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SCIENTIFIC EVALUATION OF GAJAPURISAKSARAPAKVA AVALGUJA PRALEPA- A CLASSICAL AYURVEDA FORMULATION

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ABSTRACT

Physico-chemical and phytochemical standards are of enormous consequence in assese the quality, purity and authenticity there by, efficacy of the herbal drugs. In the present scenario, the demand for herbal products is increasing exponentially throughout the world, and main pharmaceutical companies are conducting wide-ranging research on plant materials for their latent medicinal significance. Therefore, quality control for the worth and protection of herbal products is essential. Gajapurisaksarapakva Avalguja pralepa is a classical formulation mentioned in Chakradatta under kushta Adikariya. The pralepa of Gajapurisaksarapakva Avalguja consists three ingredients, viz., Elephant Dung, Elephant Urine, and Bakuchi Seed (*Psoralea corylifolia*). The attempt has been made to study Gajapurisaksarapakva Avalguja pralepa from physico-chemical and phytochemical standardization. The present study was designed to assess the quality of the Gajapurisaksarapakva Avalguja pralepa according to standard protocols. TLC fingerprint developed for Gajapurisaksarapakva Avalguja pralepa.0.1- 0.2 ml of ethanol. Gajapurisaksarapakva Avalguja pralepa was prepared according to the method described in Sushruta Samhita/*Prathisaraneeya Kshara Kalpana*. According to the results, Gajapurisaksarapakva Avalguja pralepa appeared brownish colour, sharp ordor and semi solid in nature. Physicochemical analysis of Gajapurisaksarapakva Avalguja pralepa showed water soluble ash 0.23 ± 0.02 % w/w, acid insoluble ash 0.12 ± 0.01 % w/w, total ash content 4.46 ± 0.21 % w/w, pH of 7.51±0.32 and loss on drying of 8.± 0.05 %w/w at 105 °C. The present study disclosed that the ethanol extract of Gajapurisaksarapakva Avalguja pralepa and its preliminary phytochemical screening exhibited the presence of various secondary

metabolites viz. alkaloids, phenols, steroids, saponin, tannins, flavonoids, and resin. This study would provide preliminary scientific evidence for Gajapurisaksarapakva Avalguja pralepa. Therefore, the Physicochemical and Phyto-chemical parameters of Gajapurisaksarapakva Avalguja pralepa are essential in order to achieve authentic and standard drug for therapeutic purpose and the built-up of new drug.

KEYWORDS: Gajapurisaksarapakva Avalguja pralepa, Kushta adhikariya, Physicochemical and Physico-chemical.

INTRODUCTION

Standardization plays a key role in decisive the suitability of these formulations in contemporary medicine.[1] It addresses the lack of quality control measures for herbal medicines and their formulations, which is a major challenge face by the herbal production. Physico-chemical standards are of immense significance in assuring the quality, authenticity as well as purity and there by, efficacy of the drug.[2] In the present scenario, the stipulate for herbal products is growing exponentially throughout the world, and major pharmaceutical companies are currently conducting wide-ranging research on plant materials for their potential medicinal value. Therefore, quality control for the worth and safety of herbal products is essential. Herbal medicines, particularly those applied by traditional system of medicine, have been used for thousands of years. Without analytical study, the drug study is incomplete. Analytical study of a product provides some standards to judge its quality.[3] Quality-control parameters and the developed HPTLC methods may be considered as a tool for assistance for scientific organizations and manufacturers in developing standards.[4] Clincal experience built over many centuries provides a substantialbasis for the safety and effective use of herbal medicines, not just as a main form of therapy. At present, herbal formulations have reached widespread acceptability as therapeutic agents for many diseases.[5]

Ayurveda, a part of cultural heritage of India, is widely treasured for its individuality and global recognition as it offers natural ways to treat diseases and promote healthcare. Unfortunately, standardization and quality control have remained dark shadow in the preparation of Ayurvedic medicines. The disease Switra emerged in society as colour variation in the skin and possesses a major cosmetic problem in the affected and may induce an inferiority complex in him/her. Disease of Switra affects the life of patients both biologically and psychologically due to its unsightly appearance inflicting significant

psychological stress and exerting a pernicious influence on the quality of life of patients concerning self-esteem and social interactions.

The disease Switra emerged in society as colour variation in the skin and possesses a major cosmetic problem in the affected and may induce an inferiority complex in him/her.[6] The disease Switra (Vitiligo) can be equated to 'Vitiligo' in modern medicine.[7] Disease of Switra affects the life of patients both biologically and psychologically due to its unsightly appearance inflicting significant psychological stress and exerting a pernicious influence on the quality of life of patients concerning self-esteem and social interactions.[8] The sense of being stigmatized may affect a person's interpersonal and social behaviour, which in turn increases the risk of depression.[9] But unfortunately, no scientific study has been carried out to prove the efficacy of this drug yet. Gajapurisaksarapakva Avalguja pralepa[10] is a classical formulation mentioned in Chakradatta under kushta Adikariya. It contains three ingredients, viz., Elephant Dung, Elephant Urine, and Bakuchi Seed (*Psoralea corylifolia*).

The Ayurveda Medical system has lot of medicines to cure Switra. But most of drugs have contained only herbal components. Gajapurisaksarapakva Avalguja pralepa present animal sources also. Gajapurisaksarapakva Avalguja pralepa is used as drug of choice for Switra (Vitiligo). There is lack of information regarding scientific analysis of curing of Switra, therefore characterisation of Gajapurisaksarapakva Avalguja pralepa was planned to conform its identity, quality and purity. The present study, planned to assess physiochemical screening of Gajapurisaksarapakva Avalguja pralepa.

OBJECTIVE OF THE STUDY

Evaluate the physico-chemical and phytochemical standardization of Gajapurisaksarapakva Avalguja pralepa.

METHODOLOGY OF THE STUDY

Collection of Ingredients

Bakuchi Seed (*Psoralea corylifolia*) of Gajapurisaksarapakva Avalguja pralepa was procured from the market at Gabos Lane in Pettah, Colombo, Sri Lanka, and Dung of elephant and fresh elephant's urine collected from Weediyagoda Rajamaha viharaya, Weediyagoda, at the time of preparation of drugs.

Method of Preparation of Gajapurisaksarapakva Avalguja pralepa[11]

Test drug of Gajapurisaksarapakva Avalguja pralepa prepared according to the Chakrapani Datta, Chakradatta with Vaidya Prabha. Gajapurisaksarapakva Avalguja pralepa prepared as per the standard Ayurvedic procedure of 'Prathisaraneeya Kshara Kalpana'[12] [su/su/11]. The quantities of ingredients of Gajapurisaksarapakva Avalguja pralepa calculated as per the Ayurvedic text.

Preparation of Hot -Ethanol Extract of Gajapurisaksarapakva Avalguja pralepa

Preparation of Hot -Ethanol Extract from Gajapurisaksarapakva Avalguja pralepa by using hot extraction method. A weight of 20g of powdered drug was taken into a round bottom flash containing 250ml of ethanol and refluxed for 3hr. After the extract comes to room temperature it was filtered using a muslin cloth first and then using Whatman 0.45 μm filter paper. Then the extraction was taken into a round bottom flask and solvent evaporated using a rotary evaporator. Then the round bottom flask kept in a desiccate for 24hr and weighted. The extract was used for screening of physico chemical and phytochemical analysis.

Chemicals, reagents and solvents

All chemicals, reagents and solvents used during the experimentation were of analytical grade.

Organoleptic and Physicochemical Analysis

Organoleptic characteristics of Gajapurisaksarapakva Avalguja pralepa were assessed as per the guidelines mentioned by Ayurvedic Pharmacopoeia of India[13,14,15] and phytochemical analysis, TLC was done as per the globally accepted standard guidelines. (Anonymous, 1998; Khandelwal 2007, WHO, 2011).

Preliminary phytochemical analysis of Gajapurisaksarapakva Avalguja pralepa

The extract of Gajapurisaksarapakva Avalguja pralepa was subjected to preliminary phytochemical investigations to determine the different phytoconstituents like carbohydrates, flavonoids, phenolic compounds, alkaloids, proteins, saponins, lipids, steroids and tannins using different standard methods (WHO, 2011).

Tests for Alkaloids -Two tests for Alkaloids such as Picric acid Test and Wagner's test were performed. Picric Acid Test - Yellow crystals formed proves the presence of alkaloids. Wagner's test- To 5ml of extract, added few drops of picric acid and mix well. The

appearance of reddish colour change proves that alkaloids are present.

Tests for Tanins and Phenolic Compounds- 02 drops of the Wagner's reagent were added to the 2ml of Test Solution. Ferric chloride test was performed to prove the presence of Tanins and phenolic compounds. Ferric Chloride Test- A deep blue/ dark green precipitate proves the presence of Tanins and phenolic. Few drops of 10% FeCl3 were added to 2 ml of Test Solution.

Tests for Flavonoids - Dark yellow precipitate was observed proves the presence of Flavonoids.

Tests for Saponins- To 1 ml of extract, 1 ml of 10% NaOH solution was added. Foam Test - 5 ml aqueous solution (test solution), was shaken vigorously until form a stable. Froth formation was observed proves the presence of Saponins. Kept for 10 minutes, persistent of foam. Test for Carbohydrate Benedict's test was done in to 01 ml of extract, treated with Benedict's reagent and heated gently. Purplish blue percipitate was observed proves the presence of Carbohydrates. Test for Proteins test of Xanthoproteic Test was done and yellow precipitate was observed proves the presence of proteins.

RESULTS OF THE STUDY

Organoleptic Evaluation

Table 1: Organoleptic characteristics of Gajapurisaksarapakva Avalguja pralepa.

Organoleptic characters	Observation
Colour	Dark brown
Odour	Sharp odour
Texture	Semi solid rough

Physicochemical standardization

Physicochemical parameters include extractive values, total ash value, acid insoluble ash value and water soluble ash value, moisture content, loss on drying, pH values (1% and 10% solutions) were determined to check the purity of the drug. The results of physicochemical parameters are summarized in table 2.

Table 2: Physicochemical Parameters of Gajapurisaksarapakva Avalguja pralepa.

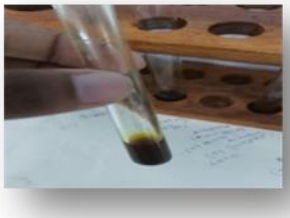
No	Parameters Values	%w/w
01	Moisture content (Loss on drying)	8 ± 0.05
02	pH values	7.51±0.32
03	Total ash	4.46 ± 0.21

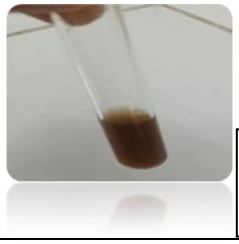
04	Acid insoluble ash	0.12 ± 0.01
05	Water soluble ash	0.23 ± 0.02
06	Alcohol soluble extractive value	3.6 ± 0.03
07	Water soluble extractive value	9.8 ± 0.03
08	Alcohol soluble extractive value	3.6 ± 0.03
09	Water soluble extractive value	9.8 ± 0.03

Preliminary phytochemical analysis

The seeds extracts in different solvents were qualitatively examined for the presence of major phytochemicals (carbohydrates, phenolic compounds, flavonoids, alkaloids, proteins, saponins, lipids, steroids and tannins). The screening of the preliminary phytochemicals showed the presence of carbohydrates, phenolic compounds, flavonoids, alkaloids, proteins, saponins, lipids, steroids and tannins. The results of phytochemicals screening are represented in table 3.

Table 3: Preliminary phytochemical tests for plant extracts.

Name of the test	Results
Picric Acid Test- Alkaloids	 Yellow crystals formed - Positive for Alkaloids.
Wagner's Test- Alkaloids	 Reddish colour appearance- Positive for Alkaloids
Benedict's Test- Carbohydrates.	 Purplish blue percipitate presented- Positive for Carbohydrates
Froth Test- Saponins	 Frothy appearance was observed- Positive for Saponins

Alkaline reagent test	 Dark yellow percipite was observed- Positive for Alkaline
Ferric Chloride Test- Tannins	 Mild green dark blue precipitate was observed -Positive for Tannins.
Xanthoproteic Test- Proteins	 Yellow precipitate was observed- Positive for Proteins

TLC fingerprint of ethanolic extract of Gajapurisaksarapakva Avalguja pralepa

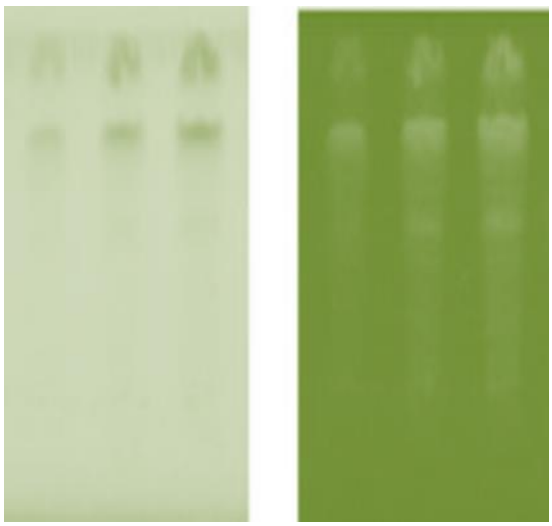


Fig. 01: TLC fingerprint.

TLC fingerprint developed for Gajapurisaksarapakva Avalguja pralepa using 0.1- 0.2 ml of ethanol. The preliminary TLC reveals that solvent system showed good separation of components.

DISCUSSION

Standardization is an important contemporary tool of analysis used to make sure the quality of herbal drugs. There are various physicochemical parameters used for the quality evaluation

of the herbal drugs. The extractive values were used to find out the active principles of the herbal drugs. The organoleptic properties indicated that Gajapurisaksarapakva Avalguja pralepa present browenish colour, sharp ordor and semi solid in nature (table 1). The ash values were used to detect the presence of any foreign matters e.g. sand and soil, water soluble salts adhering to the surface of the drugs. There is always a significant difference in the ash values of different drugs but frequently the difference varies within fine limits. The Gajapurisaksarapakva Avalguja pralepa found to be 8 ± 0.05 moisture content. Loss on Drying and Moisture Content determines the amount of moisture as well as volatile components present in a drug. Higher moisture content in the drug sample may causes hydrolysis of active ingredients of the drug and decreases its quality and efficacy. The pH values of Gajapurisaksarapakva Avalguja pralepa investigated using digital pH meter as per the standard method. The pH of the drug was found to be 7.51±0.32. (table 2). The pH of the extracts reveals that Gajapurisaksarapakva Avalguja pralepa was acidic basic compounds. Total ash of Gajapurisaksarapakva Avalguja pralepa was found to be 4.46 ± 0.21. The acid insoluble ash consists mainly of silica and low acid insoluble ash (0.12 ± 0.01) thereby indicating the no much contamination with earthy materials. The water-soluble ash (0.23 ± 0.02) was used for the quantity of inorganic elements. The validation of acid insoluble ash (0.12 ± 0.01) was also done which showed lowest content of acid insoluble ash in the extract of the seeds (table 2). The final dryness of the drug and rate of moisture removal are equally important and it was observed that the moisture content in the Gajapurisaksarapakva Avalguja pralepa was found to be 8 ± 0.05 (table 2) which showed that the drug was well dried and stored. The secondary metabolites present in the Gajapurisaksarapakva Avalguja pralepa responsible for therapeutic effect. The results of preliminary phytochemical screening showed the presence of carbohydrates, phenolic compounds, flavonoids, alkaloids, proteins, saponins, sterols and tannins, in the methanolic extract of Gajapurisaksarapakva Avalguja pralepa (table 3). Phenolic compounds, flavonoid and Tannins reported to have anti-inflammatory, antimicrobial, anticancer and anti-allergic activities. Phenolic compounds are most widely occurring groups of phytochemicals and derivatives of the pentose phosphate, and phenylpropanoid pathways in the drug. The flavonoid and phenolic compounds act as antioxidants. These active compounds are secondary metabolites which have imperative role in reconstruction and development, gives defense mechanisam as harmful pathogens (Eleazu et al., 2012). Tannins act for necessary attachment to proline rich proteins and hold up with the protein synthesis. These compounds are secondary metabolites which have vital role in reproduction and growth, gives protection against harmful predators and pathogens (Eleazu et

al., 2012). Therefore, this preliminary phytochemical screening may be useful in the detection and further quantitative analysis of such compounds. TLC fingerprint developed for Gajapurisaksarapakva Avalguja pralepa using 0.1- 0.2 ml of ethanol. The preliminary TLC reveals that solvent system showed good separation of components. Therefore, quantitative analysis of these compounds is very important to check the quality of drug. The results of preliminary phytochemical screening showed the presence of carbohydrates, phenolic compounds, flavonoids, alkaloids, proteins, saponins, sterols and tannins, in the methanolic extract of Gajapurisaksarapakva Avalguja pralepa (table 3).

CONCLUSION

Present study established the quality control parameters of Gajapurisaksarapakva Avalguja pralepa for the first time and can be used as a reference. Therefore, Organoleptic properties and physico-chemical parameters of Gajapurisaksarapakva Avalguja pralepa is essential in order to evaluate active constituents responsible for its medicinal actions and the manufacturing of new drugs.

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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