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Pharmacognostic and physicochemical analysis on the leaves of *Brunfelsia americana* L.

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ABSTRACT

Objective: To evaluate pharmacognostic properties including macroscopic, microscopic and physicochemical characters of the leaves of *Brunfelsia americana* (*B. americana*). **Methods:** Micro and macroscopic characters of fresh and dried leaf samples were analyzed. Physicochemical studies were done by using WHO recommended parameters and fluorescent behavior of the leaf sample were also tested. **Results:** Microscopic studies revealed the presence of anisocytic stomata, small non-glandular hairs, bicolateral vascular bundles and calcium oxalate crystals. Physicochemical parameters such as foreign matters, moisture content, extractive values, ash content, pH and fluorescent behavior of leaf powder were also determined. **Conclusions:** This is the first report on the pharmacognostic studies of *B. americana* and is helpful in the characterization of the crude drug.

1. Introduction

Plants have formed the sophisticated traditional medicine systems that have been in existence for thousands years[1-3]. The use of plants as medicines is dated back to early man[4]. They constitute an effective source of traditional and modern medicines and play an important role in health care programs.

Pharmacognosy is a simple and reliable tool, by which complete information of the crude drug can be obtained[5-8]. Today with the present surge of interest in the phyto-therapeutics, the availability of genuine plant material is becoming scarce. Since crude plant drugs form the basis for the manufacture of numerous medicinal preparations, accurate determination of drug identity forms an essential part of its study. It becomes extremely important to make an effort towards standardization of the plant material as medicine. The process of standardization can be achieved by stepwise pharmacognostic studies[9]. These studies help in identification and authentication of the plant material.

Brunfelsia americana L. (*B. americana*) (Solanaceae), is an exotic ornamental plant having medicinal properties. Its fruits are astringent and are used to cure chronic diarrhea and stomach disorders. Phytochemical investigations of the plant revealed the presence of many bioactive compounds such as steroids, flavonoids, tannins and sapoinins and its leaf extracts showed antioxidant activity[10]. It is also reported that it contains unusual fatty acids such as ricinolic acid together with cyclopropenoid and normal fatty acids[11]. However, no pharmacognostic study has been carried out on this plant and hence the objective of the present study is to evaluate various pharmacognostic properties including macro and microscopic and physicochemical characterization of the leaves of *B. americana*.

2. Material and methods

2.1. Plant material

B. americana was maintained in the botanic garden, Department of Botany, University of Kerala, Kariavattom and a voucher specimen (KUBH 5798) was deposited in the herbarium of the same department for reference.

2.2. Macroscopic and microscopic analysis

Macroscopic analysis of the plant was studied according to the method of Evans[12]. For microscopic studies, free hand sections of leaf were taken and stained with toluidine

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blue. Photomicrographs were taken using Image analyzer (OLYMPUS-BX51TF, Japan).

2.3. Physicochemical analysis

The leaves were shade dried and powdered using mechanical grinder for powder analysis. The physicochemical characteristics of powdered leaf were determined as per WHO guidelines[13]. The fluorescence characters of the plant material in different solvents were observed using visible, short UV and long UV light[14].

3. Result

3.1. Macroscopic and microscopic analysis

B. americana was a shrub, 2-3 m tall and sparsely appressed. Leaves were alternate, oblong-elliptic to obovate in nature. Leaf length to width ratio was 6-12 cm × 3-5cm with acute leaf base and obtuse apex. They were firmly coriaceous, subglabrous and yellowish green or light green in colour.

Table 1.
Physicochemical parameters.

WHO parameters	Average values %w/w leaves
Foreign matter	3.2
Moisture content	15.2
Water soluble extractive	20.9
Alcohol soluble extractive	21.2
Total ash content	10.5
Water soluble ash	14.8
Acid insoluble ash	55.2

Table 2.
Fluorescence characteristics.

Extractives	Visible light	Short UV	Long UV
Petroleum ether	Light yellow	Light green	Dark brown
Benzene	Pale green	Dark green	Brown
Acetone	Light green	Bluish green	Brown
Ethyl acetate	Green	Dark green	Reddish brown
Ethyl alcohol	Light green	Light green	Bluish brown
Methyl alcohol	Light green	Dark green	Dark blue
Distilled water	Reddish green	Bluish green	Light blue

Tranverse section of leaf through midrib showed a single layer of epidermis on both surface and was covered with cuticle. The epidermal cells were much larger on the adaxial surface than on the abaxial side. Leaves were hypostomatic and anisocytic (Figure 1). Small non-glandular hairs were seen on the adaxial surface (Figure 2). The mesophyll was divided into palisade and spongy tissue. Palisade cells were large, elongated chlorenchyma cells and were arranged in a single layer. The spongy tissue composed of loosely arranged parenchyma cells and was arranged in 3-5 layers. The midrib region of the leaves showed large bicolateral vascular bundles with patches of sclerenchyma tissues. The rest of the midrib was composed of closely arranged parenchyma cells (Figure 3). Some cells of the leaves also contained calcium oxalate crystal deposition (Figure 4).

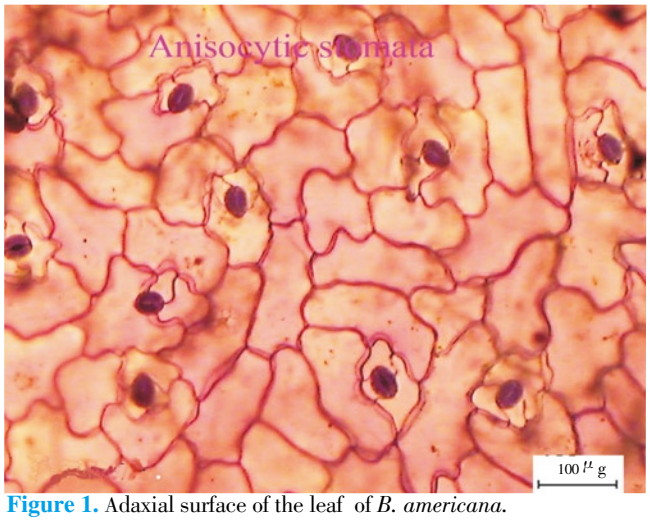

Figure 1. Adaxial surface of the leaf of *B. americana*.

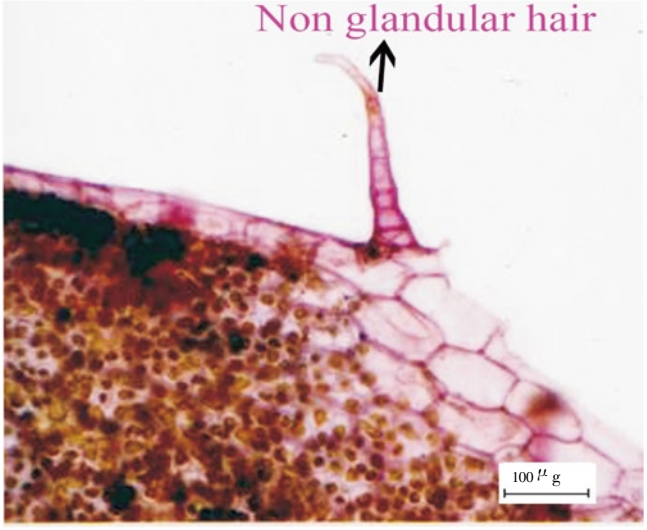

Figure 2. Adaxial surface of *B. americana* showing non glandular hair.

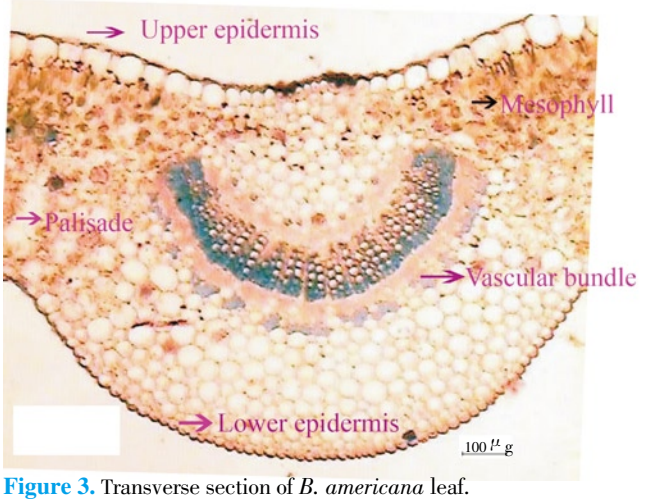

Figure 3. Transverse section of *B. americana* leaf.

3.2. Physicochemical analysis

Powder analysis showed the presence of fibres, stomata, stone cells, cork cells *etc* (Figure 5A-C). The

physicochemical characterization including foreign matter, moisture content, extractive values and ash contents were measured and shown in Table 1. The pH of the sample was noted to be 7.55. Fluorescence characteristics of leaf powder under visible, short and long UV light were determined and shown in Table 2.

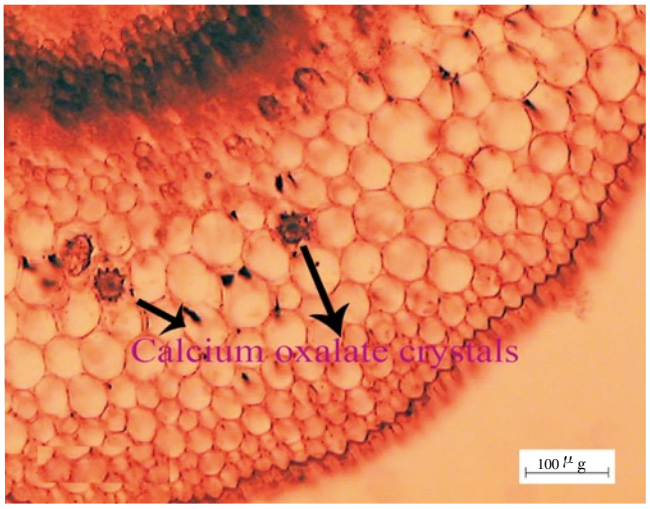

Figure 4. T.S. of leaf showing calcium oxalate crystal deposition.

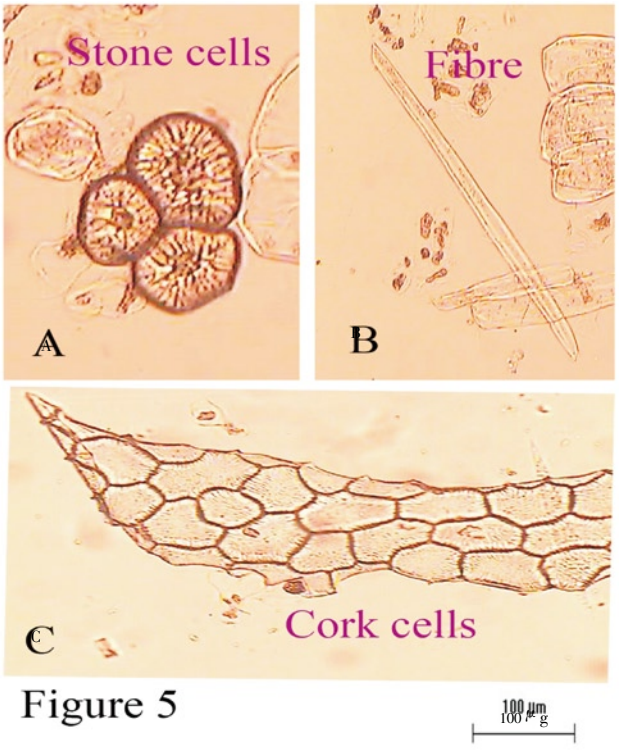

Figure 5. Powder analysis of *B. americana* leaf.

4. Discussion

The evaluation of a crude drug is an integral part of establishing the correct identification of a plant material. For this, pharmacognostic and physicochemical parameters must be determined. In this regard, the microscopic and macroscopic features of leaf have been studied. Studies revealed the presence of anisocytic type of stomata, non-glandular trichomes, bicolateral vascular bundles and calcium oxalate crystals which are the characteristic features of solanaceae family. Studies of physicochemical constants can serve as a valuable source of information and are usually used in judging the purity and quality of the drug. The extractive values give an idea about the chemical constitution of the drug and from the study, the extractive value of alcohol was highest followed by water. The ash value determines the earthy matter or inorganic composition and other impurities present along with the drug. The pharmacognostic standard for the leaves of *B. americana* is laid down for the first time in this study. To conclude, this study could be used as a diagnostic tool for the standardization of this medicinal plant and will helpful in characterization of the crude drug.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgements

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