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A detail pharmacognostic, physicochemical and phytochemical study of Kulattha churna for the management of Mutrashmari (Urolithiasis)

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Abstract
A large number of the plants are claimed to possess the medicinal properties in the traditional system and are also used extensively by the people of Assam. Kulattha (*Dolichos biflorus* L) is a very well known and reputed medicinal herb in Ayurvedic system of medicine. It is known to possess a high potential to cure many diseases like renal calculi, Fever Cough and asthma Gulma etc. So it is a very important plant in the world of Ayurvedic medicine. The plant is commonly known as Horsegram. Such traditionally used herbs Formulations are needed to be standardized for the proper use by the people and also for the establishment of a unique identification data among the common species. Present study was carried out to get a standardised data of Kulattha. Even though this plant has gained scientific importance, there is a need of standardized data. Hence, in the present work the kulattha seeds churna were subjected to various pharmacognostical and phytochemical evaluations. In the microscopical studies, the different cell structures and arrangements were studied and in physical evaluation the ash values and extractive values were studied. The various pharmacognostical constants were obtained which could help in the development of a suitable monograph for the formulation. These studies are important in the way of acceptability of herbal drugs in present scenario of lacking regulatory laws to control quality of herbal drugs.

Keywords: kulattha, pharmacogonistic study, phytochemical screening, TLC

1. Introduction
Horse gram [*Macrotyloma uniflorum* Lam. (Verdc.)], previously *Dolichos biflorus*] is an underutilized [1] and unexplored [2] food legume. It is considered as a good source of protein, carbohydrates, energy [3] It is tolerant to drought [4], salinity [5] and heavy metal stresses [6]. Horse gram mainly grown in India, Africa, Australia, Burma, Malaysia, Mauritius, and the West Indies under low soil fertility status with few inputs. It is adapted to wide range of temperature regimes [7] where other crops invariably fail to survive. In India, it is generally sown late in the rainy season by resource-poor farmers in marginal and drought-prone condition. However, sowing of seeds on the first fortnight of August and September recorded higher grain and straw yields than those sown on the first fortnight of October) [8].
The soup extract from *kulattha* (Horse gram), called *yusa*, was consumed commonly during the Sutra period (c. 1500–800 BC) are the rasams of today [9]. Horse gram is widely grown for human food as a pulse and fodder crop for livestock [10] as well as green manure and medicinal crop. In rural areas, seeds of horsegram are consumed after parched followed by boiling or frying [11] along with cooked rice, sorghum or pearl millet. Sprouted seeds, having high nutritional quality, are widely consumed by the indigenous tribal peoples [12]. Even now, in addition to its nutritive value, the consumption of sprouted seed become increasingly popular due to the excellent source to reduce the risk of various diseases and exerting health promoting effects [13]. In Indian traditional medicine, seeds of horse gram are used for treatment of urinary stones [14]; urinary diseases and piles [15], regulate the abnormal menstrual cycle in women [16] act as astringent, tonic and also used to treat calculus afflictions, corpulence, hiccups, and worms [17]. Furthermore, the cooked liquor of the horse gram seeds with spices is considered to be a potential remedy for the common cold, throat infection, fever and the soup said to generate heat [18].
There is an utmost need to standardize such traditionally used herbs formulations for the proper use by the people and also for the establishment of an unique identification data among the common species. Keeping all these in view, the present study was carried out to get a

standardized data of kulattha churna.

2. Materials and Methods

(a). Collection of plant material

Kulattha seeds were collected from Jaipur,Rajasthan The collected dried seeds were powdered after keeping some seeds in fixatives for microscopic studies and the dried material was stored in an air tight container for future use.

(b). Place of work

Pharmocognostic and phytochemical studies were carried out in State Drug Testing Laboratory, AYUSH, Govt. Ayurvedic College and Hospital, Ghy-14.

2.1 Pharmacognostic studies

Coarse powder (60 #) was used to study microscopical characters, physicochemical parameters and phytochemical investigation. For the powdered microscopical studies,slides were prepared and stained as per standard procedure [19]. The powder microscopy was performed according to the method of Khandelwal.

2.1.1 Macroscopic study

It refers to evaluation through organs of sense and includes the macroscopic appearance, color, odour, taste etc. of the drugs [20].

2.1.2 Microscopy study

The sample was treated with chloral hydrate solution and different staining reagents and chemicals were used to detect the lignified cells in the powder drugs [21]. The section was mounted on slides and studied under Trinocular Research Microscope.

2.2 Quantitative estimation

Different physicochemical properties like LOD, PH value, total ash, acid insoluble ash, extractive values of the seeds were determined using the methods described in the British Pharmacopoeia and Ayurvedic Pharmacopoeia.

2.3 Phytochemical Screening

The aqueous and methanolic extracts along with other solvent extracts of plant fruit materials were studied for various phytochemicals like alkaloids, carbohydrates, flavonoids, glycosides, gums and mucilages, phenols, tannins , reducing sugars, saponins, steroids, tannins and terpenoids by using precipitation and coloration reactions [10].

2.4 Extraction

300gm of powdered kullatha was extracted successively with solvents like petroleum ether, benzene, chloroform, acetone and methanol respectively in a Soxhlet apparatus [22]. Each solvent extract was then concentrated by distilling off the solvent under reduced pressure.

2.5 Thin layer chromatography

Thin layer chromatography was carried out with the methanolic extract and maximum spots been separated on precoated silicagel G TLC plate with trial and error methods.

2.6. Physicochemical properties

Physicochemical parameters were determined as per guidelines of WHO. Total ash value, loss on drying, water soluble ash, acid insoluble ash, solubility of the extract in different solvents, melting point, boiling point, pH, heavy metal analysis, petroleum ether soluble extractive, alcohol soluble extractive and water soluble extractive values were determined.

3. Result and observation

The kulattha churna was investigated in a systematic way covering pharmacognostical, phytochemical, and physicochemical aspects to rationalize its use as a drug of therapeutic importance.

3.1 Macroscopic characteristics

Table 1: Macroscopic characteristics

Characters	Kullatha seed (dried)
Colour	buff colour
Odour	characteristic
Touch	Rough
Size	5–6mm long, 3–4mm broad and 2–3 mm in thickness
Surface	Rough

Fig 1: Kullatha seed

3.2. Microscopical characteristics

3.2.1. Kulattha microscopy

Diagnostic characters of dried powder of kullatha under the Microscope were prismatic crystals of Calcium oxalate from epidermis, dark brown coloured content; which was confirmed to be Tannin by adding Ferric chloride solution to it, which turned black; from Sub-epidermal region, simple starch grains with Hilum, Iodine stained starch grains, Loosely arranged parenchymatous cells. Clumped masses may be protein content, with some Aleurone grains.

3.3. Determination of Quantitative standards

Table 2: Determination of Quantitative standards

Sl No	Parameters	Value
1	Ash value	
	Total ash	4.80%
	Acid insoluble ash	0.94%
2	Extractives	
	Water soluble extractive	21.34%
	Alcohol soluble extractive	11.20%
3	Loss on drying	7.14%
4	Foreign matter	0%

3.4 Chromatographic Profile of Crude Extract of Kullatha ethanolic extract

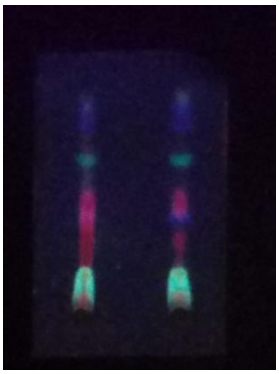

Fig 2: TLC under UV chamber

The details of solvent system and the Rf values are mentioned in the Table-3.

Table 3: The details of solvent system and the Rf values are mentioned

Extract	Solvent system	No. of spots	Rf values
Ethanol	Dichloromethane: nHexane	4	0.60 0.50 0.30 0.10

3.5 Phytochemical analysis of Pippali fruit extract

Table 4: Phytochemical analysis of Pippali fruit extract

Alkaloids	+ve
Flavonoids	+ve
Glycosides	-ve
Saponins	-ve
Tannins and phenols	+ve
Steroid	-ve
Tarpenoid	-ve

4. Discussion

The standardization of a crude drug is an integral part for establishing its correct identity. Before any crude drug can be included in an herbal pharmacopoeia, pharmacognostic parameters and standards must be established. Microscopic method is one of the simplest and cheapest methods to start with for establishing the correct identity of the source materials. The physical constant evaluation of a drug is an important parameter in detecting adulteration or improper handling of drugs. The macroscopical characters of the seed can serve as diagnostic parameters. Ash values and extractive values are important in the evaluation of purity of drugs i.e, the presence or absence of foreign inorganic matter. Extractive values are also useful to evaluate the chemical constituents present in the crude drug and also help in estimation of specific constituents soluble in particular solvents. Phytochemical analysis of the drug showed presence of alkaloids, flavonoids, sterols, tannins and phenols.

5. Conclusion

The ability to provide timely, accurate and reliable data is an essential part of discovery, development and manufacture of Pharmaceuticals. Here an attempt was made to get a standardized data of kulattha churna. The pharmacognostical, phytochemical and physicochemical characters of kulattha churna will be useful to generate standards to assess the quality and purity of the drug. The information provided by this study may be useful to carry out further study of Ayurvedic drugs of traditional medicinal practice of present era.

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