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Comparative Pharmacognostical Evaluation of *Ashodhita* and *Gomutra Shodhita Bakuchi* (*Psoralea corylifolia* Linn.) Seed

Sarika Makwana, Nikhil Mehere, Swapnil Chaudhary, Prashant Bedarkar, Pro Biswajyoti Patgiri[1], C. R. Harisha[2]

Abstract

Background: *Bakuchi* (*Psoralea corylifolia* Linn.) has immense therapeutic attributes and widely prescribed drug in skin disorders especially *Shvitra* (vitiligo). Its medicinal usage is found depict in Indian pharmaceutical codex, the Chinese, British and the American pharmacopoeias and in different traditional system of medicines such as Ayurveda, Unani and Siddha. Classic emphasized the *Shodhana* of *Bakuchi* seed as pre-treatment before its therapeutic use. Considering this, study was planned for pharmacognostical evaluation of *Shodhita* and *Ashodhita Bakuchi* seeds. **Materials and Methods**: Pharmacognostical study, physicochemical analysis, and psoralen chemical test were performed for the *Ashodhita* and *Gomutra Shodhita Bakuchi* seeds powder. **Results and Conclusion:** Endosperm cells, palisade cells, and trichomes were found disturbed from their normal structure in *Ashodhita* sample. Oil globules are released from their site. Crystalline structure was observed in *Shodhita* sample. These changes were found due to the process of alkaline hydrolysis during *Shodhana* procedure. Alkaline pH (7.4) is found in suspension of *Shodhita Bakuchi* powder due to employed *Shodhana* process. Results of ash value, acid insoluble Ash, water-soluble extractives, and alcohol soluble extractives are found within normal limit as described in Ayurveda pharmacopoeia of India. These observations can be of use in future studies.

Keywords: *Bakuchi*, *Gomutra*, Pharmacognostical study, *Psoralea corylifolia* Linn., *Shodhana*

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Introduction

Bakuchi (*Psoralea corylifolia* Linn.) is one such promising drug indicated in the management of *Kushtha* (skin diseases) especially in *Shvitra* (vitiligo).[1] *Bakuchi* and its formulations have been used considerably not only by Ayurvedic physicians but also by different Indian systems of medicine. The number of researches have been reported on clinical efficacy of *Bakuchi* formulations in *Shvitra* as external application and internal administration. Although *Bakuchi* has been used in Ayurvedic therapeutics for a long without noticeable side effects, *Bakuchi* containing formulations have been reported for adverse drug reactions such as itching, blisters on applied sites in some published studies. Here, the concept of *Bakuchi* seed *Shodhana* may be important in the occurrence of such reactions. But, it is not prevalent in clinical practice of therapeutic use of *Bakuchi* seed after *Shodhana* for internal administration and external applications. Procedure for *Shodhana* of *Bakuchi* is described in Astanga Sangraha in the context of *Bakuchi Rasayana* formulation as pre-treatment of *Bakuchi* seeds.[2] Although, there is equivocation regarding requisite and process of *Shodhana* of *Bakuchi* in Ayurvedic classics. Bakuchiol and Psoralen being the major constituents and are reported to possess a number of biological properties.[3]

Consequently, proper identification is one very important part for drug development. Precise characterization and quality assurance of raw material is a requisite step to establish reproducible quality of herbal medicine which will help to justify its safety and efficacy.[4] Published report is available on pharmacognostical evaluation of *P. corylifolia* Linn. seed.[5] However, Review of literature reveals that *Shodhita* and *Ashodhita Bakuchi* (AB) seeds have not been studied in detail for pharmacognostical characters. Hence, the present work was undertaken to establ ish certain identification standards of *Shodhita* and *Ashodhita* seeds. This work will be helpful for further research with special context to *Shodhana* procedure of *Bakuchi* seeds in Ayurvedic pharmaceutics and therapeutics.

[1]Department of Rasashastra and Bhaishajya Kalpana including Drug Research

[2]Department of Pharmacognosy, Institute of Teaching & Research in Ayurveda, Gujarat Ayurved University, Jamnagar, Gujarat, India

Corresponding Author: Vd. Sarika Makwana, MD (Ayu), PhD Scholar, Department of Rasashastra and Bhaishajya Kalpana, Gujarat Ayurved University, Jamnagar - 361 008, Gujarat, India. E-mail: sarika.makwana74@gmail.com

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Materials and Methods

Collection and Authentication of Raw Materials

Bakuchi (*P. corylifolia* Linn.) seeds were collected from its geographical source at Anjangaon Surji (District: Amaravati), Maharashtra in (October month) monsoon season at the time of flowering and fruiting. At the time of collection; average temperature and humidity are 32°C and 74% respectively. It was identified and authenticated at the Pharmacognosy laboratory of the Institute and botanical identification was done with the help of various floras. *Gomutra* (cow's urine) required for *Shodhana* of *Bakuchi* seeds was collected from local cow shed, Jamnagar. Pharmaceutical procedure of *Shodhana* was performed at the Bhaishajya Kalpana laboratory of RS&BK Department.

Procedure for *Bakuchi Shodhana*

Precisely weighed AB seeds were taken in a cleaned stainless steel vessel. *Gomutra* (cow's urine) was poured on *Bakuchi* seeds till their complete immersion. Then, the mixture was left undisturbed at room temperature for overnight. On next morning, remaining mixture was filtered and cow's urine decanted through cotton cloth. Fresh cow's urine was added in the first day's *Nimajjita* (immersed) *Bakuchi* seeds. This process was repeated for further 5 days. On the 8th day, mixture was filtered through cotton cloth and *Bakuchi* seeds were washed with water for 7 times. *Bakuchi* seeds were subjected to dry at room temperature (32°C). After complete drying, they were packed in an airtight plastic container and labeled as *Shodhita Bakuchi* (SB) [Figure 1].

Macroscopic Evaluation

Powder of *Ashodhita* (AB) and *Shodhita* (SB) *Bakuchi* seeds were analyzed for the macroscopic evaluation. Organoleptic characters such as color, taste, touch, and odor were recorded. Color test was done under diffuse daylight. Surface characteristic, texture characteristic was checked out in both samples. The material was touched to determine its softness or hardness. For odor determination, firstly the strength (none, weak distinct, strong) and then the odor sensation (aromatic, fruity, musty, moldy, rancid, etc.) were assessed. Taste was recognized by taking little quantity of the powdered material.

Powder Microscopy

Powder microscopy of shade-dried powder (60#) of AB and SB was carried out. During drying process; 34°C average temperature and 50% humidity was noted. A total of 5 days was taken for shade drying process of test materials. The drugs were individually extending on glass slides and observed under microscope at different magnifications. The powder was stained with iodine solution for the detection of starch grains.[6] Photomicrographs were taken using Carl zeiss trinocular microscope attached to camera.

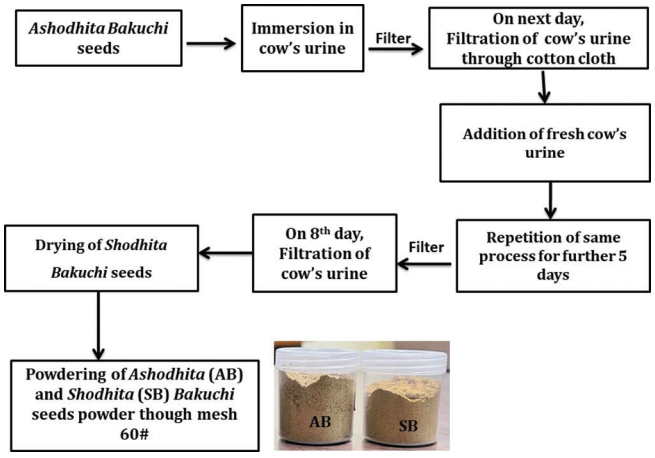

Figure 1: Procedure of Bakuchi Shodhana

Analytical Study[7]

Both the samples AB and SB were analyzed on the following parameters to develop standards. Physicochemical parameters such as pH, loss on drying (LOD % w/w), ash value (% w/w), acid insoluble ash (% w/w), water-soluble extractives (% w/w), alcohol soluble extractives (% w/w) were performed for analysis of AB and SB. All these parameters were evaluated at the Institutional pharmaceutical laboratory [Table 1].

Chemical Test of Psoralen

AB and SB were subjected for chemical test of Psoralen.[8] 2.5 g of AB and SB powder was subjected in individual conical flask. Then 50 ml of methanol was poured in it and two drops of NaOH added too. This mixture was subjected to undisturbed place for 12 h of duration. After it was filtered through filter paper and collected liquid was taken in test tubes. Test tubes were subjected in UV chamber.

Results and Discussion

Comparative organoleptic characters such as color, odor, taste, and touch for AB and SB powder were depicted at Table 2. Diagnostic characters such as bilobed trichome, brown contents, endocarp cells, oil globule, palisade cells, rhomboidal crystal, mesocarp cells were observed in AB powder. Stomata are visible in cells after staining [Figure 2]. SB powder shows black debris, brown contents, crystalline material, disturbed endosperm cells, disturbed palisade cells, disturbed Trichomes, and starch grains as diagnostic characters [Figure 3].

Cow's urine was used as *Shodhana* media for *Bakuchi* seeds. *Nimajjana* (immersion) principle is applied here for *Shodhana* process. This pharmaceutical process takes 7 days for complete *Shodhana*. As per the organoleptic characters; color, smell, and taste were differed in both the test drug samples (AB and SB). It may be due to pre-treatment (*Shodhana*) was applied on *Bakuchi* seeds.

SB powder was observed dark in color as compared with AB powder. Characteristic odor of cow's urine was observed in SB sample. It may be due to *Shodhana* of *Bakuchi* seeds in cow's urine. Pharmacognostical characters of AB sample was observed normal as mentioned above. However, pharmacognostical characters in SB sample were observe differ as compared to AB sample. Cell wall is swollen and other structures are disturbed in SB sample. As per the concentration gradient, *Shodhana* process possesses normal osmosis activity that occurs between solid and liquid system. Hence, liquid (cow's urine) is entered in the cell wall. Crystalline structure was noted in diagnostic characters of SB sample [Figure 3d]. These crystalline materials come out from the cow's urine and deposited in the cell wall. Oil globules are

Table 1: Physicochemical analysis of AB and SB powder

Samples/Tests	*Sample code*	*pH*	*LOD (%w/w)*	*Ash value (%w/w)*	*Acid insoluble Ash (%w/w)*	*Water soluble extractives (%w/w)*	*Alcohol soluble extractives (%w/w)*
Ashodhita Bakuchi seed's powder	AB	6.0	9.6	5.54	2.00	12.3	27.84
Shodhita Bakuchi seed's powder	SB	7.4	9.3	6.21	2.04	7.9	26.16

Table 2: Organoleptic characters of AB and SB powder

No.	*Organoleptic characters*	*Color*	*Touch*	*Odor*	*Taste*
1.	*Ashodhita Bakuchi* powder	Brown	Fine	Aromatic smell	Bitter-astringent
2.	*Shodhita Bakuchi* powder	Dark Brown	Fine	Strong aromatic smell with addition of cow's urine smell	Strong bitter-astringent

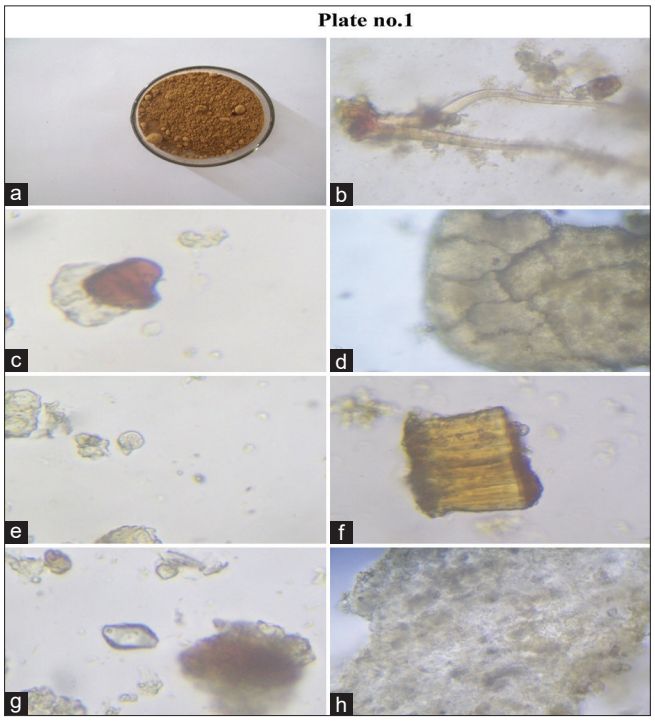

Figure 2: Pharmacognostical characters of *Ashodhita* (unprocessed) *Bakuchi*. (a) *Ashodhita Bakuchi* powder, (b) Bilobed trichome, (c) Brown contents, (d) Endocarp cells, (e) Oil globule, (f) Palisade cells, (g) Rhomboidal crystal, (h) Mesocarp cells

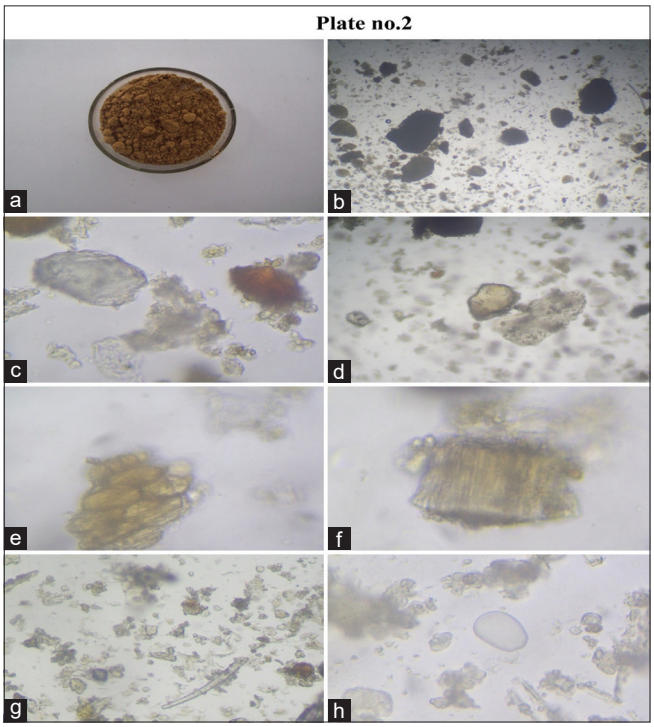

Figure 3: Pharmacognostical characters of *Shodhita* (processed) *Bakuchi*. (a) *Shodhita Bakuchi* powder, (b) Black debris, (c) Brown contents, (d) Crystalline material, (e) Disturbed endosperm cells, (f) Disturbed palisade cells, (g) Disturbed Trichomes, (h) Starch grains

released from the normal place. Endosperm cells, palisade cell, and trichomes were found disturbed [Figure 3e-g]. These changes were found may be due to employed *Shodhana* process. Principles of *Nimajjana* process may be associated to the steps of extraction procedure. Where the solvent enters through the pores into the cells and it resulting in the swelling of the tissues.

Results of comparative physicochemical analysis of AB and SB powders were summarized at Table 1. As per comparative physicochemical analysis of both the samples, more pH (7.4) is found in suspension of SB powder as compared to suspension of *Ashodhita* powder (6.0). Because alkaline hydrolysis process takes place there and alkaline hydrolysis altered the pH from 6.0 to 7.4. LOD was found nearer in both the sample drugs. Results of ash value, acid insoluble Ash, water-soluble extractives, and alcohol soluble extractives are found within the normal limit as mentioned in Ayurveda pharmacopoeia of India.[9] Percentage of Alcohol soluble extractives for AB (27.84) and SB (26.16) samples are found more than percentage of Water-soluble extractives of AB (12.3) and SB (7.9) samples. Findings denote the presence of more amounts of alcohol-soluble contents in both the samples (AB and SB). Ash usually represents the inorganic part of the plant. Ash value determination as a salient parameter to standardize the herbal drugs. High ash value is indicative of contamination, substitution, adulteration, or carelessness in preparing the drug or drug formulation. Ash values were found low indicating low contamination for both the samples.[9]

Both samples (AB, SB) were not shown any color of fluorescence at 254 nm. However, they were shown color of fluorescence at 366 nm. As per the result, powder of AB and SB were shown with yellow fluorescence at 366 nm. Powder of AB and SB was shown with yellow fluorescence at 366 nm as both contain psoralen.[10]

Conclusion

Comparative pharmacognostical study of *Ashodhita* and *Shodhita Bakuchi* powder achieved specific confirmative diagnostic characters that may be referred as standard for future researches. The process of alkaline hydrolysis possesses alteration in powder of *Shodhita Bakuchi* seeds as compared to powder of *Ashodhita Bakuchi* powder. As no published reports are available on this plant part with regard to *Shodhana*, the results obtained in the current study may be referred as a standard in future studies.

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