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Preparation and evaluation of fermented ayurvedic formulation: vasakasav

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Abstract
Growing awareness about harmful side effects of modern medicine has led to interest in Ayurveda at the international level as well as within India. Ayurvedic system of medicine consist of different types of dosage form such as Asava, Arishta, Taila, Ghrita, Churna, Gutika, Vati, Kwath and much more. Asava and arishtas are regarded as unique and valuable therapeutics due to their efficacy, stability and desirable features. It prepared using decoction of herbal drug and contains self-generated alcohol. Although these formulations are mentioned in traditional literature and used regularly their scientific investigation and reporting is essential to strengthen Ayurveda in global market. In present study attempt has been made for preparation and evaluation of Vasakasav. *Adhatoda vasica* is an active constituent of Vasakasav and it is mainly used to treat bronchitis and asthma. 7.20% v/v alcohol was generated after fermentation of traditionally prepared Vasakasav formulation.

Keywords: ayurveda; fermented formulation; vasakasav

Introduction
Ayurveda is combination of two Sanskrit word 'Ayu' means 'life' and 'Veda' means 'Science' and whole word means "Science of Life". Ayurveda is the ancient science known to human beings since more than 5000 years for their healing, prevention and longevity. Ayurveda has been recognized by World Health Organization (WHO) and became immensely popular in the US, Germany, Italy and many western countries. Asava and Arishtas are one of the important dosage forms in Ayurvedic system of medicine. Fermented dosage forms shows high palatability and stability [1].

Asava and arishta are the preparations that come under madhya kalpana are popular since samhita period due to their better absorption in human body and thereby quick action, longer shelf life and palatability. References of these preparations are available even during Vedic period. The emergence of sandhanakalpana was a revolutionary innovation at that period. In the course of time, various formulations indicated for multitude of disease came into existence in the form of asava/arishta [2].

Asava and Arishtas are medicinal preparations made by soaking the drugs, either in coarse powder form or in the form of decoction (Kashaya), in a solution of jaggery, as the case may be for a specified period of time, during which it undergoes a process of fermentation generating alcohol, thus facilitating the extraction of the active principles contained in the drugs. The alcohol, so generated, also serves as a preservative [2, 3].

Vasakasav is also known by the terms vasakasava, vasasav, vasarishta, vasakasavam. Vasakasav is a famous fermented ayurvedic liquid medicine and it contains about 5-10% of self-generated natural alcohol in it. This self-generated alcohol and the water present in the product acts as media to deliver water and alcohol, soluble the active herbal component of body. Its main ingredient is Vasaka (Adhatoda vasica). It is potent mucolytic and antiasthamatic. Hence, Vasakasav is used in many respiratory conditions [4, 5].

In the present study attempt has been made for formulation of Vasakasav by traditional method and its quality assessment.

Materials and Methods
Collection, identification and authentication of raw materials
The herb i.e. vasaka plant collected from the local region of Khanapur and other herbal drugs were procured form Welankar Ayurvedic Store, Sangli. In table no. 1 shows the required herbal drugs which are used for the preparation of Vasakasav formulation. Vasaka plant and other ingredients were authenticated by Mr. M. D. Wadmare sir, Dept. of Botany, Smt. Kasturbai Walchand College, Sangli.

Preparation of vasakasav by fermentation method

The coarse powder of vasa was added with required amount of water and it was boiled in a wide mouthed vessel to a quarter parts. This was filtered. Kashayam (decoction) was formed. To this kashayam jaggery was added and mixed till fully dissolved and filtered again. Now fine powders of rest of the ingredients were added inside a vessel, which was already smeared inside with ghee and stirred to form a homogeneous mixture. The vessel was sealed and kept for fermentation for 15 days. After the fermentation process is completed, the decoction was filtered and stored in clean bottles away from heat, light and moisture [1].

Table 1: composition of Vasakasav formulation

Sr. No.	Ingredient	Quantity (%)
1	Vasaka	92.9
2	Guda	46.4
3	Dhataki	3.7
4	Tvak	0.4
5	Ela	0.4
6	Tejpatra	0.4
7	Nagkeshra	0.4
8	Kankola	0.4
9	Sunthi	0.4
10	Maricha	0.4
11	Pippali	0.4
12	Hrivera	0.4

Physico-chemical evaluation

Preliminary evaluation

Determination of organoleptic characteristics viz. odour, taste, colour and clarity of prepared vasakasav was carried out.

Determination of Total phenolic content

Absorbance of standard tannic acid solutions are recorded on UV-visible spectrophotometer (Jasco, V550) at 725 nm and standard curve is plotted. Sample of vasakasav prepared as per Ayurvedic Pharmacopoeia of India and processed for estimation of total phenolic content [5, 6].

Determination Total solid content

Solid contents were determined by heating the products until the solvent was removed and heated on water bath until the residues were apparently dry, dried at 1050C to constant weight and solid contents were calculated [7].

Determination of Specific gravity

Specific gravity was measured using density bottle of 25 ml and taking mass of 25 ml of formulation and water [8].

Determination of pH

Calibrated pH meter was used to check the pH of formulation [7- 9].

Determination of alcohol content

25 ml of preparation were mixed separately with about 100 ml of water and saturated with sodium chloride. Then 100 ml of hexane was added and the mixtures and vigorously shacked for 2 to 3 minutes and allowed to stand for 15 to 20 minutes. Lower layers were run into the distillation flask, the hexane layer was washed by shaking vigorously with about 25ml of sodium chloride solution, allowed to separate and washed liquors were run into the first saline solution. Mixed solutions were made just alkaline with 1M sodium hydroxide using solid phenolphthalein as indicator. Little pumice powder and 100 ml of water were added. Mixture was distilled and not less than 90 ml of distillates were collected into 100 ml volumetric flasks and made up the volume with distilled water. Specific gravities of both the mixtures were determined and alcohol contents were calculated from the table [8, 2].

Determination of Reducing and non-reducing sugars

Clarifying reagent

Solution 1: Dissolve 21.9 g of zinc acetate and 3 ml of glacial acetic acid in purified water and make the volume to 100 ml.

Solution 2: Dissolve 10.6 g of potassium ferrocyanide in water and make up to 100 ml.

Reducing sugars

The suitable amount of the sample was taken and neutralize with sodium hydroxide solution (10% in water). Evaporate the neutralized solution to half the volume on a water bath at 50°C to remove the alcohol. Cool the solution add 10 ml of the clarifying solution 1 followed by 10 ml of the clarifying solution 2. Mix, filter through a dry filter paper and make up the volume to 100 ml. Take 10 ml of the Fehling's solution and from a burette and add sugar solution (above prepared sample) in a drop wise manner and heat to boiling over the hot plate (maintained at 800) until the mixture of Copper (Fehling's solution) appears to be nearly reduced. Add 3-5 drops of 1% methylene blue and continue the titration till the blue colour is discharged. Note down the readings and calculate the percentage of glucose [1, 7].

Non-reducing sugars

The suitable amount of sample was taken and neutralize with sodium hydroxide solution (10% in water). Evaporate the neutralized solution to half the volume on a water bath at 50°C to remove the alcohol. Cool the solution add 10 ml of the clarifying solution 1 followed by 10 ml of the clarifying solution 2. Mix, filter through a dry filter paper. To the Filter add 15 ml of 0.1 N hydrochloric acid. Cover with stopper and heat to boiling for two minutes. Add phenolphthalein and neutralize with sodium hydroxide solution (10%). Transfer to 100 ml volumetric flask and make the volume to 100 ml and perform the titration as done for the reducing sugars. Calculate the percentage of the total sugars. Subtract the percentage of the reducing sugars from the sugars to obtain non-reducing sugars [1, 7].

Results and Discussion

Organoleptic characteristics of prepared vasakasav formulation (Table no. 2) revealed that the taste of the formulation was slightly alcoholic and the appearance was clear. Measured pH indicates that vasakasav formulation have more acidic properties (Table no. 3). Self-generated alcohol itself act as preservative and improve stability of formulation, no addition of external preservative is essential. All remaining parameters were found to be within the normal range.

Table 2: Organoleptic evaluation of formulation

Parameters	Description
Colour	Brown
Odour	Aromatic
Taste	Alcoholic
Appearance	Clear

Table 3: Physicochemical evaluation of Vasakasav

Parameters	Observed value
Total phenolic content (% w/v)	0.0642±0.0008
Total solid content (% w/v)	65.94±0.005
Specific gravity	1.040± 0.0005
pH	3.39± 0.005
Alcohol content (% v/v)	7.2081±0.003
Reducing sugar (% w/v)	17.55± 0.045
Non-reducing sugar(% w/v)	0.78±0.001

Conclusion

Traditional Ayurvedic formulations are losing their value in international market. Total phenolic content, total solid content, specific gravity, pH, alcohol content, reducing and non-reducing sugar content can be considered as the basic tools for the quality control measures of asava and arishtas to improve their acceptance.

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