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Original Research Article

Pharmacognostic analysis of Tudri Surkh (*Cheiranthus cheiri*)

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ABSTRACT

Background: Tudri Surkh *(Cheiranthus cheiri)* is an important medicinal plant species used in Unani system of medicine. Despite having immense medicinal importance, little information is available on the standardization parameters of the species. For this reason, present work was carried out to establish pharmacognostical standards and generate comprehensive report on the quality control and standardization parameters of Tudri Surkh *(Cheiranthus cheiri)*.
Methods: Seeds of the plant were examined using microscopy and macroscopy, physicochemical parameters, extractive values, and fluorescence analysis.
Results: The macroscopic, microscopy, and physicochemical parameters of seeds of Tudri Surkh *(Cheiranthus cheiri)* revealed various diagnostic characteristics in the species.
Conclusions: This study provides complete pharmacognostic profile of Tudri Surkh *(Cheiranthus cheiri)* and hence will be useful for correct identification and authentication of the species for future studies.

Keywords: Tudri Surkh, *Cheiranthus cheiri*, Physicochemical, Fluorescence, Extractive values

INTRODUCTION

World Health Organization (WHO) has recognized the imperative role played by traditional systems of medicine in providing essential health service to a major portion of world's population particularly in rural areas. Majority of Indian population rely on indigenous system of medicine including Ayurveda, Siddha and Unani or Greco-Arabic medicine. In Unani system of medicine drugs based on natural products like plants, minerals and animal parts are the mainstay of treatment.[1] Medicinal plants or their products are best source of large number of pharmacologically useful compounds or constituents which alleviate various disease including acquired immuno-deficiency syndrome (AIDS), cancer, and many degenerative diseases. WHO (traditional medicine division) recognizes that the use of certain plants since centuries as therapeutical agents should be taken into account as proof of their efficiency.[2] Phytochemicals including lipid, protein, starch, sugars, phenols are synthesized by plants for their normal growth and development. These plants are used as therapeutic agents for combating disease and health related problems due to the presence of various bioactive chemical constituents.[3] Extraction and phytochemical analysis helps to separate the medicinally active portions of plants by using universal solvents including water and alcohol through standard procedure.[4]

A number of medicinal plants are consumed by humans as therapeutic agents and their evaluation by way of proper identification and quality assurance is of utmost importance. It is imperative to establish pharmacognostic parameters to ensure use of quality plant material for manufacturing traditional herbal drugs and basic steps for evaluation of the material include macroscopic and microscopic characterization, physicochemical properties, and fluorescence analysis.[5]

According to WHO determination of macroscopic and microscopic characteristics of medicinal plants is the first step towards establishing their identity and the degree of purity and is recommended to be carried out before any further investigation is undertaken.[6]

Tudri Surkh (*Cheiranthus cheiri*) is a perennial plant species belonging to family Brassicaceae (Cruciferae). According to classical Unani literature the seeds and oil of Tudri Surkh (*Cheiranthus cheiri*) are used in the management of various disease that include cancer. Tudri Surkh (*Cheiranthus cheiri*) possess remarkable activity against various physiological disorders. It was formerly used as diuretic. Smaller doses of seeds of Tudri Surkh (*Cheiranthus cheiri*) are cardiotonic supporting a failing heart in a manner similar to foxglove but in more than small doses it is toxic.[7] Seeds are also considered as tonic, diuretic, expectorant and stomachic. They are good remedy for fevers and eye injuries and are used in dry bronchitis.[8]

Discorides has described its potential role in cancer, jaundice, sciatica, and drug toxicity.[9-11] Even though seeds of Tudri Surkh (*Cheiranthus cheiri*) possess good medicinal properties, information available on its standardization parameters is not enough. Therefore, current work is an effort for generating comprehensive report on the quality control and standardization parameters of seeds of Tudri Surkh (*Cheiranthus cheiri*). Accordingly, methods of microscopy and macroscopy, physicochemical parameters, fluorescence analysis and extractive values were utilized to determine pharmacognostical standards of seeds of Tudri Surkh (*Cheiranthus cheiri*). The physicochemical analysis of seeds of Tudri Surkh (*Cheiranthus cheiri*) will establish its pharmacognostical standards to ensure quality control and will aid to assemble appropriate monograph for proper identification.

METHODS

Plant material and sample preparation

The seeds of Tudri surkh (*Cheiranthus cheiri*) were purchased from Khari Baoli, New Delhi and taxonomically identified by taxonomist at National Institute of Science Communication and Information Resources (NISCAIR), New Delhi.

The study was carried out in the department of Pharmacology (Ilmul advia), School of Unani Medical Education and Research (SUMER), Hamdard University, Jamia Hamdard, New Delhi from January 2017 to June 2017.

Chemicals and reagents

All chemicals used were of analytical grade. Acetic acid, chloroform, diethyl ether, ethyl alcohol or ethanol, ferric chloride, gallic acid, hematoxylin-eosin, methanol, Molisch's reagent, petroleum ether, sodium phosphate buffer, sodium acetate, toluene from S D fine chemical Pvt. Ltd (Mumbai India), butylated hydroxy toluene, Dragendroff's reagent, EDTA, orthophosphoric acid, Mayer's reagent from E Merck Pvt. Ltd. (Mumbai India) were used.

Microscopic examination

Seed of Tudri surkh (*Cheiranthus cheiri*) was embedded in a paraffin block. A small hole was made at the centre with a thin glass rod in which seed was placed. Sections were made with the help of microtome and were observed for cell structure and contents under microscope with a magnification power of 4x, 10x, and 40x.[6]

Loss on drying

5 gm of powdered drug was placed on a tarred evaporating dish. Tarred evaporating dish was dried at 105 ^{o}C for 6 hours and weighed. The drying continued until two successive readings matched each other or the difference between two successive weighing was not more than 0.25% of the constant weight.[6]

Determination of swelling index

To determine swelling index 1 gm of drug material was placed in 25 ml glass stoppered measuring cylinder. After adding 25 ml of water, mixture was shaked thoroughly for 10 minutes and allowed to stand for 3 hours. Volume in ml occupied by the drug was measured including sticky mucilage. Mean value of individual determination was calculated in relation with 1gm of drug.[6]

Determination of ash values

Ash which is recovered after the ignition of herbal material is determined by three methods which measure total ash, acid–insoluble ash and water-soluble ash.[6]

Total ash

Air dried crude drug (5 gm) was placed in a tarred silica crucible and was incinerated at a temperature of 500-600 ^{o}C until it turned white, indicating absence of carbon. Then crucible was cooled on desiccator and weighed. The percentage of ash with reference to the air-dried drug was calculated.[6]

Acid insoluble ash

The total ash was boiled with 25 ml of hydrochloric acid (HCl) for 5 minutes. The insoluble matter was collected on an ash less filter paper and washed with hot water until filtrate was neutral. Then filter paper was placed in crucible and ignited. The residue was cooled and weighed. The percentage of acid insoluble ash with reference to the air-dried drug was calculated.[6]

Water soluble ash

About 25 ml of water was added to the crucible containing total ash and boiled for 5 min. The insoluble matter was collected on filter paper and was ignited for 15 min at a temperature not exceeding 450 ºC. The weight of residue was subtracted from the weight of ash. The content of water-soluble ash was calculated with reference to the air-dried drug.[6]

Determination of pH

pH 1% solution

1 gm drug was taken and dissolved in 100 ml of distilled water and then filtered. pH of filtrate was determined with glass electrode.[6]

pH 10% solution

10 gm drug was taken and dissolved in 100 ml of distilled water and filtered. pH of filtrate was determined with standard glass electrode.[6]

Fluorescence analysis

The powdered drug material was subjected to different chemicals like acetic acid, picric acid, sulphuric acid, ferric chloride, 1N HCl, nitric acid, sodium hydroxide and water. Then they were observed under visible light and UV light at 245 nm and 366 nm.[12]

Determination of extractive values

Cold extraction

10 gm of coarsely powdered air-dried material accurately weighed in a glass stoppered conical flask was macerated with 100 ml of solvent, frequently shaken and allowed to stand for 18 hours. The solution was filtered rapidly and transferred to a tarred flat-bottomed dish and evaporated to dryness on water bath. Extract was dried at 105 ºC for 6 hours and cooled on a desiccator for 30 min. Then it was weighed and content of extractable matter in mg per g of air-dried material was calculated. Cold maceration was done by different solvents which include petroleum ether, water, methanol and chloroform.

Hot extraction

10 gm. of coarsely powdered air-dried material was extracted with different solvents including petroleum, water, methanol and chloroform by soxhletion method or hot continuous extraction. Drug material was kept in a porous bag or thimble made of cotton placed in thimble chamber of soxhlet apparatus. Extraction solvent was heated in the round bottom flask, vaporizes into the sample present in thimble condenses in the condenser and dripped back. After the liquid content reached the siphon arm, the liquid content gets emptied into flask again and the process was continued.[13] Hot extraction was done with different solvents which include petroleum ether, water, methanol and chloroform.

Successive extraction

The dried and coarsely powdered drug material (50 gm) was subjected to successive extraction in a soxhlet apparatus with different solvents like petroleum ether, chloroform, methanol and water. The extracts were evaporated to dryness and their constant extractive values were recorded.[6]

Statistical analysis

Data was entered in Microsoft excel 2016 and analysed by GraphPad prism statistical software. Means were calculated for each of the variables, with three values obtained for each variable.

RESULTS

Macroscopic characteristics

Seeds of Tudri Surkh (*Cheiranthus cheiri*) are globular in shape, about 2.0-3.0 mm long and 1.5-2.0 mm wide with mucilaginous warty surface. Testa is dark reddish brown and minutely pitted. The odour is musky and taste is mucilaginous (Figure 1a and b).

Figure 1: (a) and (b) Seeds of Tudri Surkh (*Cheiranthus cheiri*).

Microscopic

The microscopic examination of seeds of Tudri Surkh (*Cheiranthus cheiri*) revealed presence of various diagnostic features as shown in Figure 2. The cells of the epidermis of the testa contain mucilage. The embryo is oily and greenish yellow. It consists of two cotyledons folded along their mid ribs to enclose the radicle. The starch is present in the seeds.

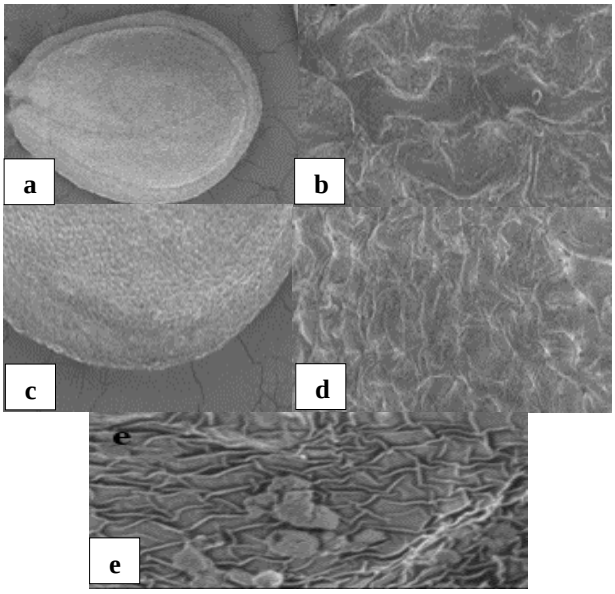

Figure 2: (a)-(e) Seed morphology of Tudri Surkh (*Cheiranthus cheiri*).

General physicochemical screening

The results of various physicochemical parameters of Tudri Surkh (*Cheiranthus cheiri*) seeds are presented in Table 1. Results obtained on cold extraction, hot extraction, and successive extraction values are given in Figure 3. The fluorescence characteristics of powdered seeds were observed in day light and UV light at 254 nm and 366 nm and the observations are presented in Tables 3 as colour variations.

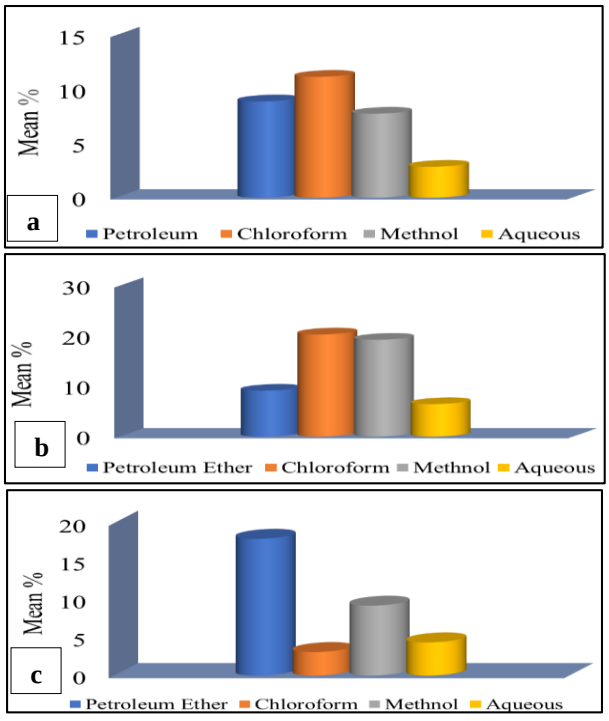

Figure 3: (a) Cold extractive value, (b) hot extractive values, and (c) successive extractive values.

Table 1: General physicochemical screening of seeds of Tudri Surkh (*Cheiranthus cheiri*).

S. no.	Physicochemical constants	Mean value
1	% age of loss on drying	7.6
2	% age of ash content	6
3	% age of acid insoluble ash	1.33
4	% age of water soluble ash	1.73
5	Swelling constant of crude drug	1.1
6	pH of the drug at 1%	7.01
7	pH of the drug at 10 %	7.04

Table 3: Fluorescence analysis of crude drug powder with different chemical reagents.

S. no.	Treatment	Day light	UV light (254 nm)	UV light (366 nm)
1	Powder as such	Light brown	Blackish brown	Whitish yellow
2	Powder with distilled water	Dark brown	Blackish brown	Dark black
3	Powder with 1N NaOH in water	Black	Dark black	Dark brown
4	Powder with HNO_3	Light brown	Greenish brown	Dark black
5	Powder with H_2SO_4	Black	Dark black	Brown
6	Powder with iodine	Dark brown	Blackish brown	Dark black
7	Powder with conc. HCl	Dark brown	Greenish brown	Dark brown
8	Powder with ammonia	Brown	Dark greenish brown	Greenish brown
9	Powder with ferric chloride	Black	Dark black	Blackish brown
10	Powder with Ferric chloride	Black	Dark black	Blackish brown
11	Powder with glacial acetic acid	Brown	Whitish black	Light yellow
12	Powder with picric acid	Yellow	Greenish brown	Blackish brown

DISCUSSION

Tudri surkh (*Cheiranthus cheiri*) belongs to the family *Brassicaceae* (*Cruciferae)* and is originally a native of European countries. Tudri surkh is used either alone or as an ingredient in various pharmacopeial formulations in Unani system of medicine. Formulations containing Tudri surkh is used in dry bronchitis, orchitis, mastitis, cancer and jaundice.[9] Precise identification and determination of

physicochemical standards of plant material are crucial for quality assurance of polyherbal formulations used in various traditional systems of medicine. Moreover, in addition to their use in traditional systems of medicine currently around 40% of the drugs used globally originate directly or indirectly from natural products including plants.[14] The proper evaluation of pharmacognostic standards of medicinal plants and generation of monographs is therefore very critical.

The seeds of Tudri surkh (*Cheiranthus cheiri*) were subjected to macro and microscopical examination. Microscopic examination of plant material is crucial for its accurate identification and the revealed anatomical traits significantly aid in taxonomical characterization of a plant. The observed anatomical characteristics of a plant part provide diagnostic characteristics imperative for quality control and standardization of herbal formulations.[5]

Physicochemical screening of the test drug that include parameters such as ash value, loss on drying, pH, swelling index and powdered drug reaction with different reagents is equally vital for standardization and quality control of raw plant material employed in the manufacturing of drugs.[6] Loss on drying indicates the amount of moisture content of the drug and in powdered seeds of Tudri surkh (*Cheiranthus cheiri*) it was found to be 7.6%. Drying decreases the risk of growth of microbes (bacteria, yeast, or fungi), hydrolysis and oxidation during storage.[15] To stop the enzymatic processes, the water content must be brought down to about 10%.[6] The swelling index of test drug was 1.1% and indicates the presence of gums and mucilage, hemicellulose, or pectin in the natural drug.[5]

The ash value of seeds of Tudri surkh (*Cheiranthus cheiri*) was found to be 6% of the dry weight of the drug. The water soluble and acid insoluble ash values were found to be 1.73% and 1.33% respectively. Ash values are essential in determination of quality and purity of crude drug and indicates existence of impurities like carbonate, oxalate, and silicate. The water-soluble ash is used to estimate the number of inorganic compounds present in drugs. The acid insoluble ash consist mainly silica and indicate contamination with earthy material.[15] However, the ash content is possibly due to the Na^+ and Ca^{2+} salts which are not harmful.[6] The pH values of the drug indicate acidic or basic nature of the chemical constituents of the crude drug and seeds of Tudri surkh (*Cheiranthus cheiri*) at both 1% and 10% concentration were slightly alkaline.

The quantity of compounds in a drug extracted with solvents from a given amount of medicinal plant material denotes extractive values.[16] To estimate amount of the active constituents in a given quantity of plant material extractive values are calculated using a particular extraction solvent. The yield extract contains different chemical constituents depending upon the nature of the drug and the solvent used. It also provides an indication whether the crude drug is exhausted or not.[5,15,17] Extractive values of test drug were determined with different solvents including petroleum ether, chloroform, methanol and water. Individual cold extractive value of test drug (petroleum ether, chloroform, methanol, water) were found to be 8.9%, 11.17%, 7.7% and 2.86% respectively. While as individual hot extractive values with same solvents were 9.13%, 20.33%, 19% and 6.46% respectively. Successive extractive values by using solvents of petroleum ether, chloroform, methanol, and water were 17.96%, 3.09%, 9.18%, 4.36% respectively. The variation in the extractive values for different solvents may be due to difference in solubility of chemical constituents in these solvents.[17]

Fluorescence analysis is also an important pharmacognostic parameter. In daylight. Some constituents show fluorescence in the visible range. Many natural products which do not visibly fluoresce in day light produces fluorescence in ultraviolet light. Substances which are not fluorescent, may often be converted into fluorescent derivatives or decomposition products by applying different reagents. Hence it is an important parameter for pharmacognostic evaluation and crude drugs are often assessed qualitatively in this way.[5]

The present study carries significance as there is scarce data available about pharmacognostic description and phytochemical analysis of Tudri surkh (*Cheiranthus cheiri*) even though this drug is extensively used in Unani and other traditional systems of medicine. The limitations of the present study included study of different samples of the drug harvested at different geographical locations and grown using different agro techniques. Besides it will be of potential interest to study samples of the Tudri surkh (*Cheiranthus cheiri*) grown under controlled conditions.

CONCLUSION

Physicochemical standards are of great significance in assuring the quality, authenticity as well as purity and thereby efficacy of the drug. A major portion of global population depend on traditional systems of medicine mainly using plant origin drugs either singularly or in compound formulations for treatment of various diseases. We conclude that the data of this study can be utilized for proper identification, authentication and standardization of seeds of Tudri Surkh (*Cheiranthus cheiri*) an important drug used either in singular form or as an important constituent of many Unani and Ayurvedic compound formulations.

The data form this study will be valuable in prevention of adulteration and will help in maintaining purity of the drug. Further research to evaluate phytochemical constituents of the drug can provide exact mechanism of the pharmacological properties of the drug.

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Ethical approval: The study was approved by the Institutional Ethics Committee

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