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Standardization of Tukhm-E-Konch (*Mucuna Pruriens*): An Important Unani Drug

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Abstract:- Tukhm-e-Konch (Mucuna pruriens) belongs to the family Fabaceae. It is also known as Velvet bean. It is a prime drug of Unani Medicine commonly used in the treatment of impotence, diabetes mellitus, bones fractures, cough, dog bite, cancer and madness. Due to natural variations a number of natural products have significantly different biological activity and varied clinical efficacy. Therefore, it becomes essential to standardize the herbal drugs to ensure their identity, quality and purity so as to ascertain their therapeutic efficacy. In the present study an attempt has been made to determine the physicochemical characters helpful in identification, standardization and quality control of Tukhm-e-Konch. It includes the parameters used in National Unani Pharmacopeia i.e. Total Ash values, Acid insoluble ash, Water soluble ash, successive extractive values , loss of weight on drying at 105ºC , pH at 1% & 10%, bulk density (poured & tapped density) and moisture content. Qualitative analysis and Chromatographic study (TLC) were also performed.

Keywords:- *Standardization, Tukhm-e-Konch, Mucuna pruriens, TLC.*

I. INTRODUCTION

Mucuna is the Brazilian name mentioned by "Marcgraf" in 1648, and pruriens means itching. The rigid pointed hairs on the pods if touched enter the skin and produce itching. The action appears to be purely mechanical (from the effect of the hairs on the skin) [1, 2]. *Mucuna pruriens* the drug under study has been mentioned by ancient scholars like Dioscorides (78 C.E), Galen (129-200 C.E), Ibne Baitar (1197-1248). In India this drug has been used much earlier even by Sushrut as aphrodisiac. In Europe it was introduced by Bancroft in 1769 and later it was incorporated in British Pharmacopoeia in (1783-1809)[3]. Mucuna is a wild plant found mostly in Rajasthan and Orissa regions. It is an annual climbing shrub found in bushes and hedges throughout the plains of India and in Andmans & Nicobar Island[4]. Habulkulai, Akolshi, Alkusa, Bichchoti, Kamch, Kachkuri, Gauch, Kaunch, Adhyanda, Ajada, Ajarcha, Arshabhi, Atmagupta, Kapikachhu, Kapiprabha and Kapiromaphala are some of its vernacular names[5, 6, 7]. The genus Mucuna belongs to the family Fabaceae. About 15 species are found in India. Pruriens is one of the important medicinal species used in Unani system of medicine[3]. Seeds are mainly used for medicinal purpose. Seeds are 4 or 5 in number, separated by cellular partitions, about ¼ of an inch long, ovoid, somewhat compressed, smooth brownish, mottled with black, hilum large and oblong . The Temperament or Mizaj of Tumkhm-e-konch is hot in second degree and dry in first degree[8, 9]. Seeds are used as aphrodisiac and in the formulations used to cure spermatorrhoea. It is also used in cough to expel phlegm[8], to increase appetite[10], in hemorrhoids[8], dropsy[11] and Gonorrhoea[12, 13]. Being an important medicinal herb, Konch *(Mucuna pruriens)* attracts worldwide attention for its phytochemical studies. It is a good source of Levodopa (3-4 dihydroxyphenylalanin) which is a standard drug for Parkinson′s disease. Seeds contain a number of alkaloids including nicotine, prurieninine, prurindin, prurinine etc. It also contains phosphorus, calcium, iron and manganese[1].

Due to its varied therapeutic efficacy and uses in Unani system of medicine the present study was designed to study on Tukhm-e-Konch (*Mucuna pruriens*) for certain physicochemical parameters in order to set the standards of its quality and purity.

II. MATERIAL AND METHODS

The drug was procured from the Dawakhana Tibbiya College, A.M.U, Aligarh. It was identified by Pharmacognosy Section, Department of Ilmul Advia, A. K. Tibbiya College, Aligarh Muslim University, Aligarh. The sample of the test drug was submitted to Museum of the department for future reference with the voucher No of Sc-0247/18.

The seeds of Tukhm-e-Konch (*Mucuna pruriens*) were grounded to get coarse powder. The powder was then subjected for analysis.

- *Determination of Organoleptic Characteristics*

Organoleptic evaluation includes characteristics like appearance, colour, odour, taste and texture of crude drug (Table-1).

- *Physicochemical Study*

The Physicochemical study included the estimation of successive extractive values of the test drug in various solvents, ash values, moisture content, loss of weight on drying, bulk density and pH values (Table-2).

❖ *Ash values*

➢ *Total Ash*

2 to 3 gm of powdered drug was incinerated in silica dish at a temperature not more than 450°C, until free from carbon. Then it was cooled and weighed. The total ash was calculated in terms of percentage with reference to weight of powdered drug subjected under study[14].

➢ *Water Soluble Ash*

The ash was boiled with 25 ml of water for 5 minutes. Then it was filtered and insoluble matter was collected on an ash less filter paper and then ignited for 15 minutes at a temperature not more then 450°C. The weight of the insoluble matter was subtracted from the weight of the ash and the difference in weight represented the water soluble ash.

The water soluble ash was calculated in terms of percentage with reference to weight of powdered drug subjected under study[14].

➢ *Acid Insoluble Ash*

The ash was boiled for 5 minutes with 25 ml of dilute Hcl. The insoluble matter was collected on ash less filter paper, washed with hot water and ignited to temperature less than 450°C.

The acid insoluble ash was calculated in terms of percentage with reference to weight of powdered drug subjected under study[14].

➢ *Moisture Content*

The drug was kept in a flask along with sufficient quantity of toluene. The level of toluene was kept above the level of drug to allow the later to get submerged. Then it was distilled for sufficient time. The distillate was collected in a measuring receiver along with the toluene, and a separated upper layer was measured in the receiver[15].

➢ *Loss of weight on drying*

The known weight of the test drug was taken and uniformly spread in a shallow Petri dish. It was heated at a constant temperature of 105^0C. Then it was cooled in a desiccator and weighed. The process was repeated many times till two consecutive weights were found constant. The percentage of loss in weight was calculated with respect to weight of powdered drug subjected under study[16].

➢ *pH Value*

It was carried out by a synchronic digital pH meter equipped with a combined electrode. The pH meter was standardized by using buffer solution of 4.0, 7.0, and 9.20 prior to the experiment. The pH value of 1% and 10% aqueous solution of powdered drug was determined[14].

➢ *Bulk Density*

It was measured by Digital Bulk Densitometer. A clean, dry and previously washed bottle of 250 ml capacity was filled with 100 gm of powdered test drug. It was allowed to tap till the time when no further decrease in level of drug was observed. It was calculated by the formulae-

Poured Bulk Density = Mass of powdered drug / Volume (poured) of test drug.

Tapped Bulk Density = Mass of powdered drug / Volume (tapped) of test drug[16].

➢ *Successive extractive values*

It measures the amount of a certain constituent or a group of related constituents in a particular solvent, the drug contains. The drug was extracted in different solvent in order of ascending polarity by using Soxhlet apparatus[15].

➢ *Qualitative Analysis*

It was carried out according to the scheme proposed by Bhattacharjee and Das (1969)[17] (Table-3).

➢ *Test for Alkaloids*

A drop of Dragendroff's reagent was added in the aqueous extract. The brown precipitate showed the presence of alkaloids.

❖ *Test for Carbohydrate / Sugars*

➢ *Fehling's Test*

A mixture of equal parts of Fehling's solution A and B previously mixed and added in the aqueous extract and then heated. A brick red precipitate of cuprous oxide indicates the presence of carbohydrates or sugars.

➢ *Molisch test*

In this test α-napthol was added in the aqueous extract. Then, concentrated sulphuric acid was gently poured. At the junction of the two solutions brown colour ring indicates the presence of the sugar.

➢ *Test for Flavonoids*

In the alcoholic extract of the drug a piece of magnesium ribbon was added followed by drop wise addition of concentrated Hydrochloric acid. The colour ranging from orange pink to red indicates the presence of flavonoids.

➢ *Test for Glycosides*

The test solution was filtered and sugar was removed by fermentation with baker's yeast. The acid was removed by precipitation with magnesium oxide. The remaining alcoholic extract that contained the glycosides was subsequently detected by the following method:

The hydrolysis of the solution was done with concentrated sulphuric acid and after the hydrolysis sugar was determined with the help of Fehling's solutions.

➢ *Test for Tannin*

In the aqueous extract of the drug Ferric chloride solution was added. A bluish-black colour, which disappeared on addition of dilute sulphuric acid followed by a yellowish brown precipitate indicates the presence of tannin.

❖ *Test for Proteins*

➢ *Xanthoproteinic reaction*

The concentrated nitric acid was added in the test solution. A yellow precipitate appeared. Strong solution of ammonia was added to it. Reappearance of yellow colour indicates the presence of proteins.

➢ *Biurette´s reaction*

1ml concentrated NaOH was added in the heated test solution, followed by one drop of copper sulphate solution. A violet or red colour shows the presence of proteins.

❖ *Test for Sterol/Terpenes*

➢ *Salkowski reaction*

2 ml of concentrated sulphuric acid was added in chloroform extract of drug from the side of the test tube. A red colour ring at the junction of solution indicates the presence of the sterols/terpenes.

➢ *Test for Amino Acids*

In the alcoholic extract of drug ninhydrin solution (0.1% in acetone) was added. After heating gently on water bath for few minutes it gives a blue to red-violet colour that indicates the presence of amino acids.

➢ *Thin Layer Chromatography*

Thin Layer Chromatography of different extract of drug was performed on aluminium plates precoated with Silica gel (Thickness 0.20-0.25 mm) visualized by different technique. The R_t (Retention time) of spots was calculated by the following formulae[14].

R_t Value - Distance travelled by the spot / Distance travelled by the solvent

III. RESULTS AND DISCUSSION

Nowadays Standardization is mandatory to ensure the identity of the drugs and to establish the quality of the drugs which ensures proper efficacy. So that the test drug was undergone physicochemical examination which includes Organoleptic studies of the drug which are shown in table 1, Physicochemical studies are shown in table 2. Qualitative analysis of Tukhm-e-Konch showed the presence of alkaloids, glycosides, flavonoids, proteins, amino acids, tannins and steroids (table 3). TLC of petroleum ether extract showed the presence of 3 spots (table 4).

S.NO.	Organoleptic characters	Observations
1.	Appearance	Solid and bean shape
2.	Colour	Brown to black
3.	Odour	Inodorous
4.	Texture	Firm and smooth
5.	Taste	Tasteless

Table 1:- Organoleptic characters

S.NO.	Parameters	Results (%)
1.	Ash value	Total Ash: 4.235±0.0046 Water soluble: 1.65±0.0147 Acid Insoluble Ash: 0.95±0.0073
2.	Moisture content	0.68±0.01642
3.	Bulk density: Poured Density Tapped Density	0.6±0.0142 0.8±0.0164
4.	Loss on drying at 105^0C	4.37±0.2082
5.	pH values	1 % pH- 5.8± 0.1049 10 % pH- 6.2 ± 0.3606
6.	Extractive values	Petroleum ether 4.44±0.515 Diethyl ether 0.28±0.713 Chloroform 0.59±0.397 Acetone 0.74±0.548 Alcohol 4.50±0.849 Distilled water 14.95±0.787

Table 2:- Physicochemical parameters

S.NO.	Chemical constituents	Tests/reagent	Inference
1.	Alkaloid	Dragendroff´s reagent	+
		Hager´s test	+
		Mayer´s reagent	+
2.	Carbohydrate	Molisch´s Test	-
		Fehling´s test	-
3.	Glycoside	NaOH Test	+
4.	Flavonoids	Mg ribbon and Dil. Hcl	+
5.	Tannin	Ferric chloride test	+
6.	Protein	Xanthoproteinic test	+
		Biurette´s test	+
7.	Steroid	Salkowski reaction	+
8.	Amino acid	Ninhydrin solution	+

Table 3:- Qualitative analysis of Tukhm-e-Konch (*Mucuna pruriens*)

Extract	Solvent system	Treatment	Number of spots	Rf Value
Petroleum ether	Petroleum ether: Diethyl ether (4:1)	UV Long UV Short Visible light	3 3 3	0.78, 0.52 and 0.29

Table 4:- TLC profile of Tukhm-e-Konch (*Mucuna pruriens*)

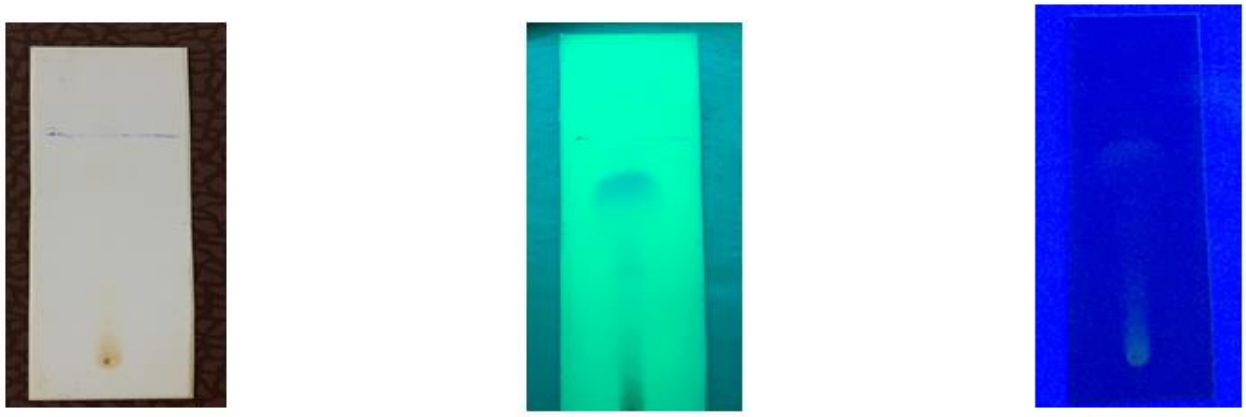

Fig 1:- TLC of Petroleum ether extract of Tukhm-e-Konch (*Mucuna pruriens*)

Standardization is very much necessary for ensuring the quality control of the herbal drugs. For the world wide acceptance of any system of medicine standardization is very important tool prior exploring the therapeutic efficacy of drug of that system of medicine. India can emerge as the major country and play the lead role in the production of standardized, therapeutically effective Unani drugs and its formulations. Standardization of herbal formulation is essential in order to assess the quality of drugs, based on the concentration of their active principles. It is an essential tool to ensure identity, purity and quality of herbal drugs. Pharmacognostical studies are the first step of standardisation which helps in identification, characterization and distinguishing the drug from confounding varieties. Since the therapeutic efficacy of a drug mainly depends upon its physicochemical characteristics therefore, the determination of physicochemical characters for the authenticity of a drug is imperative before studying it for pharmacological activity. Physicochemical study helps in characterization of constituents or groups of constituents which interact at molecular level in human beings.

Standardization of Tukhm-e-Konch (*Mucuna pruriens*) which is an effective aphrodisiac drug will ensure its proper identification, purity and quality and thereby its therapeutic efficacy. The findings of the present study will also help in distinguishing it from similar varieties which possess few common characters. The present study determines a comprehensive range of physicochemical characters of the drug according to the parameters used in National Formulary of Unani Medicine. Therefore, these findings may be used as the standards for ensuring the purity and quality and thereby the predictable efficacy and safety of Tukhm-e-Konch (*Mucuna pruriens*). The estimated information of the present study will assist in the proper identification, authentication and preventing the adulteration of medicinal plant subjected under study i.e. Tukhm-e-Konch (*Mucuna pruriens*).

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