to be less than that of water-soluble ash. The results are presented in table 2 and 3.

Determination of Extractive values of the aerial parts of plant

The extractive values of the aerial parts of plant (both alcohol soluble and water soluble) were determined as per the method given in the earlier. The water-soluble extractive value of the aerial part of plant was found to be more than that of alcohol soluble extractive value. This may be due to the presence of polar compounds like polyphenols and tannins in the plant. The results are shown in table no 4, 5.

Determination of foreign matter

The foreign matter present in the aerial parts of plant was determined according to the accepted method. The results are shown below in table 6.

Microscopic evaluation

Histology of stem

- Transverse section of the Stem differentiated into epidermis, cortex and ground tissue
- Epidermis: single layered wavy rectangular cells covered by thick cuticle
- Cortex: below the epidermis two to three layers of compactly arranged palisade like cells. Groups of lignified and nonlignified fibres are scattered
- Ground tissue: large thin-walled polygonal parenchyma with intercellular space. Closed collateral vascular bundles are present which are surrounded by bundle sheath shown in figure 2

Histology of leaf

- Transverse section of leaf through midrib and lamina.
- Lamina: dorsiventral leaf,
  - Epidermis: upper and lower epidermis are single layered rectangular of cells covered by cuticle.
  - Mesophyll: differentiate into upper palisade and spongy parenchyma with air cavities
- Midrib: The upper epidermis continues over midrib and associated with bulliform cells
- Collenchyma cells were present below the bulliform cells and above the lower epidermis. Patches of lignified sclerenchyma tissue were present just above the lower collenchyma cells, Vascular bundles are collateral, surrounded by bundle sheath. calcium oxalate crystals are present shown in figure 3.

Powder analysis

The powder when observed under microscope exhibit the following characters

EXTRACTION OF AERIAL PARTS OF PLANT

The coarse powder of the aerial parts of plant *Cyperus iria* was subjected to successive solvent extraction using soxhlet apparatus. After extraction the percentage yield of each extract was calculated with reference to the air-dried drug used for the study. The percentage yield and other characteristic features of the extract are shown in table 9.

Table No 1: Results showing Moisture content of the aerial parts of plant Cyperus iria

Sl. No	Weight of drug + dish before drying (g)	Weight of drug + dish after drying (g)	Loss on drying (g)	Percentage loss on drying (%w/w)	Average (%w/w)
1	30.067	30.382	0.315	15.7	15.7
2	31.162	31.472	0.310	15.5	
3	29.689	29.371	0.318	15.9	

Table 2. Results showing Total Ash Value of the aerial parts of plant Cyperus iria

Sl. No	Weight of empty crucible (g)	Weight of crucible+ Sample (g)	Weight of crucible + Ash (g)	Weight of Ash (g)	Percentage Yield (%w/w)	Average (%w/w)
1	39.330	41.330	39.529	0.199	9.95	
2	38.768	40.768	38.954	0.186	9.30	9.8
3	40.850	42.850	41.053	0.203	10.15	

Table No 3: Results showing Acid insoluble Ash Value of the aerial parts of plant Cyperus iria.

Sl. No	Weight of empty crucible (g)	Weight of crucible+ Acid insoluble Ash (g)	Weight of Acid insoluble Ash (g)	Percentage Yield (%w/w)	Average (% w/w)
1	39.330	39.442	0.112	5.60	
2	38.768	38.865	0.097	4.85	5.2
3	40.850	40.954	0.104	5.20	

Table No 4: Results of Alcohol Soluble Extractive Values of aerial parts of the plant Cyperus iria

Sl. No.	Weight of dry powder (g)	Weight of empty dish (g)	Weight of dish + Extract (g)	Percentage yield (%w/w)	Average (%w/w)
1	1.25	47.884	47.983	7.92	
2	1.25	46.265	46.375	8.80	8.10

3	1.25	52.710	52.805	7.60	

Table No. 5: Results showing Water Soluble Extractive Values of aerial parts of the plant Cyperus iria

Sl. No.	Weight of dry powder (g)	Weight of empty dish (g)	Weight of dish + Extract (g)	Percentage yield (%w/w)	Average yield (%w/w)
1	1.25	47.884	48.024	11.2	
2	1.25	46.265	46.447	14.4	12
3	1.25	52.710	52.840	10.4	

Table No 6: Results showing the foreign matter present in the aerial parts of plant Cyperus iria

Sl. no:	Weight of crude drug (g)	Weight. of foreign matter present(g)	Percentage yield(%w/w)	Average (%w/w)
1	25	0.080	0.32	
2	25	0.120	0.48	0.393
3	25	0.095	0.38	

Table No. 7: Quantitative microscopy

Sl. No	Length of Fibre (µm)	Width of Fibre (µm)	Calibration Factor (CF)	Actual length of Fibre (µm)	Actual width of Fibre (µm)
1	45	5	5.28	237.6	26.4
2	52	2		274.56	10.56
3	63	5		332.64	26.4
4	40	4		211.2	21.12
5	46	5		242.88	26.4
6	43	2		227.04	10.56
7	79	3		417.12	15.84
8	83	2		438.24	10.56
9	39	3		205.92	15.84
10	80	4		422.4	21.12
11	47	4		248.16	21.12
12	72	2		380.16	10.56
13	89	4		469.92	21.12
14	40	4		211.2	21.12
15	81	5		427.68	26.4
16	57	4		300.96	21.12
17	78	3		411.84	15.84
18	91	3		480.48	15.84
19	65	3		343.2	15.84
20	62	2		327.36	10.56

Table No 8: Result showing length and width of fibres of C. iria

Sl. No	Parameters of Fibres (µm)	Average (µm)	Maximum (µm)	Minimum (µm)
1	Length	309.408	480.48	205.92
2	Width	17.16	26.4	10.56

Table No 9: Results showing the Nature and Percentage yield of the aerial parts of plant extracts of Cyperus iria

Sl. no	Solvent used for extraction	Colour	Consistency	Percentage yield (%w/w)
1	Petroleum ether	Light green	Semisolid	2.376
2	Chloroform	Dark green	Semisolid	2.774
3	Acetone	Brown	Semisolid	4.198
4	Methanol	Dark brown	Semisolid	4.876
5	Aqueous	Black	Powder	6.968

Table No 10: Results of qualitative phytochemical screening of petroleum ether, chloroform, acetone, methanol and aqueous extract of Cyperus iria

Sl.no	Qualitative Test	Pet.Ether	Chloroform	Acetone	Methanol	Aqueous
1	**Alkaloids**					
a.	Mayer's test	-	++	+	+	+
b.	Wagner's test	-	++	+	+	+

c.	Hager's test	-	++	+	+	+
d.	Dragendorff's test	-	++	+	+	+
2.	**Glycosides**					
a.	Legal's test	-	-	-	-	-
b.	Baljet's test	-	++	-	+	+
c.	Libermann Buchard test	+	++	+	-	+
d.	Borntrager's test	-	-	-	-	-
e.	Modified Borntragers' test	-	-	-	-	-
3.	**Phenolics**					
a.	Ferric chloride test	-	-	+	+	+
b.	lead acetate test	-	-	+	++	+
c.	Decolourisation	-	-	++	++	++
4.	**Flavones& Flavonoids**					
a.	Aqueous NaOH test	+	++	-	-	+
b.	Ammonia test	-	-	-	+	-
5.	**Carbohydrates**					
a	Molisch's test	-	-	+	+	+
b.	Benedict's test	-	-	+	+	+
c.	Fehling's test	-	-	+	++	+
6.	**Proteins & Amino acid**					
a.	Millon's test	-	-	-	-	-
b.	Biuret test	-	-	-	-	-
c.	Ninhydrin test	-	-	-	-	-
7.	**Terpenoids**					
a.	Salkowski's test	+	-	+	+	+
8.	**Sterols**					
a.	Libermann buchard test	+	++	+	-	+
b.	Salkowski's test	+	+	+	+	+
9.	**Saponins**					
a.	Foams test/ Froth test	-	-	+	++	++

Table no. 5.13: Results of the TLC analysis of chloroform extract of Cyperus iria

Sl. no	Solvent system	No. of spots	Detection	Colour of spot	Rf value
1.	Chloroform: Methanol: Acetone	3	Iodine chamber	Yellow	0.25 (Spot 1) 0.59 (Spot 2) 0.64 (Spot 3)

Figure- 1- Cyperus iria

Fig no. 2 TS of stem of Cyperus iria

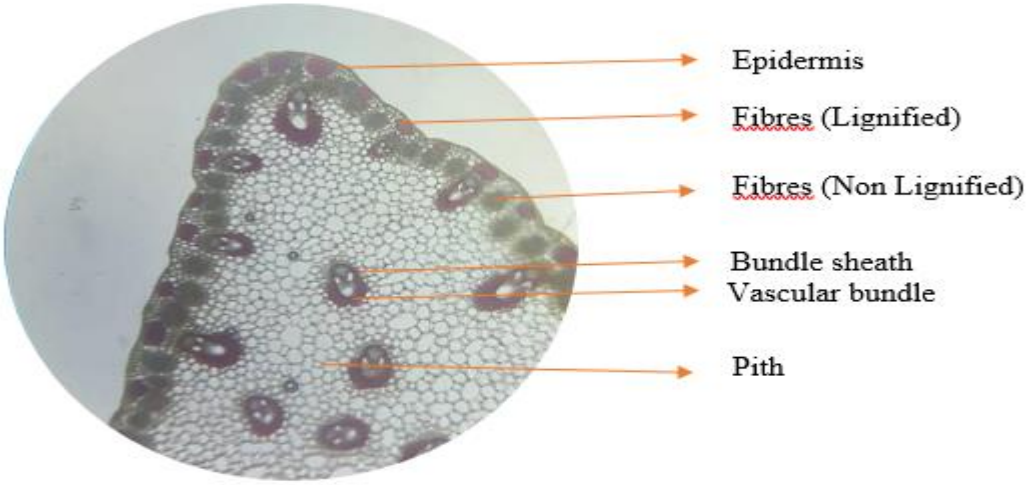


Fig no 3: TS of leaf of Cyperus iria

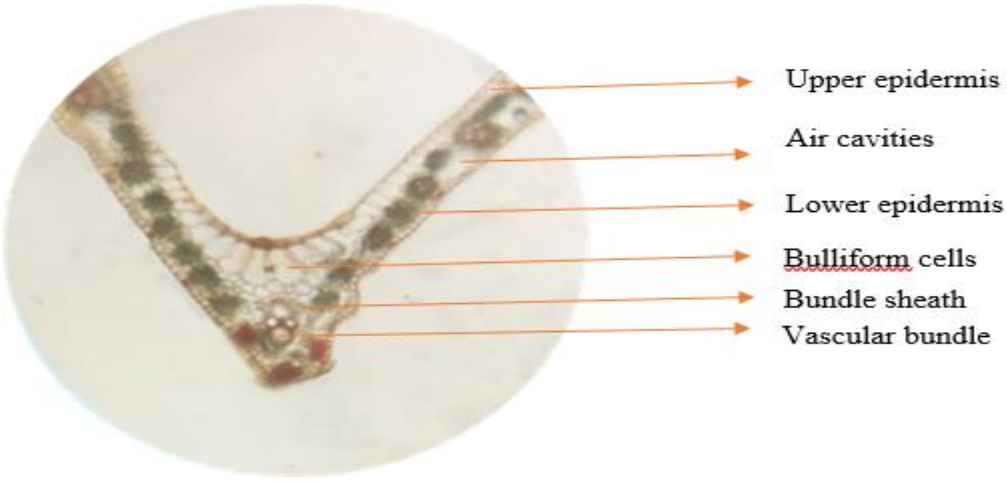
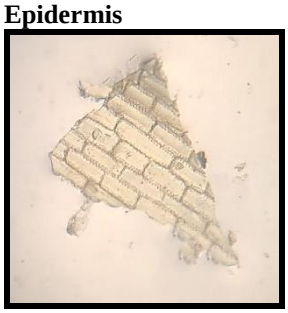


Fig no. 4: Characteristic image of powder analysis

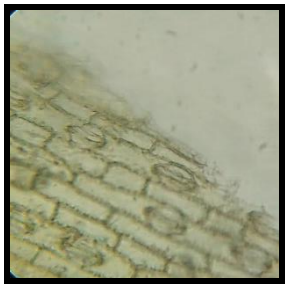
Epidermis



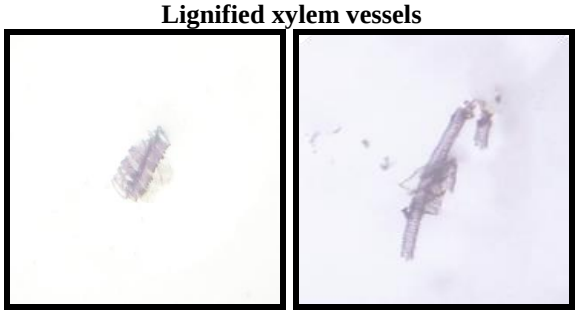
Phloem fibres

Lignified xylem fibres

Epidermal cells with stomata



Lignified xylem vessels



Calcium oxalate crystals

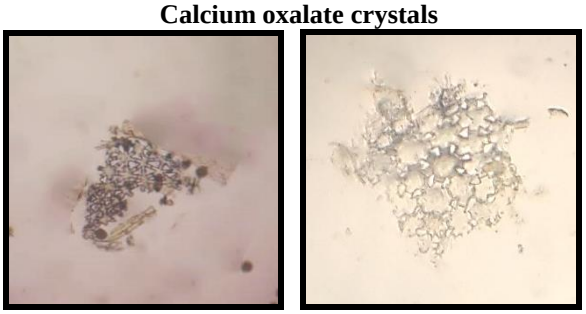
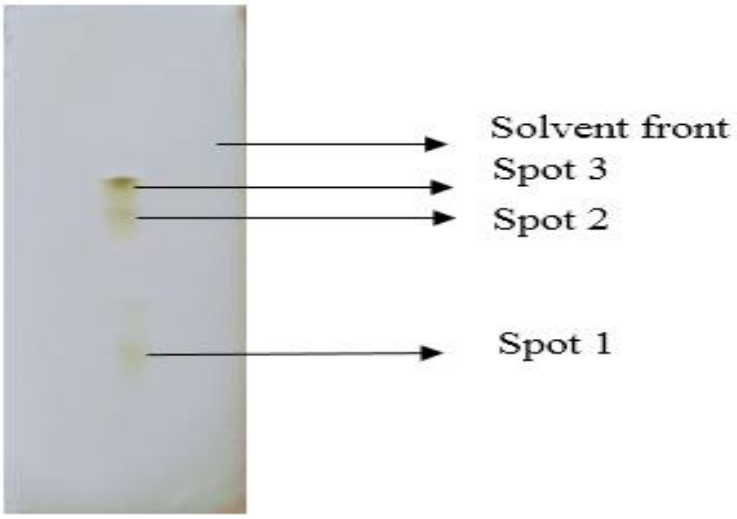


Fig no. 5: TLC of chloroform extract of Cyperus iria



Preliminary Phytochemical Screening of Extracts

The extracts were subjected for qualitative chemical analysis for the identification of various phytoconstituents, like alkaloids, glycosides, phenolics, flavanoids, carbohydrates, proteins and amino acids, terpenoids, sterols and saponins etc. Petroleum ether extract contains terpenoids, steroids and glycosides. Chloroform extract contain alkaloids, glycosides, flavonoids, terpenoids and steroids. Alkaloids, glycosides, phenolics, terpenoids, steroids and saponins are present in acetone extract. While in methanol extract and water extract, it shows the presence of alkaloids, glycosides, phenolics, flavonoids, carbohydrates and saponins. The results of the chemical tests for each extract are recorded and tabulated in the following table 10.

TLC of chloroform extract of *cyperus iria*

TLC of chloroform extract using solvents Chloroform: Methanol: Acetone (8:1:1), the solvent system suitable as a screening system for the TLC investigation of alkaloids and flavonoid glycosides. The solvent system showed good separation and three yellowish green clear spot. The results of the TLC analysis of chloroform extract are tabulated below in table 11 and Fig 5.

CONCLUSION

The aerial parts of plant *Cyperus iria* was collected, authentified, dried and powdered. Pharmacognostical studies were carried out like moisture content, foreign matter, ash value (total ash, acid insoluble ash and water-soluble ash), extractive value (water soluble extractive value and alcohol soluble extractive value). The organoleptic evaluation was done to identify the colour, odour, taste, size, shape, texture etc. Microscopical studies were done to identify the microscopical characters of the plant and powder microscopy was performed for the identification of powder characteristics. The powdered drug material was then subjected to successive solvent extraction using solvents of increasing polarity viz. petroleum ether, chloroform, acetone, methanol and water. After each extraction, the same dried marc was used for the subsequent extraction. Each extract was filtered, the solvent distilled off and finally the dried extract was obtained. These extracts were used for further phytochemical screening and pharmacological studies. The phytochemical studies reveal the presence of alkaloids, carbohydrates, glycosides, flavonoids, phenolics, steroids, terpenoids and saponins. From the TLC analysis of chloroform extract by using solvent system like Chloroform: Methanol: Acetone (8: 1: 1) showing a good separation and three clear spots with Rf value 0.25, 0.59 and 0.64 respectively.

REFERENCES

1. Reelsaltlife.blogspot.in:2012;02:role-of-herbs-in-human-life.html
2. Kokatae CK. Practical Pharmacognosy;4th ed. Delhi: Vallabh prakashan; 2008.21
3. Mojab F, Kamalinejad M, Ghaderi N, Vahidipour HR. Phytochemical Screening of Some Species of Iranian Plants. *Iranian Journal of Pharmaceutical Research.* 2, 2003, 77-82.
4. Molyneux RJ, Stephen T Lee, Dale R Gardner, Kip E Panter, Lynn FJ. Phytochemicals: The good, the bad and the ugly. *National Library of Medicine*. 68, 2007, 2973-2985.
5. Sasidharan S, Chen Y, Saravanam D, Sundram KM, Yoga L. Extraction, Isolation and Characterisation of Bioactive Compounds from Plant Extracts. *Afr J Tradit Complement Altern Med*. 8(1), 2011, 1-10
6. Cyperus iria- Invasive species compendium:CABI:Cabi.org/isc/datasheet
7. The Ayurvedic Pharmacopoeia of India. Part 1, vol. 2, 1st edition, 1999, 190-191

8. Pulok K M. Quality control of herbal drugs, an approach to evaluation of botanicals. *Business horizon*. 2002, 529-534 and 554-559
9. Sass J E. Elements of Botanical Microtechnique. New York: Mc Grow Hill Book, 1940.
10. Kandelwal KR. Practical pharmacognosy techniques and experiments. 18th ed, Pune, *Nirali Prakashan*, 2007, 10-29.
11. Kokatae CK, Purohit AP, Gokale SB. Pharmacognosy, 24th ed. Pune: *Nirali Prakshan*, 2003, 97-113
12. WHO, Geneva. Quality control methods for medicinal plant material. 1st edition. Delhi, AITBS Publishers and distributers, 2002.
13. Sethi PD and Dilip C. Identification of drugs in pharmaceutical formulations by thin layer chromatography;2nd edition/CBS publishers and distributors, 1999, 1-19
14. Antara S and Amla B. Evaluation of antimicrobial activity of different solvent extracts of medicinal plant *Melia azedarach* L. *Int J Current Pharma Res*. 4(2), 2012, 67-73
15. Egon S. Thin layer chromatography:A laboratory hand book;2nd edition/Springer- Verlag Berlin,1969, 1-5.