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A COMPARATIVE PHYTOCHEMICAL ANALYSIS OF *CHURNA* AND *BHAVITA CHURNA* OF STEM BARK OF *ASOKA – SARACA ASOCA* (ROXB.) DE WILDE

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Abstract

Bhavana is a process for potentiating or purification of drugs. *Bhavana* should be done for seven times by completely soaking the *churna* (powder) in any liquid (*swarasa, kashaya* etc) for one whole night and then drying the next day under sunlight. This increases the efficacy of the drug and hence the effective dose can be reduced. In the present study, *bhavana* of *churna* (powder) of stem bark of *Asoka* [*Saraca asoca* (Roxb.) de Wilde] was done in *kashaya* prepared of stem bark of the drug and *bhavita churna* was made. The preliminary phytochemical evaluation of *churna* and *bhavita churna* of stem bark of *Asoka* [*Saraca asoca* (Roxb.) de Wilde] was carried out. On analysing the presence of phytochemical constituents both *churna* and *bhavita churna* revealed the presence of alkaloids, flavonoids, saponins, carbohydrates, phenols, steroids and tannins. *Bhavita churna* showed more extractive and successive solvent extractive values than *churna* of the drug indicating quantitative enhancement of phytoconstituents which substantiate the fact that process of bhavana help to increase the potency and efficacy of the drug.

Key words: *Bhavana*, *Churna*, *Bhavita churna*, Stem bark, *Asoka*

I. Introduction

"*Asoka"* botanically identified as *Saraca asoca* (Roxb.) de Wilde is a well-known plant for its utility in gynaecological diseases. It is a drug which has got special consideration in treating the diseases of women. *Bhavana* is a process done in *churna* (powder) of a drug in order to change its potency. Based on the drug used for *bhavana*, one can potentiate the powder or decrease its toxicity. Acharya Caraka opines that the potency of the drug can be increased by subjecting to *bhavana* (processing) with its own liquid (*swarasa / kashaya*). The most important benefit of *bhavana* is that even a small dose of a drug can bring good effect.[1] In the present study, a comparative preliminary phytochemical analysis of *churna* and *bhavita churna* of stem bark of *Asoka* - *Saraca asoca* (Roxb.) de Wilde was undertaken to prove the increased efficacy of *bhavita churna*. The identity, purity and strength of *churna* of stem bark of drug through acceptable levels of physical parameters and extractive values are provided in Ayurvedic pharmacopoeia of India and Quality standards of Quality Standards of Indian Medicinal Plants by ICMR.The preliminary phytochemical analysis of *churna* of Asoka - *Saraca asoca* (Roxb.) de Wilde has been conducted by Suja et al and Dr. Asha S Raj. The results of phytochemical evaluation of *churna* were also compared with the above references. The preliminary phytochemical evaluation of *bhavita churna* of stem bark of the drug has not been reported yet.

II. Materials and methods

A. Collection of the drug

The *churna* (powder) and *bhavita churna* (processed powder) of stem bark of *Asoka*-*Saraca asoca* (Roxb.) de Wilde were used for preliminary phytochemical evaluation. The stem barks were freshly collected from the trees in the courtyard of Kodakara village in Thrissur district.

B. Preparation of *churna* (powder)

The fresh stem barks of the drug were washed with water thoroughly to remove residual physical impurities. The barks were kept under sunlight till the barks got properly

dried. A sufficient quantity of dried stem barks was finely powdered to obtain *churna* of the drug.

C. Preparation of *bhavita churna* (processed powder)

The *churna* of the stem bark was subjected to *bhavana* according to reference quoted in *Bhaishajya ratnavali*.[2] A sufficient quantity of whole dried stem barks was crushed and was used for the preparation of *kashaya* for *bhavana.* The crushed dried stem barks were taken in the quantity equal to that of *churna* in a mud pot. It was then added with 8 times water. The pot was kept over a firewood stove for boiling. Once started boiling, it was then reduced to 1/8th by providing mild fire. Then the *kashaya* was left undisturbed till it got completely cooled. The *churna* of the drug was taken in a clean plastic tray. It was evenly spread in a thickness of 1 cm. The prepared *kashaya* was filtered and added slowly to the *churna* after cooling such that *Kashaya* got drained into the *churna.* The powdered drug got completely submerged in *kashaya* and a thin layer of *kashaya* was left above the fine powder. The tray was shaken slowly for even distribution and the *churna* was mixed using a clean rod to ensure uniform mixing and was left undisturbed for whole night. The next day the soaked powder was dried in the sunlight and tray was covered with a clean thin cloth to avoid dust and contamination. When the top layer of soaked drug got dried, it was stirred using a clean rod to ensure uniform drying. Once properly dried, it was again powdered to remove lumps. Again, *kashaya* was prepared with crushed stem barks and added to this *churna* for next *bhavana,* which was then dried under sunlight. The *bhavana* was done in the same manner for 7 times and the process of powdering of *churna* was done after each *bhavana*. After whole procedure, the properly dried *bhavita churna* was made into fine powder and sieved through mesh with size-120.

Figure 1. ***Asoka – Saraca asoca*** **(Roxb.) de Wilde Figure**

2. Dried stem bark of ***Saraca asoca*** **(Roxb.) de Wilde**

Figure 3. ***Churna*** **(powder) of stem bark of** ***Saraca asoca*** **(Roxb.) de Wilde**

Figure 4. ***Bhavita churna*** **(processed powder) of stem bark of** ***Saraca asoca*** **(Roxb.) de Wilde**

D. Reagents

Concentrated and dilute hydrochloric acid, xylene, concentrated and dilute sulphuric acid, concentrated and dilute nitric acid, sodium hydroxide solution, lead acetate solution, sodium oxalate, potassium permanganate, anhydrous sodium carbonate, petroleum ether, cyclohexane, acetone, alcohol, Fehling's solution A&B, chloroform water, Dragendorff's

reagent, Mayer's reagent, Wagner's reagent, neutral ferric chloride, magnesium ribbon, methylene blue reagent, sodium bicarbonate solution and copper sulphate, catechol, Folin-ciocalteu phenol reagent.

E. Apparatus

Dean and stark's apparatus, Clevenger's apparatus, Soxhlet apparatus, Bunsen burner, water condensers, hot air oven, muffle furnace, heating mantle, silica crucible, round bottom flask, glass beakers, petri dishes, standard flask, measuring jars, conical flask, funnel, glass rods, watch glass, burettes, pipettes, shaker, centrifuge.

F. Procedure

Determination of the physicochemical parameters like foreign matter, total ash, acid insoluble ash, water insoluble ash, volatile oil, moisture content, fibre, tannin, total sugar, reducing sugar, phenol and pH was evaluated in *churna* and *bhavita churna* of stem bark of *Asoka* - *Saraca asoca* (Roxb.) de Wilde. The ash of both powders was subjected to qualitative analysis to confirm the presence carbonate, phosphate, chloride, sulphate and potassium. Extractive values including cold and hot watersoluble extractives and cold and hot alcohol soluble extractive values of both test drugs was evaluated in the study. Successive solvent extraction of both powders was also carried out using the solvents petroleum ether, cyclohexane, acetone and alcohol. The presence or absence of phytochemical constituents like alkaloids, flavonoids, phenols, saponins, carbohydrates, proteins, steroids and tannins were evaluated. Petroleum ether, cyclohexane, acetone and alcohol extracts of both powders were subjected to qualitative analysis for the presence of steroids, alkaloids, flavonoids and phenols. The physical and preliminary phytochemical analysis was done by standard procedures mentioned in the Ayurvedic Pharmacopoeia of India.[3]

III. Results

The preliminary phytochemical evaluation of *churna* and *bhavita churna* of *Asoka* – *Saraca asoca* (Roxb.) de Wilde obtained showed the following results.

Table 1. Physico-chemical parameters of *churna* and *bhavita churna* of stem bark of *Asoka -Saraca asoca* (Roxb.) de Wilde

Sl.No	Parameters	*Churna* (powder)	*Bhavita churna* (processed powder)
1.	Foreign matter	Nil	Nil
2.	Total ash	8.2%	7 %
3.	Acid insoluble ash	0.15%	0.65%
4.	Water insoluble ash	7 %	8.5%
5.	Moisture content	8 %	13 %
6.	Volatile oil	Nil	Nil
7.	Tannin content	43.56%	62.68%
8.	pH	5.52	5.70
9.	Fiber	24.19%	27.59%
10.	Total sugar	6.92%	1.88%
11.	Reducing sugar	1.74%	1.30%
12.	Phenol	4.91%	6.94%

Table 2. Qualitative analysis of ash of *churna* and *Bhavita churna* of stem bark of *Asoka - Saraca asoca* (Roxb.) de Wilde

Sl.No	Experiment	*Churna* (powder)	*Bhavita churna* (processed powder)
Acid radicals			
1.	Carbonates	+	+
2.	Phosphate	+	+
3.	Chloride	-	-
4.	Sulphate	+	+
Basic radicals			
1.	Potassium	-	-

Table 3. Extractive values (water soluble and alcohol soluble) of *churna* and *bhavita churna* of stem bark of *Asoka - Saraca asoca* (Roxb.) de Wilde.

Sl.No	Type of extractive	*Churna* (powder)	*Bhavita churna* (processed powder)
1.	Cold water soluble	12.06%	14.78%
2	Hot water soluble	37.3%	42.4%
3.	Cold alcohol soluble	15.54%	17.36%
4.	Hot alcohol soluble	39.1%	66%

Table 4. Extractive values (in different solvents) of *churna* and *bhavita churna* of stem bark of *Asoka - Saraca asoca* (Roxb.) de Wilde.

Sl.No	Solvents	*Churna* (powder)	*Bhavita churna* (processed powder)
1.	Petroleum ether	10.4%	13.6%
2.	Cyclohexane	1.2%	4.7%
3.	Acetone	30.4%	49.0%
4.	Alcohol	41.5%	113.6%

Table 5. Qualitative phytochemical analysis of *churna* and *bhavita churna* of stem bark of *Asoka-Saraca asoca* (Roxb.) de Wilde.

Sl.no	Experiment	*Churna* (powder)	*Bhavita churna* (processed powder)
1.	Alkaloids a. Dragendorff's test	-	-
	b. Meyer's test	+	+
2.	Flavonoids	+	+
3.	Saponins	+	+
4.	Carbohydrates a. Fehling's test	+	+
	b. Benedict's test	+	+
5.	Proteins	-	-
6.	Phenols a. Ferric chloride test	+	+
	b. Lead acetate test	+	+
7.	Steroids	++	++
8.	Tannins a. Ferric chloride test	++	++
	b. Lead acetate test	++	++

Table 6. Qualitative analysis of successive solvent extracts of both *churna* and *bhavita churna* of stem bark of *Asoka* - *Saraca asoca* (Roxb.) de Wilde.

Sl. No	Extract	Steroids	Alkaloids	Flavonoids	Phenols
1.	Petroleum ether	+	+	-	-
2.	Cyclohexane	+	+	-	-
3.	Acetone	+	+	+	+
4.	Alcohol	+	+	+	+

IV. Discussion

The *churna* and *bhavita churna*of stem bark of *Asoka* - *Saraca asoca* (Roxb.) de Wilde were subjected to preliminary phytochemical evaluation to confirm their quality and purity. The observed results in *churna*were then compared with references available in authentic textbooks and published research articles. The preliminary phytochemical evaluation of *bhavita churna* has not been reported yet and therefore references were not available to compare the results.

The physicochemical parameters such as foreign matter, total ash, acid insoluble ash of *churna* (powder) of stem bark were available from Ayurvedic Pharmacopoeia of India[4], Quality Standards of Indian Medicinal Plants by ICMR[5], research work of Suja et al[6] and Dr. Asha S Raj[7]. Foreign matter was absent in both *churna* (powder) and *bhavita churna* (processed powder) which proved these were not contaminated. Ash value is a measure of inorganic matters present as impurity. Total ash is the residue obtained after incineration of powder and was found 8.2% for *churna* and 7% for *bhavita churna* of the drug. Acid insoluble ash value mainly indicates presence of silica and silicates. Acid insoluble ash was present in 0.15% in *churna* and 0.65% in *bhavita churna* of the drug. The observed result was comparable to all four available references. Water insoluble ash was present in 7% in *churna* and 8.5% in *bhavita churna* of the drug. Water insoluble ash was similar to that of Suja et al. The three ash values were within limits and suggested that the *churna* and

bhavita churna of the drug is devoid of sand and confirmed its purity. Moisture content, volatile oil, fibre, tannin, total sugar, reducing sugar, phenol and pH were mentioned in research work of Dr. Asha S Raj and results obtained were found comparable in all parameters except for fibre and tannin content. The moisture content was 8% in *churna*and 13% in *bhavita churna* of the drug. The moisture content of *bhavita churna* had increased probably because of repeated soaking of *churna* in *kashaya* (aqueous extract) of the drug. The volatile oil content was absent in both *churna* and *bhavita churna* of the drug. Tannin content was estimated to be 43. 56% in *churna* and 62.68% in *bhavita churna* of the drug. The tannin content in *bhavita churna* might have increased as a result of repeated soaking of the powder in *kashaya* prepared with its stem bark itself. As phenolic compounds are submissive to change under the influence of biotic and abiotic factors like water availability, light availability, carbon dioxide level, nutrient content in soil and temperature, the tannin content of *churna* estimated in the present study was less compared to that obtained in research work by Dr. Asha S Raj. Both *churna* and *bhavita churna* turned blue litmus paper to red and were acidic in nature. The pH was 5.52 in *churna* and 5.70 in *bhavita churna*. The observation was similar to that obtained for research work of Dr. Asha S Raj. The fibre content in *churna* was 24.19% which has increased to 27.59% in *bhavita churna* by repeated *bhavana* (processing). When matched up with research work by Dr. Asha S Raj, the fibre content in *churna* obtained was less probably because of comparatively lower proportion of fibrous part of stem bark used in present study. The total sugar content in *churna* was 6.92% which decreased to 1.88% in *bhavita churna*. Reducing sugar was 1.74% in *churna* while in *bhavita churna* it was slightly less (1.30%). The observed total and reducing sugar were comparable to those obtained in previous research work of Dr. Asha S Raj, though a slight increase of total sugar content was noticed in *churna*. Total sugar content and reducing sugar got decreased in *bhavita churna*perhaps because of increase in polyphenolic contents like tannin and phenol as a result of *bhavana.* Phenol was 4.91% in *churna* and it increased to 6.94% in *bhavita churna* mainly because of repeated *bhavana*. As phenolic compounds in plants respond to seasonal variations, the phenol content of *churna*

estimated in the present study was more compared to that obtained in research work by Asha S Raj.

The observations of qualitative analysis of ash of powdered stem bark were obtained from research work of Dr. Asha S Raj.[7]There was presence of acid radicals like carbonates, phosphates and sulphates in both *churna* and *bhavita churna*of the drug. Acid radical chloride and basic radical potassium were absent in both powders. It was comparable except that presence of chloride and absence of phosphate was reported in above reference. Whereas in a research article published by Dharmender Rathee et al, phosphate was found in ash obtained from powdered bark.[8]

Extractive values by different solvents are used to detect adulteration. Less extractive value indicates addition of exhausted and incorrectly processed crude drugs. Extractive values of powdered drug were mentioned in Ayurvedic Pharmacopoeia of India, Quality Standards of Indian Medicinal Plants by ICMR, research article published by Suja et al and in research work of Dr. Asha S Raj.[4,5,6,7] In the present study, cold watersoluble extractive value was 12.06% in *churna* and 14.78% in *bhavita churna* of the drug. Cold alcohol soluble extractive values were 15.54% in *churna* and 17.36% in *bhavita churna* of the drug. The observed values were in normal range and comparable to references available. The quantitative estimation of hot water and hot alcohol soluble extractive values of powdered drug was conducted by Dr. Asha S Raj for her research work. Hot water soluble extractive value was 37.3% and 42.4% respectively for *churna* and *bhavita churna*in the present study. The extractive values were higher when compared to previous study indicating better quality of the drug. Hot alcohol soluble extractive value was 39.1% in *churna* while it was 66% in *bhavita churna* of stem bark. The extractive values are higher for *bhavita churna* compared to *churna* of the drug indicating its increased efficacy. In both *churna* and *bhavita churna* of the drug, the alcohol soluble extractive values were higher than water soluble extractive values indicating better extraction medium as alcohol over water. Hot extractive values were greater than cold extractive values in both *churna* (powder) and *bhavita churna* of the drug.

Successive solvent extraction is done with solvents of increasing polarity from a non-polar solvent to a highly polar solvent so that maximum phytoconstituents in the drug can be extracted out into the solvents. The values of quantitative successive solvent extractive values in petroleum ether, cyclohexane, acetone and alcohol were available in research work of Dr. Asha S Raj.[9] The extractive values of *churna* and *bhavita churna* were 10.4% and 13.6% in petroleum ether, 1.2% and 4.7% in cyclohexane, 30.4% and 49% in acetone and 41.5% and 113.6% in alcohol respectively. The values of successive solvent extraction were higher in comparison to previous research work indicating better extraction of phytoconstituents. The maximum extractive values were obtained in alcohol solvent followed by acetone in both *churna* and *bhavita churna* of stem bark. This observation was similar to those mentioned for *churna*in research work of Dr. Asha S Raj. All the values were greater for *bhavita churna* indicating its better potency compared to *churna* of the drug.

Qualitative analysis of phytoconstituents like alkaloids, flavonoids, saponins, carbohydrate, proteins, phenols, steroids, tannins of *churna* of drug was available in research work of Dr. Asha S Raj and research article of Suja et al.[6,7]Alkaloids, flavonoids, saponins, carbohydrates and phenols were present in both *churna* and *bhavita churna*. Steroids and tannins were markedly present in both the drug samples. The observed results in *churna* were supported by findings mainly in research work of Dr. Asha S raj. Protein was absent in both *churna* and *bhavita churna* of the drug. This observation in *churna* was supported by research article of Suja et al.

The observations of qualitative analysis of successive solvent extractives in powdered drug were available in research work of Dr. Asha S Raj.[9]The presence of steroids, alkaloids, flavonoids, phenols were evaluated in *churna* and *bhavita churna* of the drug. Steroids was present in all the four extracts with marked amount in acetone and alcohol extracts in both *churna* and *bhavita churna* of the drug. Presence of alkaloids was detected in all the extracts in both powdered drug samples. Flavonoids and phenols were present only in acetone and alcohol extracts in both extracts. The observed results were

similar as obtained in the research work of Asha S Raj. In addition, steroids were detected in cyclohexane extract and alkaloids in acetone solvent in the present study.

V. Conclusion

In the preliminary phytochemical evaluation, quantitative increase in tannin, fibre, phenol, extractive values and successive solvent extractive values was found in *bhavita churna* when compared to *churna* of drug, which uphold the concept of increase in therapeutic efficacy of a drug by *bhavana*. Through *bhavana* procedure, the dosage of the drug can be reduced considerably. By powdering the *bhavita churna* further, the bioavailability of the drug can also be increased.

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