

PHYTOCHEMICAL INVESTIGATION AND PHARMACEUTICAL APPLICATION OF SACRED PLANT *OCIMUM CANUM*

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ABSTRACT

The medicinal plants are widely used by the traditional medical practitioners for curing various diseases in their day to day practice. In traditional systems of medicine, different parts (leaves, stem, flower, root, seeds and even whole plant) of *Ocimum canum Linn* (known as kalaTulsi in Hindi), a small herb seen throughout India, have been recommended for the treatment of bronchitis, bronchial asthma, malaria, diarrhea, dysentery, skin diseases, arthritis, painful eye diseases, chronic fever, insect bite etc. The *Ocimum canum L.* has also been suggested to possess antifertility, anticancer, antidiabetic, antifungal, antimicrobial, hepatoprotective, cardioprotective,

antiemetic, antispasmodic, analgesic, adaptogenic and diaphoretic actions.

KEYWORDS: Tulsi, medicinal plant, *Ocimum Canum Linn*, phytochemical constituents, traditional uses.

INTRODUCTION

Natural products are important sources for biologically active drugs.^[1] There has been an increasing interest in the medicinal plants as natural products in different parts of the world.^[2] Medicinal plants containing high antioxidant properties play an important role in the prevention of various degenerative diseases in the society. The medicinal value of these plants depends on bioactive phytochemical constituent's action in the human body. Some of the most important bioactive phytochemical constituents include alkaloids, flavonoids, essential oils, tannins and saponins.^[3] Phenolics are commonly found in medicinal plants and their biological effects, include antioxidant activity. Due to synthetic antioxidants such as butylated hydroxyl anisole (BHA), butylated hydroxyl toluene (BHT) and tert-butyl hydro

quinone (TBHQ), which are widely used in food industry and cosmetic, have been growing concern over the possible carcinogenic effects.^[4] Thus interest in natural antioxidant has increased considerably. Nowadays, it is well known that natural antioxidants extracted from herbs and spices have high antioxidant properties and are used in many food applications⁵. Natural antioxidants from plant sources are potent and safe due to their harmless nature; wild herbs have their antioxidant properties. Tulsi, the Queen of herbs, the legendary 'Incomparable one' of India, is one of the holiest and most cherished of the many healing and healthy giving herbs of the orient. The sacred Tulsi, is renowned for its religious and spiritual sanctity, as well as for its important role in the traditional Ayurvedic and Unani system of holistic health and herbal medicine of the East. It is mentioned by Charaka in the Charaka Samhita; an Ayurvedic text. The important advantages claimed for therapeutic uses of medicinal plants in various ailments are their safety besides being economical, effective and their easy availability.^[7,8] Because of these advantages the medicinal plants have been widely used by the traditional medical practitioners in their day to day practice. According to a survey (1993) of World Health Organization (WHO), the practitioners of traditional system of medicine treat about 80% of patients in India, 85% in Burma and 90% in Bangladesh.^[8,9] In traditional systems of medicine the Indian medicinal plants have been used in successful management of various disease conditions like bronchial asthma, chronic fever, cold, cough, malaria, dysentery, convulsions, diabetes, diarrhea, arthritis, emetic syndrome, skin diseases, insect bite etc and in treatment of gastric, hepatic, cardiovascular & immunological disorders.^[10]

2.1- PLANT PROFIL	
Botanical Name:	<i>Ocimum canum L,</i>
Family Name:	Lamiaceae
Synonym:	Manjari, wild basli
Taxonomy:	-Kingdom - <i>Plantae</i> – Plants Subkingdom- <i>Tracheobionta</i> – Vascular plants, Superdivision <i>Spermatophyta</i> – Seed plants
Division:	<i>Magnoliophyta</i> – Flowering plants, Class - <i>Magnoliopsida</i> – Dicotyledons Subclass - <i>Asteridae</i> , Order – <i>Lamiales</i> Genus - <i>ocimumL.</i> R--bsali. Species - <i>ocimum canum</i> Sims hoary basli
Vernacular name -	Hindi- Kali tulsi, English-Holi tulsi

Members of the genus

About some species, including

Ocimum americanum, *Ocimum basilicum*, *Ocimum camechiamum*, *Ocimum fruticosum*, *Ocimum gratissimum*, *Ocimum kilimandscharicum*, *Ocimum tenuiflorum*, *Ocimum minimum*.

2.2-CHEMICAL CONSTITUENTS

The phytochemical and nutrient compositions of its leaves were investigated using established analytical procedures. The phytochemical analysis of the plant revealed high concentrations of flavonoids (10.00%), saponins and tannins, but low levels of phenolics and alkaloids. The leaves showed high carbohydrate content (639.6 g/kg), ash, crude fat and crude fiber, but very low in protein content. The study reveals high concentration of calcium (50.72 g/kg) with appreciable levels of potassium, sodium, phosphorous and magnesium. In addition, the plant was found to be a good source of iron, zinc and manganese. Furthermore, the concentrations of cadmium (0.01 g/kg) and lead (0.02 g/kg), which are toxic metals were very low, while the vitamin C content of the leaves was found to be high (0.05 g/kg). The result of this study therefore revealed that the leaves of *O. canum* are a good source of phytochemicals and nutrients that can be harnessed to combat nutritional deficiencies, especially in the rural communities. Leaves and the seeds contain an essential oil consisting of Ursolic acid, apigenin and luteolin. Some other main chemical constituents of Tulsi are oleanolic acid, Rosmarinic acid, Eugenol, carvacrol, Linolool and beta caryophyllene.

MATERIALS AND METHODS

3.1. Plant collection

Leaves of *Ocimum canum* were collected in the month of November 2011 from its natural habitat from nearby Dasapalla forest division, Nayagarh district of Odisha. The plant was authenticated from National Botanical Research Institute (NBRI) Lucknow by Dr C.H.VRao. The leaves were cleaned and dried under the shade to avoid degradation of volatile oil.

3.2. Extraction

The dried leaves were coarsely powdered and extracted with Petroleum ether and water by a Soxhlet apparatus at 50°C. The solvent was completely removed and obtained dried crude extract which was used for investigation. Further the extracts were subjected for the phytochemical study as well as pharmacological screening.

2.3. Phytochemical screening

Phytochemical screenings were performed using standard procedures.^[11,12,13]

1) **Test for alkaloids:** To the extract dilute hydrochloric acid will be added and filtered. The filtrate will be treated with various alkaloidal reagents.

- a) **Mayer's test:** The filtrate will be treated with Mayer's reagent: appearance of cream colour indicates the presence of alkaloids.
- b) **Dragendroff's test:** The filtrate will be treated with Dragendroff's reagent: appearance of reddish brown precipitate indicates the presence of alkaloids.
- c) **Hager's test:** The filtrate when treated with Hager's reagent, appearance of yellow colour precipitate indicates the presence of alkaloids.

2) Test for carbohydrates and reducing sugar

The small quantities of the filtrate will be dissolved in 4ml of distilled water and filtered. The filtrate will be subjected to

- a) **Molisch's test:** A small portion of the filtrate will be treated with Molisch's reagent and sulphuric acid. Formation of a violet ring indicates the presence of carbohydrates.
- b) **Fehling's test:** The extract will be treated with Fehling's reagent A and B. The appearance of reddish brown colour precipitate indicates the presence of reducing sugar.
- c) **Benedict's test:** The extract will be treated with Benedict's reagent; appearance of reddish orange colour precipitate indicates the presence of reducing sugar.
- d) **Barfoed's test:** The extract will be treated with barfoed's reagent and heated. Appearance of reddish orange colour precipitate indicates the presence of non-reducing sugars.

3) Test for steroids

Liebermannburchard's test: The extract will be treated with 3ml of acetic anhydride, few drops of glacial acetic acid followed by a drop of concentrated sulphuric acid. Appearance of bluish green colour indicates the presence of steroids.

4) Test for proteins

Biuret test: The extract will be treated with copper sulphate solution, followed by addition of sodium hydroxide solution; appearance of violet colour indicates the presence of proteins.

- a) **Millon's test:** The extract will be treated with Millon's reagent; appearance of pink colour indicates the presence of proteins.

5) Test for tannins

The extract will be treated with 10% lead acetate solution; appearance of white precipitate indicates the presence of tannins.

6) Test for phenolic compounds

- a) The extract will be treated with neutral ferric chloride solution; appearance of violet colour indicates the presence of phenolic compounds.
- b) The extract will be treated with 10% sodium chloride solution; appearance of cream colour indicates the presence of phenolic compounds.

7) Test for flavonoids

- a) 5ml of extract will be hydrolyzed with 10% sulphuric acid and cooled. Then, it will be extracting with diethyl ether and divided in to three portions in three separate test tubes. 1ml of diluted sodium carbonate, 1ml of 0.1N sodium hydroxide and 1ml of strong ammonia solution will be added to the first, second and third test tubes respectively. In each test tube, development of yellow colour demonstrated the presence of flavonoids.
- b) Shinoda's test: The extract will be dissolved in alcohol, to which few magnesium turnings will be added followed by concentrated HCL drop wise and heated, and appearance of magenta colour shows the presence of flavonoids.

8) Test for gums and mucilage

The extract was treated with 25 ml of absolute alcohol and filtered. The filtrate will be examined for its swelling properties.

9) Test for glycosides

When a pinch of the extract was treated with glacial acetic acid and few drops of ferric chloride solution, followed by the addition of conc. Sulphuric acid, formation of a ring at the junction of two liquids indicates the presence of glycosides.

10) Test for saponins

Foam test About 1 ml of the extract was diluted to 20 ml with distilled water and shaken well in a test tube. The formation of foam in the upper part of the test tube indicates the presence of saponins.

11) Test for Triterpenoids

The substance was warmed with tin and thionyl chloride. Pink colour indicates the presence of triterpenoids.

Table -Phytochemical analysis of *Ocimumcanum*leaves

S. No.	Phytoconstituents	Petroleum ether	Aqueous
1	Alkaloids	-	-
2	Carbohydrates	+	+
3	Glycosides	-	-
4	Phytosterols	-	-
5	Fixed oils	-	-
6	Saponins	-	-
7	Tannins	+	+
8	Protein and amino acids	-	-
9	Gums and mucilage	-	-
10	Flavonoids	+	+
11	Terpenoids	+	+

+ = presence, – = absence.

PHARMACEUTICAL APPLICATION OF OCIMUM 4.1-Healing power

The tulsi plants have many medicinal properties. The leaves are used as nervine tonic and also enhance memory power. They promote the removal of the catarrhal matter and phlegm from the bronchial tube. The leaves strengthen copiously the stomach and induce preparation. The seed of the plant are mucilaginous.

4.2-Skin Disorders

Applied locally, ocimum juice is beneficial in the treatment of skin diseases. It has also been tried successfully by some naturopathy in the treatment of leucoderma.

4.3-Coughs

Water boiled with ocimum leaves can be taken as drink in case of sore throat. This water can also be used as a gargle.

4.5-Respiratory Disorder

The herb is useful in the treatment of respiratory disorder. A decoction of leaves with Honey and ginger is an effective remedy for bronchitis, asthma, influenza, cough and cold, decoction of leaves, cloves and common salt also gives immediate relief in case of influenza. They should be boiled in little quantity of water and then taken.

4.6-Kidney stone

ocimum has strengthening effect on the kidney. In case of renal stone the juice of ocimum leaves and honey, If taken regularly for 6 months it tract. will expel stones via the urinary.

4.7-Heart disorder

Ocimum canum has beneficial effect in cardiac disease and the weakness resulting from them. It reduces the of blood cholesterol. Common pediatric problems like cough, cold, fever diarrhea and vomiting respond favorably to the juice of basil leaves, If pustules of chicken pox delay their appearance, ocimum leaves taken with saffron will hasten them.

4.8-Stress

Ocimum canum leaves are regarded as an adeptogen or anti stress agent. Recent studies have shown that the leaves afford significant protection against stress. Even healthy person can chew 12 leaves of ocimum, two a day, to prevent stress. It purifies blood and help prevent several common eliments.

4.9-Mouth infection

The leaves are quite effective for the ulcer and infection in the mouth. A few leaves chewed will cure these element.

4.10-Insect bites

The herb is a prophylactic or preventive and curative for insect stings or bites. A tea spoonful of Fresh leaves juice may be applied to the affected parts, a paste of fresh root is also effective in case of bites of insects.

4.11-Teeth disorder

The herb is useful in teeth disorder. The dried leaves powder can be used for brushing teeth. It can also be mixed with mustered oil to make a paste and used as a toothpaste. This is a very good herbal drug for maintaining dental health, counteracting bad breath and for massaging the gum. It is also useful in pyorrhoea and other teeth diseases.

4.12-Headaches

Ocimum is a good medicine for headache. The decoction of the leaves can be given for this disorder. Powder leaves mixed with sandal wood paste can also be applied on the forehead for getting relief from headache.

Ocimum juice is an effective remedy for sore eye and night –blindness, Which is generally caused by deficiency of vitamin. Two drops of black basil juice are put into the eye daily at bed time.

4.13-Anti cancer activity

The anti cancer activity of ocimum has been proved. The alcoholic extract(ALE) OF leaves of ocimum has a modulatory influence on carcinogen metabolizing enzyme stouch as cytochrome P450, Cytochrom b aryle hydrocarbon, hydroxylase and glutathione S – transferase (gst), which are important in detoxification of carcinogen and mutagen. The anti cancer activity of os has been reported against human fibrosarcoma cells culture.

4.14-Lipid lowering activity

Fresh oimum leaves causes significance change in the lipid profile of normal albino rat. This results significant lowering in serum total cholesterol triglycerides, phosholipid LDP Cholestrol.

4.15-Anti malarial effect

Essential oil of ocimum has been reported to possess 100% larvicidal activity against the culex mosquitoes. Ocimum have excellent anti malarial activity.

4.16-Anti allergic and immunomodulatory effect

Essential oil of ocimum was found to have anti allergic properties. When administered to laboratory animal, the ocimum was found to inhibit mast cell degranulation and histamine release in the presence of allergen. These studies reveal the potential role of ocimum canum extracted in the management of immunological disorder including allergic and asthma.

4.17-Anti fertility effects

One of the major constituents of the leaves, urosolic acid possess anti fertility activity in rats and mice. This effect has been attributed to its anti estrogenic effect which may be responsible for arrest of spermatogenesis in male and inhibitory effect on implantation of ovum in female. This constituent may prove to be a promising anti fertility agent devoid of side effect.

4.18-Anti diabetic effect

Ocimum shows prominent effect in lowering blood sugar level.

4.19-Other uses

Many people wears the ocimum, which is said to have certain physical medicinal properties. Its wood is considered as more powerful than any other gem that in protecting one from the negative influence. One can also buy several handicraft jewellery items made of tulsi wood.

CONCLUSIONS

The present study clearly indicates that *Ocimumcanumis* a rich source of phyto-constituents having lotes of pharmaceutical application like treatment of bronchitis, bronchial asthma, malaria, diarrhea, dysentery, skin diseases, arthritis, painful eye diseases, chronic fever, insect bite etc. The *Ocimum canum L.* has also been suggested to possess antifertility, anticancer, antidiabetic, antifungal, antimicrobial, hepatoprotective, cardioprotective, antiemetic, antispasmodic, analgesic, adaptogenic and diaphoretic actions.

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