

STANDARDIZATION AND PRELIMINARY PHYTOCHEMICAL INVESTIGATION ON *CYPERUS ROTUNDUS* LINN RHIZOME

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ABSTRACT

Cyperus rotundus Linn vernacularly called “Nagarmotha” is a medicinal plant belonging to the family of the *Cyperaceae* and was widely distributed in the Mediterranean basin areas. This plant, which grows naturally in tropical, subtropical and temperate regions, is widespread in northeast appearing among Indian, Chinese, Japanese natural drugs used as home remedy against spasms, stomach disorders and irritation of bowel. *Cyperus rotundus* has many different uses and used as cooling, intellect promoting, nervine tonic, diuretic, antiperiodic, diarrhoea, dysentery, leprosy, bronchitis, amenorrhoea, blood disorders, analgesic, anti-inflammatory and antipyretic activity. The present study was aimed to evaluate the parameter to determine the quality of the *Cyperus rotundus* rhizome. These studies comprise to investigate macroscopy, powder analysis, physicochemical parameters, preliminary phytochemical screening and fluorescence characteristics. The studies will provide referential information for the correct identification of the crude drug.

KEYWORDS: *Cyperus rotundus*, Fluorescence characteristics, Physico-chemical parameters, Phytochemical screening.

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INTRODUCTION

Cyperus rotundus Linn has many different uses were based on the different parts of the plant. The medical use of *Cyperus* has been used in medicine for thousands of years. The parts of the *Cyperus* used are its leaves, seeds, rhizome and oil¹. *Cyperus rotundus* Linn is widely distributed in the Mediterranean basin areas. This plant, which grows naturally in tropical, subtropical and temperate regions, is widespread in northeast². *Cyperus rotundus*, vernacularly called “Nagarmotha” is a medicinal plant belonging to the family of the *Cyperaceae* and appearing among Indian, Chinese, Japanese natural drugs used as home remedy against spasms, stomach disorders and irritation of bowel³. In Indian system of medicine, the rhizome of the plant has been recommended for use in several clinical conditions like fever and arthritis. The rhizomes are cooling, intellect promoting, nervine tonic, diuretic, antiperiodic, and used to treat diarrhoea, dysentery, leprosy, bronchitis, amenorrhoea and blood disorders⁴. The rhizome is reported to possess analgesic, anti-inflammatory, antipyretic activity⁵. In India, the fruits were considered carminative, diuretic, tonic, stomachic, antibilious, and refrigerant. Both the leaves and seeds are rich in volatile oils that act mainly on the digestive system, stimulating the appetite and relieving irritation and as an expectorant. The oil is fungicidal and bactericidal. The leaves were widely used to flavor food, especially in the Middle East, and Southeast Asia. The seeds are also an ingredient of curries and pickling spices, dishes a la grecque, and bakery products. Medicinally, *Cyperus* was used internally for minor digestive problems, and externally for hemorrhoids and painful joints (seeds). Seeds reduce griping in laxative preparations based on *Rheum officinal* and *Cassia angustifolia*. The oil adds to the flavor of gin, vermouth and Chartreuse, and was also prized in perfumery⁶. They form an ingredient of poly herbal formulation Abana and health food Amrita Bindu, useful for prevention of nitrosamine induced depletion of antioxidant defense. The phytochemical investigations of *C. rotundus* have revealed the presence of polyphenol, flavonol glycoside, saponin, vitamin – C, sesquiterpenoids, essential oil and cardiac glycosides^{1,3}.

MATERIAL AND METHODS

Plant material

The fresh plant materials i.e. rhizomes of *Cyperus rotundus* were collected from herbal garden of north region of Haryana in the month of September. The plant was authenticated by taxonomist, Dr. Saroj Arora, Head of the Department of Botany, Guru Nanak Dev University (GNDU), Amritsar, India. The voucher specimen (6508/A.N) for *Cyperus rotundus* rhizomes was deposited at Department of Pharmacognosy & Phytochemistry, SBS College of Pharmacy, Patti (Tarn Taran), Punjab. Fresh rhizomes were used to study the macroscopy and microscopy whereas shade dried powder was used for the determination of physicochemical parameters and phytochemical screening.

Macroscopy

The fresh rhizome of *Cyperus rotundus* was subjected to macroscopic studies which comprised of organoleptic characters of the drugs viz., colour, odour, appearance, taste, smell, texture, fracture, etc⁷.

Powder Analysis

The rhizomes are oven dried at 60°C for 4-6 hrs to make moisture free and powdered the rhizomes and was passed through sieve no. 60. Powder characteristic were studied by standard methods. The powder of the drug was boiled with chloral hydrate to remove the colouring matters, mounted on the glass slides using glycerin, covered with a cover slip and viewed under microscope. The powder was also stained with phloroglucinol and hydrochloric acid (1:1) and examined under microscope.

Physicochemical parameters

The determination of various physicochemical parameters such as total ash, acid insoluble ash, water soluble ash, water soluble extractive value and alcohol soluble extractive were calculated as per Indian pharmacopoeia. Successive soxhlet extractives of the drug were carried out with various solvents like petroleum ether, benzene, chloroform, ethyl acetate and methanol and weight, colour/consistency of the extractives were observed^{8,9}.

Preliminary phytochemical screening

The different extractives were subjected to preliminary phytochemical investigation for the presence of various phytoconstituents like alkaloids, carbohydrates, glycosides, phenolic and tannins, flavonoids, protein and amino acids, saponins, acidic compounds, phenol, mucilage, resins, and lipids/ fats using method describe in^{10,11}.

Fluorescence analysis

When physical and chemical parameters are inadequate as it often happens with the powdered drugs, the plant material may be identified from their adulterants on basis of fluorescence study. Fluorescence analysis of the rhizome powder sample was carried out by treating with different chemical reagents to observe various colour instances¹².

RESULTS AND DISCUSSION

The microscopic character was useful in quick identification of plant material and also serves as an important standardization parameter. Organoleptic evaluations of *Cyperus rotundus* rhizome were reported in **(Table-1)**. Rhizomes are simple, elongated, broadly obovoid, trigonous shape, and surface is slightly tuberous at the base shown in **(Figure-1)**.

The various diagnostic characteristic of powder are coarse, yellowish brown with pleasant odour and astringent and bitter taste. Microscopic examination of powder shows presence of pitted vessels and fibre shown in **(Figure-2)**.

Various physico-chemical parameters are important in determination of adulterants and improper handling of drugs. **(Table-2)** shows the result of various physico-chemical parameters of powdered drugs carried out using standard method. Ash value used to determine quality and purity of crude drug. The extractive values are useful to evaluate the chemical constituents present in crude drug and also help in estimation of specific constituents soluble in particular solvent.

The results of extractive value of powdered drug in different solvent obtained by successive extraction is shown in **(Table-3)**. Higher extractive value of methanol extract is due to presence of alkaloids, carbohydrates, glycosides, phenolic and tannins, flavonoids, protein and amino acids, acidic compounds, phenol and lipids/ fats. **(Table-4)** shows behavior analysis of different solvent extract of *Cyperus rotundus* rhizome under visible light, short and long UV. All the extract obtained by successive extraction in different solvent subjected to qualitative chemical test and the result shown in **(Table-5)**. The result shows that maximum constituents found in methanolic extract of *Cyperus rotundus* rhizome. Such preliminary phytochemical screening was helpful in prediction of nature of drugs and also useful for the detection of different constituents present in different polarity solvent.

The fluorescence analysis of powdered drug in day light, short UV and long UV were examined by reported methods. The observations are given in **(Table-6)**.

CONCLUSION

In present investigation various standization parameters such as macroscopy, microscopy, physico-chemical parameter and phytochemical screening was carried which could helpful in authentication of *Cyperus rotundus* rhizome. The information obtained from preliminary phytochemical screening will be useful in finding out the genuity of the drug. Ash values, extractive values can be used as reliable aid for detecting adulteration. The other parameters observed may be useful for the future identification of the plant.

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Table 1: Organoleptic Characters of Rhizomes of *Cyperus rotundus*

Sr. No.	Organoleptic Character	<i>Cyperus rotundus</i> (rhizomes)
1	Type	Simple
2	Colour	Dark brown or black externally, creamish-yellow internally
3	Odour	Pleasant
4	Taste	Slightly pungent, bitter and astringent
5	Size	10-20 cm long 0.8-2.5 cm wide
6	Shape	Elongated, broadly obovoid, trigonous
7	Surface	Slightly tuberous at the base

Table 2: Physico-chemical Parameter of Rhizomes Powder of *Cyperus rotundus*

Physico-chemical parameter	Value (% w/w)
Ash values	
Total ash	8.3 % w/w
Acid insoluble ash	3.4 % w/w
Water soluble ash	4.4 % w/w
Extractive values	
Water soluble extractive	9.5 % w/w
Alcohol soluble extractive	6.5 % w/w

Table 3: Yield of Extract by Successive Solvent Extraction

Sr. No.	Extract	<i>Cyperus rotundus</i> (%w/w)
1	Petroleum ether	1.2%
2	Benzene	2.52%
3	Chloroform	2.92%
4	Ethyl acetate	1.75%
5	Methanol	6.88%

Table 4: Behavior Analysis of Different Solvent Extract of *Cyperus rotundus* Rhizome Under visible light, Short and Long UV

Sr. No.	Extract	Visible light	UV (254 nm)	UV (366 nm)
1.	Petroleum Ether	Yellowish brown	Brown	Black
2.	Benzene	Brownish yellow	Brown	Black
3.	Chloroform	Brown	Greenish brown	Black
4.	Ethyl acetate	Brownish black	Yellowish black	Black
5.	Methanol	Reddish brown	Dark brown	Dark brown

Table 5: Preliminary Phytochemical Screening of Rhizomes Extract of *Cyperus rotundus*

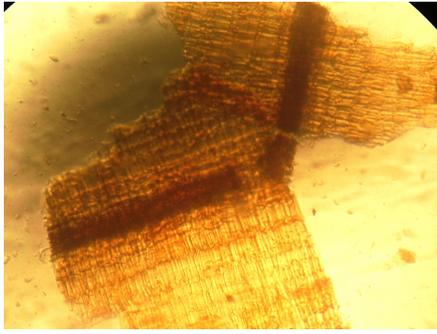
Sr. No.	Plant Constituent	Petroleum Ether Extract	Benzene extract	Chloro-Form Extract	Ethyl acetate extract	Methanol Extract
1.	Alkaloids	-	-	-	+	+
2.	Glycosides	+	+	-	-	+
3.	Proteins & Amino acids	-	+	+	+	+
4.	Carbohydrates	+	-	-	+	+
5.	Tannins	+	-	+	+	+
6.	Flavonoids	+	-	-	-	+
7.	Acids	+	+	+	+	+
8.	Saponins	-	-	-	-	-
9.	Phenols	+	+	-	-	+
10.	Lipids	++	-	-	-	+

+ Present, - Negative

Table 6: Fluorescence Analysis of Powder of *Cyperus rotundus* Rhizomes with Various Chemical Reagents

Sr. No.	Treatment of Drug Powder	Visible light	UV (254 nm)	UV (366 nm)
1.	Drug Powder	Brown	Dark green	Black
2.	H ₂ SO ₄	Brownish black	Greenish black	Black
3.	H ₂ SO ₄ + H ₂ O	Dark brown	Greenish black	Black
4.	Conc.HCl	Yellowish brown	Greenish black	Black
5.	Conc.HCl+H ₂ O	Brown yellowish	Dark yellowish	Yellowish black
6.	Conc.HNO ₃	Reddish brown	Greenish black	Greenish black
7.	Acetic acid	Brownish black	Greenish black	Black
8.	Conc.HNO ₃ + H ₂ O	Yellowish brown	Greenish brown	Black
9.	Methanol	Creamish brown	Brown	Black
10.	Ethanol	Light brown	Brown	Black
11.	Chloroform	Brownish black	Greenish brown	Black
12.	Pet. Ether	Dark brown	Greenish brown	Black
13.	D-water	Brown	Dark brown	Brownish brown
14.	10% NaOH	Brownish black	Greenish brown	Black
15.	5% Iodine	Brown	Dark brown	Greenish black
16.	Picric Acid	Greenish brown	Brownish green	Black
17.	FeCl ₃ sol.	Yellowish brown	Yellowish black	Black
18.	Ammonia sol.	Brown	Greenish brown	Black
19.	Ethyl acetate	Light brown	Greenish brown	Black

**Figure 1: *Cyperus rotundus* Rhizomes**



Fragments of Pitted Vessels



Fragments of Fibres

Figure 2: Powder Microscopy of *Cyperus rotundus* Rhizome

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