Pharmacognostic Evaluation of *Ocimum basilicum* L

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Authors’ contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

**Aim:** Standardization of *Ocimum basilicum* through pharmacognosy.

**Place and Duration of Study:** Department of Plant Biology and Plant Biotechnology, Ethiraj College for Women, Chennai “Between” Dec 2014-April 15.

**Methodology:** Organoleptic evaluation was carried out based on sensory characters. A free hand anatomical section of the stem was observed. Powder analysis, maceration, Phytochemical test and Fluorescence analysis were conducted according to standard protocol.

**Results:** The organoleptic characters of the dried leaves of *Ocimum basilicum* were green, aromatic, pungent and brittle in texture with anomocytic stomata with stomatal index 71.87% in the lower epidermis. A prominent bundle sheath in the leaf was evident. The macerated stem showed annular xylem vessels. The aqueous extract showed the presence of alkaloids, phenols, flavonoids, steroids, terpenoids, and glycosides. Alkaloids and lignins were evident in the histochemical study.

**Conclusion:** Pharmacognostic evaluation of *Ocimum basilicum* would help in identification, detection of adulterants and development of a monograph.

**Keywords:** *Ocimum basilicum*; anomocytic stomata; phytochemicals; fluorescence analysis.

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1. INTRODUCTION

Herbal drugs have been used in Ayurveda, Siddha and Unani healing systems since ancient times. According to WHO 80% of the world population depend on the herbal drugs for health care. Ancient Indian literature incorporates a remarkably broad definition of medicinal plants and considers all plants parts to be potential source of medicinal substances [1]. However a key obstacles which has hindered the acceptance of the alternative medicines in the developing countries, is the lack of documentation and stringent quality control. There is a need for documentation of research work carried out on traditional medicines [2]. With this backdrop, it becomes extremely important to make an effort towards standardization of medicinal plant. Pharmacognostic study is potential tool to standardize crude drugs. Pharmacognostic evaluation gives valuable details of the crude drug and also determines the safety and efficacy.

_Ocimum basilicum_ L. (sweet basil) a perennial herb belongs to Lamiaceae. Sweet basil has been used extensively in Ayurveda and Unani medicine [3]. The plant is rich source of phenolics, flavonoids and terpenoids [4]. It has been used to treat various ailments such as poor digestion, nausea, migraine, depression, insomnia, kidney malfunction and skin infections [5,6]. The plants is also reported to possess exceptional biological activities such as, antioxidant [7,8] antifibrotic, anticancer, radioprotectant [9], anti-inflammatory effects, immunomodulatory activity, anticonvulsant[10], anti-stress activity, anti-pyretic activity, antimicrobial, anti-arthritic activity, anti-diabetic activity, repellent[11], prophylactic agent and in cardiovascular disease [12,13]. It is also used as a kitchen herb, culinary herb and ornamental herb. Essential oils extracted from fresh leaves and flowers can be used as aroma additives in food, pharmaceuticals, cosmetics and to improve shelf life of food products [14]. Pharmacognostic evaluation on such valuable plant is very less and hence this study was conducted which will help determination of safety and efficacy of the drug.

2. MATERIALS AND METHODS

2.1 Organoleptic Characters and Microscopic Characters

The external sensory and morphology of the dried leaf sample was observed for the colour, taste, aroma and texture. The leaves of _Ocimum basilicum_ in FAA (Formalin 5 ml + Acetic Acid 5ml + 70% Ethanol 90 ml) were subjected to epidermal peeling and transverse sectioning. The section were stained in Safranin (1%) and mounted in glycerol. The stomatal index was calculated by the formula =No of stomata/No of epidermal cell× 100.

2.2 Anatomical Study

Free hand section of stem and leaves were taken, stained with safranin and mounted in glycerol and observed under light microscope and photographed.

2.3 Maceration

The stems of _Ocimum basilicum_ were cut into small piece, boiled in water repeatedly for 3-5 times to expel air until the pieces settled down. Treated pieces were soaked in Jeffery's fluid (equal volume of 10% of nitric acid and 10% of chromic acid) for 24 hours at 30-40º C, decanted, washed and stored in 50% alcohol. Pieces of macerated stem were, dehydrated through alcohol series (50%, 60%, 70%, 80%, 90%, 100%) for five minutes and passed through alcohol: xylol (1:1 ratio) series for five minutes, macerated and observed.

2.4 Histochemical Test

The sectioned stem was treated with wagners reagent (potassium iodide and iodine) for detection of alkaloid, orcinol in sulphuric acid for detection of gums, Toluidine blue O for lignin, Sulphuric acid for crystals, ferric chloride (1N Hydrochloric acid) for tannin, methylene blue test for phenols and copper acetate test for Terpenoids.

2.5 Phytochemical Analysis

The leaves washed dried and powdered. Aqueous extracts prepared was used for phytochemical test according to the standard procedures described by Kokate[15] and Horborne[16].

2.6 Powder and Fluorescence Analysis

The leaves of _O.basilicum_ were powdered and sieved to obtain fine powder. The powder was placed on a clean slide and observed under microscope. The dried leaf powder was treating with several drops of specified reagent like
Fig. 1. Pharmacognistic evaluation of Ocimum basilicum

1- Habit of Ocimum basilicum 2- Epidermal peeling – Anomocytic Stomata 3- C.S of leaf stained with safranin 4- T.S of stem stained with safranin 5- Histochemical test of stem for Alkaloids (Note the yellowish brown) 6- Histochemical test tannin in stem with Toluidene blue O  7 & 8 - Maceration of stem - Phloem Fiber and Annular thickening in the xylem vessels. 9- Powder analysis - Trachieds

hydrochloric acid, sodium hydroxide, water, nitric acid, and sulphuric acid. were observed under UV 365nm and the emitted fluorescence was observed that helps in identifying the drug in powdered form[17].

3. RESULTS AND DISCUSSION

3.1 Organoleptic and Microscopic Characters

The organoleptic characters of dried leaf were green aromatic, pungent and brittle in texture. The microscopic analysis of the leaf peel showed stomatal index of Ocimum to be 71.87. The stomata were anomocytic in nature in contrary to the observations made by Pooja et al.[18] where it was diacytic. In another variety of O. basilicum the stomatal index was only 23-33 /mm²[19]. The stomatal index varies from one variety to another. It depend on the environmental condition in which the plant has grown.

3.2 Anatomical Study

The transverse sections of presents a square contour with prominent epidermis made of isodiametric cells with internal and external tangential walls. The epidermis is covered with thin cuticle. Cortex made up of 5-6 layers of parenchymatous cell. Several sclerenchyma
patches were also evident (Fig 1). Single layer of primary endodermis with multilayered pericycle and eustele present. Pith parenchymatous in nature. Anatomical sections of Ocimum basilicum leaf shows upper and lower epidermis. Mesophyll differentiated as elongated palisade and spongy tissue. Scleranchymatous bundle sheath with Vascular bundle conjoint and exarch. Venugopalan [19] reported the absence of bundle sheath in leaf of O. basilicum L var. pilosum (willd.) Benth which was observed in the present study.

3.3 Powder Analysis and Maceration

The powder analysis showed tracheids and fibers. The macerated stem showed annular thickening in the xylem vessels and phloem fibers were observed (Fig 1).

3.4 Histochemical and Phytochemical study

The stem sections showed pigmentation indicating presence of alkaloids. Presence of lignin was found. The histochemical study was in accordance with the phytochemical test. The aqueous extract of Ocimum basilicum showed the presence of alkaloids, phenols, flavonoids, steroids, terpenoids, and glycosides. (Table 1). Pooja et al. [18] reported the presence of Triterpenoids, glycosides, carbohydrate, polyphenols and mild amounts of tannins in ethanolic extracts. These secondary metabolites are responsible for various therapeutic activities in the plants [20]. Due to its sharp pungent smell, the essential oil of this plant may have the potential to be used to combat microbial infections. In addition the aromatic principles of the plant may have mosquito properties against and commercially viable aromatic principles and phytochemicals offer wide applications in human health care.

3.5 Fluorescence Analysis

Under Whitelight the O. basilicum powder appeared to be green in Hydrochloric, dark green in Sodium hydroxide & methanol and Sodium hydroxide & water treatment. However, it was greenish orange in nitric acid brown in sulphuric acid treatment. Under Long UV light it appeared dark brown hydroxide & methanol and Sodium hydroxide & water treatment. Red and Reddish orange emitted in Nitric acid and Sulphuric acid respectively (Table 2).

4. CONCLUSION

O. basilicum L ‘King of Herbs’ has been used extensively in traditional medicine to cure various ailments. The leaves are rich source of...
phytochemicals such as alkaloids, phenols, flavonoids, steroids, terpenoids, and glycosides as evident in the histochemical and phytochemical study. The stomatal index distinct, with anomocytic stomata. The fluorescent analysis of O. basilicum showed characteristic emission. This study not only helps in detecting the adulterants but also helps in understanding the variations in pharmacognostic characters of the medicinal plant.

CONSENT
Not applicable.

ETHICAL APPROVAL
Not applicable.

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COMPETING INTERESTS
Authors have declared that no competing interests exist.

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