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Standardization of Ashwagandharishta formulation by TLC Method

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Abstract: Ayurveda is a Sanskrit term for "knowledge of longevity." It is the earliest health care system of India beginning over 5,000 years ago. More than 1,200 species of plants, nearly 100 minerals and over 100 animal products comprise the Ayurvedic Pharmacopoeia Asava and Arishta are unique dosage form discovered by Ayurveda having indefinite shelf life and it was said that the "older the better it is"
Arishtas are self-generated herbal fermentations of traditional Ayurvedic system. They are alcoholic medicaments prepared by allowing the herbal juices or their decoctions to undergo fermentation with the addition of sugars.
Ashwagandha is the herb used for rejuvenation of whole body and shows immunomodulatory, adaptogenic and several other activities.In the present study Ashwagandharishta was prepared and herb as well as arishta was standardized by TLC.
Keywords: Ashwagandha, Arishta, Ayurveda, TLC, Immunomodulatory.

A. Introduction

Ayurveda is a traditional Indian medicinal system being practiced for thousands of years. More than 1,200 species of plants, nearly 100 minerals and over 100 animal products comprise the Ayurvedic Pharmacopoeia. Asava and Arishta are unique dosage form discovered by Ayurveda having indefinite shelf life and it was said that the "older the better it is". Because this dosage form has an inherent attribute of continuous hydro-alcoholic extraction and probably formation of natural analogues of the chemical compounds present in the medicinal plants.[1]

Arishtas and *asavas* are self-generated herbal fermentations of traditional Ayurvedic system. They are alcoholic medicaments prepared by allowing the herbal juices or their decoctions to undergo fermentation with the addition of sugars. *Arishtas* are made with decoctions of herbs in boiling water while *asavas* are prepared by directly using fresh herbal juices.[2]

Preparation of arishta can be done by decoction and infusion process. In this process, the crude drug is boiled in a specified volume of water for a defined time; it is then cooled and strained or filtered. This procedure is suitable for extracting water-soluble, heat-stable constituents.[3] Fresh infusions are prepared by macerating the crude drug for a short period of time with cold or boiling water. *Woodfordia fruticosa* are mostly used in asava and aishta for fermentation.[4]

The product of arishta and asava end up with 79 products out of which 38 are arishtas. Many arishtas such as arjunarishta, ashokarishta, amirtha Viswamritha, Balamritha and Swasamrutharishta are available in market.

In the present study Ashwagandharishta was prepared. Ashwagandha is one of the reputed herb in Ayurveda and has many actions on body like anti-ageing, adaptogenic, immunomodulatory, anxiety, depression, stress, cardiovascular protection, hypothyroidism to

name a few.[5] It contains alkaloids and steroidals lactones, Many bio-chemical heterogeneous alkaloids, including choline, tropanol, pseudotopanol, cuscokygrene, 3-tigioyloxytropana, isopelletierine and several other steroidal lactories, withanolides and several sitoindosides.[6,7]

B. Experimental Work

B.1 Method of preparation

In preparation of Ashwagandharishta, Ashwagandha, Musali, Yashtimadhuka Vidari, Shatavari Bramhi, Shankhapushpi, Daruharidra,Arjuna,.Sarkara, Dhataki, Sunthi Pippali,Nagkesra, curcuma was used.

The basic were first cleaned and rinsed in water to get rid of dirt. For preparation of arishta a decoction was obtained by boiling the drugs in the specified volume of water used should be clean clear and potable. When the extracts are obtained the sugar (cane sugar), jaggery and or honey are added and completely dissolved. Sometimes any one or more of these sugary substances are omitted if so directed in the recipe. The sugar jaggery and Honey should be pure the jaggery to be added should be very old (prapurana) because fresh jaggery aggravates kapha and suppresses the power of digestion. The flavoring agents are coarsely powdered and added to sweetened extract. The earthen pot or jar intended for fermenting the medicine is tested for weak spot and cracks and similarly lid is chosen.[2]

B.2 The Fermentation process

During autumn and summer season's fermentation takes place in 6 days. In winter it takes 10 days. Arishta was prepared and earthen pot was sealed with three layers of clay smeared with cloth and kept in dark place, undisturbed for a month.

B.3 Physicochemical analysis of crude drug Ashwagandha[8]

i)Total Ash Value

Heat silica crucible to red heat for 30 min , allow to cool and weigh it. Unless otherwise specified in the individual monograph weigh accurately about 1 gm substance under examination and evenly distribute it in crucible. Dry at 100^{o} to 105^{o} for 1hr and ignite to constant weight . Allow to cool after each ignition. The material should not catch fire at any time during procedure. If after prolong ignition carbon free ash cannot be obtained as directed in method. Calculate the %wt of ash on dried basis.

ii) Acid insoluble ash

Boil the ash (Total ash method) with 25ml of *hydrochloric acid* for 5min, collect the insoluble matter in ash less filter paper(Whatmann Filter paper) ,wash with hot water, ignite, cool and weigh. Calculate the percentage of acid insoluble ash on dried basis.

iii) Water insoluble ash

Boil the ash (Total ash method) for 5 min with 25 ml water, collect the insoluble matter in ash less filter paper(Whatmann Filter paper), wash with hot water and ignite for 15 min at temp less than 450^{o} .Subtract the weight of the insoluble matter from the weight of the ash; the difference in weight represents the water-soluble ash. Calculate the percentage of water soluble ash on dried basis.

iv) Sulfated Ash

2gm of Powdered (Ashwagandha) drug was taken in silica crucible and 3 ml of sulfuric acid was added. Powdered was Incinerated by gradually increasing the heat until free from carbon. and then residue was cooled in the desiccator. Ash was weighed and calculated the percentage of sulfated ash value.

v) Moisture Content

Sample was taken in tarred china dish. Dried in oven at 100^{o}c cooled .After loss of moisture is recorded. Procedure continued for at until two common readings.

B.4 Phytochemical tests

Phytochemical tests on Ashwagandha was done and steroids, alkaloids, saponins were found to be present.

B.5 Thin layer chromatography[9]

Ashwagandha was extracted with methanol.

Mobile phase used was Benzene: Ethyl Acetate (9:1) and detection was done by keeping plate in Iodine vapour chamber.

B.6 Analysis of Ashwagandharishta[10]

i) pH of Ashwagandharishta

pH **of Ashwagandharishta** was checked by the pH meter

ii) Specific Gravity of Ashwagandharishta

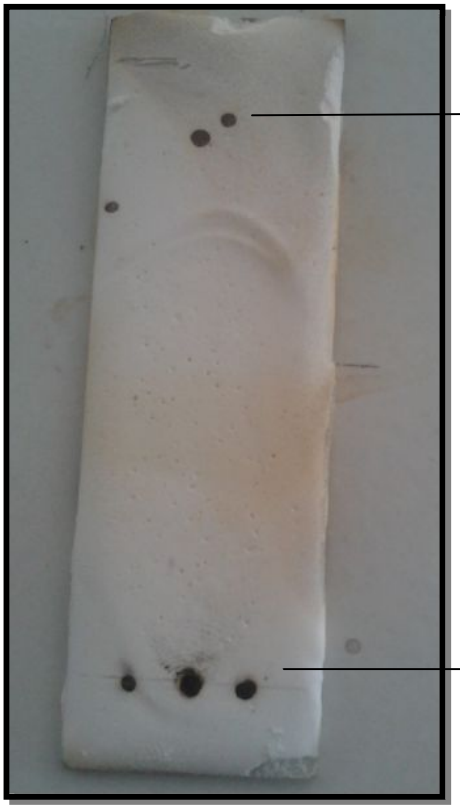
The specific gravity of **Ashwagandharishta was checked by using** specific gravity bottle.

C. RESULTS

Tests	**Results**			**Inference**
Test for extraneous matter	R_1	R_2	R_3	
Foreign organic matter	0.54	0.55	0.54	Not more than 2.0
Insect infestation	absent	absent	absent	Should be absent
Rodent contamination	absent	absent	absent	Should be absent
Physico-chemical analysis				
Total Ash content	0.2 g	0.21g	0.2g	Present in limit
Acid insoluble ash	0.23gm	0.23 gm	0.22gm	Present in limit
Water insoluble ash	0.11 gm	0.12 gm	0.11 gm	Present in limit
Moisture content	0.2gm	0.2gm	0.2gm	Present in limit

Thin layer chromatography

Solvent system used	**Detection reagent**	**Colour of spots**	**Rf value**
Benzene:EthylAcetate (9:1)	Iodine vapours	Blackish-brown	o.8



D. Summary and Discussion

The history of development of pharmaceutical dosage forms can be traced back to *Charak Samhita*, the first systematic documentation of Ayurveda. Ayurveda has recommended a comprehensive *Materia Medica* including medicinal plants, minerals, metals, and products of marine and animal origin. Medicinal plants have been used for therapeutic purposes for centuries. Initially, these were used in fresh or dried powder form, which caused the problems of high dose, high volume and low shelf life. This led to the development of extraction processes. Extracts were found to be more useful as the necessary dose was less, the volume was low and shelf life was higher. Initially the solvents used for extraction were either water or alcohol, or their mixture. Now a day's extraction procedure has become more specific depending on polarity amd solubility of compound to be extracted.

Arishtas are the unique dosage forms discovered by Ayurveda and is supposed to have indefinite shelf life and it was said that the "older the better it is". Ashwagandha is an Ayurvedic herb and many studies have been done on its theraputic potential and is very reputed drug in immunomodulation, ant-ageing and tonic for the body. Ashwagandharishta serves as general tonic for the body and helps rejuvenate mind, body and soul.

However Ayurvedic and herbal formulations are still lagging behind because of lack of standardization. In the present study Ashwagandharishta was prepared by traditional method and was standardized by TLC method. Physicochemical and phytochemical analysis was performed to confirm the chemical constituents from Ashwagandha root powder. However present study has few lacunas, formulation should be standardized by HPTLC, HPLC and pharmacokinetic profiling methods by using markers. These studies are suggested for future because of unavilibility of facilities.

This study was done with the aim to understand the benefits of Ayurvedic formulations like arishtas and need to standardize them. Study of such formulations in current scenario is of immense imporyance because asava arishtas, the self-fermented products can undergo continuous chemical transformation which goes on beyond hydro-alcoholic extraction of the suspended material. This may result in novel natural molecules with enhanced therapeutic activity.

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