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Ethanopharmacological and Phytochemical Aspects of *Ocimum sanctum* Linn- The Elixir of Life

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Authors' contributions

Both the authors contributed equally in designing the concept, literature collection and manuscript preparations. All authors read and approved the final manuscript.

Research Article

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ABSTRACT

Ocimum sanctum Linn. (Tulsi), a sacred and traditional medicinal plant of India which belongs to the family Lamiaceae possesses innumerable health benefits and therefore regarded as the "Elixir of Life". The entire plant body including its leaves, stem, root, inflorescence and seed are proved to be significant medicinal value and hence it is one among the inevitable plant used in the preparation of various ayurvedic pharmacological products. The plant is a rich source of various components including eugenol, Vicenin- 2, linoleic acid, oleic acid, rosmarinic acid, Ocimarin, isorientin, orientin, isovitexin, aesculectin, aesculin, chlorgrnic acid, galuteolin, circineol, gallic acid, Citronellal, Camphene, Sabinene, Dimethyl benzene, Myrecene, Ethyl benzene, Limocene, Vitamin C, Calcium, Phosphorous and many more. The plant truly deserves the title 'Elixir of Life' due to its Ethanopharmacological properties such as Anti- diabetic, Anti- cancerous, Analgesic, Anti- inflammatory, Radiopreotective, Hepatoprotective, Anti- microbial, Immunomodulatory effect, cardioprotective, Anti- coagulant, Anti- fertility, Anti- oxidant, Neuroprotective and the line-up found to be multitudinous. This review elucidates in-depth literature survey particularly focussing the phytochemical constituents of Tulsi as well as extrapolating its Ethanopharmacological property.

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1. INTRODUCTION

Ayurveda, “the science of life” or longevity is more than 5000 years old and is believed to be the oldest healing science in existence. This is a system of traditional Hindu medicine which is native to India and is renowned as one of the major systems of alternative and complementary medicine. According to Hindu mythology, Dhanvantari, the physician of the God’s, is attributed with the origin of ayurvedic medicine. Ayurveda traces its origin to the Vedas particularly Atharvaveda and it stresses the use of indigenous plant based medicines for the treatment of diseases [1]. India with its vast geographical diversity inhabits about 17,000 species of higher plants and among that 7500 are known for its medicinal properties. These plants were prominent in the regions of Eastern Himalayas, Western Ghats and Andaman and Nicobar Island [2]. Medicinal plants are considered to be very rich sources of secondary metabolites which are of therapeutic importance. They are the most exclusive source of life-saving drugs for majority of the world's population. The important advantages of medicinal plants in various treatments are: their safety besides being less expensive, efficacy and availability throughout the world. According to World Health Organisation, the practitioners of traditional system of medicine treat about 80% of patients in India, 85% in Burma and 90% in Bangladesh [3]. *Ocimum sanctum* Linn, a traditional medicinal plant widely known across South Asia is commonly used in Ayurvedic medicines [4]. The entire plant body is used for the treatment of various human diseases. The plant possess diverse functions as anti- diabetic, anti- fungal, anti- microbial, anti- fertility, anti- cancer, cardioprotective, expectorant, anti- spasmodic, adaptogenic, anti- helminthic, antiseptic, analgesic, tonic rejuvenator and so on. The beneficial effects of *O. sanctum* are shown in Table 1. This review details the morphology, distribution and systemic classification of *O sanctum*, its health beneficial’s, phytoconstituents of Tulsi plant and a detailed up- to- date literature survey of various research findings related to its Ethanopharmacological property.

Table 1. Medicinal properties of *O. sanctum* L.

Diseases	Plant parts used	Medicinal Properties
Fever and Cold [5]	Leaf, Juice of leaf, Decoction of leaves	Leaves of Tulsi are specific against many fevers. In case of acute fevers, a decoction of the leaves boiled with powdered cardamom in half a litre of water and mixed with sugar and milk brings down the temperature
Malaria and Dengue [6]	tender leaves	Tender leaves boiled with tea acts as a preventive against these diseases.
Cough [7]	Leaf	Tulsi is an important constituent of many ayurvedic cough syrups and expectorants.

Bronchitis [8]	Leaf	Helps to mobilise mucous
Asthma [9]	Leaf	Helps to mobilise mucous
Influenza [10]	Leaf	Chewing Tulsi leaves relieves cold and flu.
Sore throat [11]	Leaf	Water boiled with Tulsi leaves can be taken as drink, water can also be used as a gargle
Heart disorder [12]	Leaf	Reduces the level of blood cholesterol. It strengthens the heart muscles and improves blood supply to the heart.
Eye diseases [13]	dark basil juice	It is effective against night blindness and sore eyes which is caused due to deficiency of Vitamin A. Two drops of juice are dropped in to eyes daily before bed.
Stress [14]	Leaf	leaves act as “adaptogens” or anti- stress agent
Mouth infections [15]	Leaf	Tulsi leaves are effective against mouth ulcer and infections in the mouth. Regular chewing of leaves could cure these conditions
Insect Bite [16]	Paste of root and Juice of Leaf	A teaspoonful of juice of the leaves is taken and is repeated after a few hours. Fresh juice can also be applied to the affected parts. Paste of fresh root is also effective in case of bites of insects and leeches.
Kidney stone [17]	Leaf	Kidney stones can be removed if tulsi leaves are taken regularly. Taking tusli leaves along with honey and water for six months removes stones through the urinary tract. It also strengthens the kidneys
Headache[18]	Leaf	Pounded leaves mixed with sandal wood paste can be applied on the forehead for

		getting relief from pain, heat and headache by providing coolness.
Tooth disorder[11]	dried leaves	leaves are dried in the sun and powdered can be used for brushing teeth. It can counteract with bad breath and for healthy gums. It is also useful for pyorrhoea and other teeth disorders.
Skin disorder [12]	juice of Tulsi	Tulsi juice is beneficial in the treatment of ring worm and other skin diseases. It also shows effective for leucoderma.
Children's ailment [11]	juice of Tulsi leaves	common paediatric problems like cough, cold, fever, diarrhoea, vomiting could be effectively treated.
Improves memory[19]	Leaf	Daily intake of tulsi remarkably improves memory power and intelligence. The tulsi leaves acts as a nerve tonic that sharpens and boosts the memory power.

2. *Ocimum sanctum* Linn - MORPHOLOGY, DISTRIBUTION AND CLASSIFICATION

Botanically known as *Ocimum sanctum* Linn. and commonly as Tulsi, is the sacred plant of India and is also known by various names as Tulassi, Manjari, Krishna Tulsi, Trittavu, Tulshi, Thulsi. The plant is known in English as Holy Basil. *Ocimum sanctum* is perhaps the most common and most revered of all household plants in India. The plant is commonly cultivated in garden and also grown near temple. Medicinal properties attributed to the plant are not only mentioned in Ayurveda and Siddha but also in Greek, Roman and Unani system of medicine [20]. This aromatic shrub belongs to the family Lamiaceae. Tulsi has been described as vanya (wild type) and gramya (grown in homes). Properties of both types are almost similar and the main difference is the former has lighter leaves. Tulsi grows widely in tropics and warm regions. The plant is native to tropical Asia, likely having originated in India. Robust Tulsi varieties readily grow wild in many areas of Asia and Africa. It is also abundantly found in Malaysia, Australia and some of the Arab countries. The plant is distributed and cultivated throughout India.

Morphologically it is an erect, much branched subshrub which is of about 30-60 cm tall with hairy stems and simple opposite green leaves that are strongly scented. Leaves have petioles, and are ovate, up to 5 cm long, usually slightly toothed. Leaf color (Fig. 1) ranges from light green (Rama tulsi) to dark purple (Krishna tulsi). Flowers are purplish in elongate racemes in close whorls (Fig. 2). The morphology of *Ocimum sanctum* Linn is detailed in

Table 2. It is usually planted immediately after the rainy season ends. In good soil and hot sunny weather, Tulsi may grow to a meter or more in height and be ready for harvest in a few months. The plant is usually cultivated annually from seeds although it can also be propagated from tip or root cuttings.

Fig. 1. Leaves and Inflorescence of *Ocimum sanctum* Linn

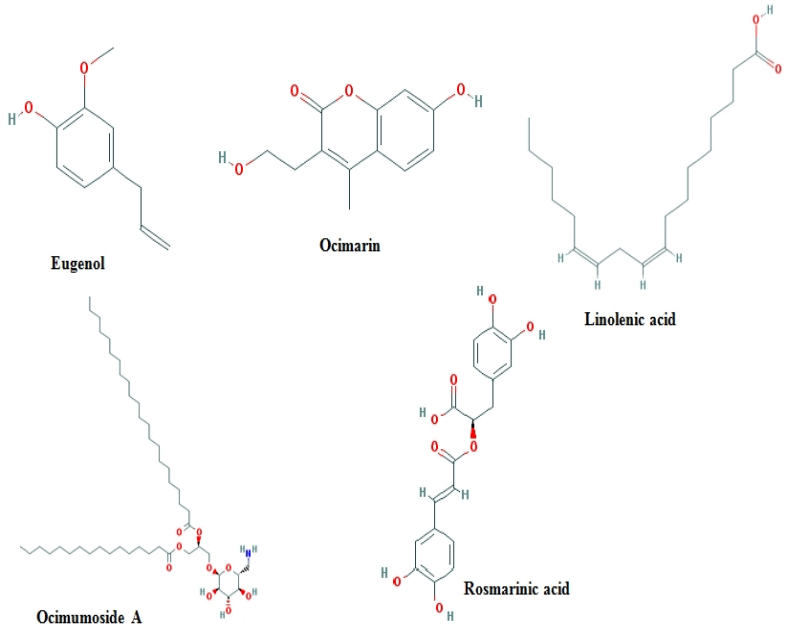

Fig. 2. Structure of few phytochemical constituents

Taxonomic Ranks

Kingdom :Plantae
Division : Magnoliophyta
Class : Magnoliopsida
Order : Lamiales
Family : Lamiaceae
Genus : *Ocimum*
Species :*sanctum*
Binomial name : *Ocimum sanctum* L.

Table 2. Morphology of *Ocimum sanctum* Linn.

Plant Parts	Description
Root	Thin, wiry, branched, hairy, soft, greenish brown externally and Pale blackish internally
Stem	Erect, herbaceous, woody, branched, hairy, quadrangular
Leaves	Cauline and ramal, opposite decussate, exstipulate, simple, petiolate, ovate, serrate, acute, pubescent, aromatic smell present, unicostate reticulate
Inflorescence	Vertecellaster
Flower	Crimson to purplish coloured, small in close whorls, Bracteate, pedicellate, complete, zygomorphic, hermaphrodite, pentamerous, hypogynous and cyclic
Calyx	sepal five, gamosepalous, calyx ¼ bilabiate, valvate, persistant
Corolla	petals five, gamopetalous, corolla 4/1 bilipped, valvate
Androecium	Stamens four, polyandrous, epipetalous, didynamous, dithecous, dorsifixed, introrse
Gynoecium	Bicarpellary, syncarpous, ovary superior, placentation axile, tetralocular with one ovule in each locule, a disc is present below the ovary, style gynobasic and stigma bifid [21]
Fruit	A group of 4 nut lets, each with one

	seed, enclosed in and enlarged, membranous, veined calyx, nut lets subglobose or broadly elliptic, slightly compressed, near smooth, pale brown or reddish with small black marking at the place of attachment to the thalamus, odour-aromatic, taste pungent, carcerulus
Seed	Rounded to oval brown mucilaginous when soaked in water; 0.1 cm long, slightly notched at the base, no odour, taste- pungent, slightly mucilaginous [22].

3. PHYTOCHEMICAL CONSTITUENTS OF TULSI: 'THE ELIXIR OF LIFE'

The unique aromatic odour of *O. sanctum* is due to the presence of essential or volatile oils. The aromatic volatile oil mainly constitutes phenols, terpenes and aldehydes. Various studies proved that chemical constituents vary due to edaphic and geographic factors [23]. Besides oil, the plant also contains alkaloids, glycosides, saponines and tannins. The volatile oils are mainly concentrated in the leaf. The leaf of *O. sanctum* contains 0.7% volatile oil comprising about 71% eugenol and 20% methyl eugenol [24, 25]. The essential oil from leaves comprise α- Thujene, Nonane, Octane, Benzene, (Z)-3- hexanol, Ethyl 2-methyl butyrate, α- pinene, β - pinene, Toulene, Citronellal, Camphene, Sabinene, Dimethyl benzene, Myrecene, Ethyl benzene, Limocene, 1,8- cineole, Cis- β - ocimene, Trans- β - ocimene, p-cymene, Terpiniolene, Allo- ocimene, Butyl- benzene, α- cubebene, γ- terpene, trans-linalool oxide, Geraniol, α- copaene, β - bourbonene, β - cubebene, linalool, eugenol, methyl eugenol, β - farnesene, β - elemene, (E)- cinnamyl acetate, Isocaryophyllene, β - caryophyllene, iso- eugenol, α- guaiene, α- amorphene, α- humulene, γ- humulene, 4, 11- selinadiene, α- terpenol, isoborneol, borneol, germacrene- D, α- salilene, β - salilene, myrtenylformat, α- muurolene, δ- cadinene, cuparene, calamenene, geraneol, neralidol, caryophyllene oxide, iedol, humulene oxide, α- guaiol, τ- cadinol, α- bisbolol, (EZ)- farnesol, cis- sesquisabinene hydrate, Elemol, tetradecanal, selin- 11- en- 4- α- ol, 14-hydroxy- α- humulene [26,27,28,29,30,31,32,33]. The alcoholic extract of leaf and other aerial parts constitute ursolic acid, asgenin, luteolin, apignin- 7- O- glucuronide, luteolin- 7- O- glucuronide, isorientin, orientin, molludistin, stigmsterol, triacontanol ferulate, vicenin-2, vitexin, isovitexin, aesculectin, aesculin, chlorgrnic acid, galuteolin, circineol, gallic acid, gallic acid methyl ester, gallic acid ethyl ester, procatechuic acid, vllinin acid, 4- hydroxybenzoic acid, vallinin, caffiec acid, chlorogenic acid, phenyl propane glucosides 1, phenyl propane glucosides 2, β- stigmsterol and rosmarinic acid [34, 35, 36, 37, 38, 39, 40]. The leaves contain ascorbic acid and carotene as well. The oil extracted from the seed of Tulsi is called fixed oil and mainly composed of fatty acids. The fixed oil from seed includes palmitric acid, stearic acid, linolenic acid, linoleic acid, oleic acid, sitosterol, dilinoleno-linolins, linolenodilinolin, hexourenicacid [41,42]. The mineral content (per 100 gram) of whole plant of *O. sanctum* L. includes Vitamin C (83 mg), Carotene (2.5 mg), Calcium- 3.15%, Phosphorous- 0.34%, Chromium- 2.9 µg, Copper- 0.4 µg, Zinc- 0.15 µg, Vanadium- 0.54 µg, Iron- 2.32 µg, Nickel- 0.73 µg and insoluble oxalate [43].

A group of organisms that produce the same chemical profile for a particular class of secondary metabolites is termed as chemotype. Variations in chemical profiles were

observed from oils produced from specimens from the same population and location, demonstrating the presence of different chemotypes within this species. The chemical composition of the *O. sanctum, O. gratissimum* and *O. basilicum* essential oils varied depending upon the origins and cultivars. Within the *O. basilicum*, there is a clear variation in phenotype and chemotype in terms of oil content and oil composition [44]. Linalool and methylchavicol were the main components of common and European basils, whereas methyl-chavicol was present at high concentration in the Reunion basil. The tropical chemotype of basil is known to have methyl cinnamate as the major component of its essential oil. Another basil chemotype, frequently grown in North Africa, Russia, Eastern Europe, and parts of Asia was high in eugenol [45]. On the basis of chemical composition, Telci et al. [46] classified the basil into seven chemotypes: (1) linalool, (2) methyl cinnamate/linalool, (3) methyl cinnamate, (4) methyl eugenol, (5) methyl chavicol (estragol), (6) citral, and (7) methyl chavicol/citral. *Ocimum sanctum* also varies considerably in terms of Methyl Eugenol (ME) and eugenol contents in leaf and inflorescence essential oils. Seven varieties of holy basil in Malaysia and Indonesia can be grouped into three chemotypes based on the phenylpropanoid content in leaf essential oils: two as eugenol chemotypes with 66–73% eugenol and 0.5–3.1 % ME, four ME chemotypes with 78–81% ME and 2.7–5.8 % eugenol, and one ME—eugenol chemotype with 52% ME and 27% eugenol [47]. The phenylpropanoids in the leaves of both sweet and holy basils are not released naturally. They are stored in the numerous oily glands (characteristic of Lamiaceae (formerly Labiatae)). More glands per unit surface area are found on the lower surfaces of leaves in the basils.

4. ETHANOPHARMACOLOGICAL PROPERTIES OF *Ocimum Sanctum* Linn.

4.1 Anti-Fatigue Activities

The aqueous suspension of 70% alcoholic extract of *Ocimum sanctum* L. was investigated for antifatigue activity in rats. Swimming time, change in body weight, lipid peroxidation, Lactic Acid (LA), glycogen and blood biochemical parameters namely heamoglobin (Hb%), Blood Urea Nitrogen (BUN) and Creatine Kinase (CK) were evaluated as biomarkers of physical fatigue. Leaf extract of *O. sanctum* at 300 mg kg^{-1} b. wt would be the optimum concentration to act against fatigue in rats [48]. Treatment at 300 mg kg^{-1} b.wt. facilitated the aerobic glucose metabolism and promote swimming time, suggesting that *O. sanctum* ameliorates the various impairments associated with physical fatigue. Furthermore validation is needed regarding the optimal concentration.

4.2 Adaptogenic Properties

An adaptogen is a herb product that is a plant derivative. *O. sanctum* is an important supplement used in combination with other plants for the treatment of various stress-induced disorders in India and other Asian countries [8]. An interesting study by [49] on *Ocimum sanctum* leaf extract reveals three new compounds namely ocimumoside A, ocimumoside B and ocimarin, together with eight known substances, apigenin, apigenin-7-*O*-β-d-glucopyranoside, apigenin-7-*O*-β-d-glucuronic acid, apigenin-7-*O*-β-d-glucuronic acid 6″-methyl ester, luteolin-7-*O*-β-d-glucuronic acid 6″-methyl ester, luteolin-7-*O*-β-d-glucopyranoside, luteolin-5-*O*-β-d-glucopyranoside, and 4-allyl-1-*O*-β-d-glucopyronosyl-2-hydroxybenzene and two known cerebrosides. The new compounds and the known compounds apigenin-7-*O*-β-d-glucuronic acid and 4-allyl-1-*O*-β-d-glucopyronosyl-2-hydroxybenzene were screened at a dose of 40 mg/kg body weight for acute stress-induced

biochemical changes in male Sprague–Dawley rats. Compound ocimumoside A displayed promising antistress effects by normalizing hyperglycemia, plasma corticosterone, plasma creatine kinase, and adrenal hypertrophy. Compounds ocimumoside B and 4-allyl-1-*O*-β-d-glucopyronosyl-2-hydroxybenzene were also effective in normalizing most of these stress parameters. In contrast, compounds ocimarin and apigenin-7-*O*-β-d-glucuronic acid were ineffective in normalizing any of these effects.

4.3 Anti- Microbial Properties

Several studies were conducted to prove the antimicrobial activity of *O. sanctum.* Antibacterial activity of Tulsi against *E. coli* and *S. aureus* at various plant concentrations ranging from 15%, 30%, 50% and 100% showed inhibition showed maximum activity against *S. aureus* followed by *E. coli* [50]. A study [51] on the crude extracts prepared from dry leaves of *Ocimum sanctum* with methanol, hexane, acqueous, ethanol and ethyl acetate shows that the most active extract was found to be of methanol which inhibited a total of four bacteria (*E. coli, S. aureus, B. subtilis* and *B. cereus*) studied in the range of 11.86 mm to 18.50 mm size of inhibition zone. Ethanol, ethyl acetate, and aqueous extracts were found to be effective only against *Staphylococcus aureus* and *Escherichia coli* but, with comparatively lower activity than that of methanol extract. Hexane extract was found completely inactive against all the organisms tested. *Staphylococcus aureus,* a Gram-positive bacterium was observed as most susceptible bacterium as it was inhibited by almost all the extracts except hexane extract. Another interesting study on extract with chloroform from leaves were found to be most effective against *P. aeruginosa* [52]. A study suggested that higher content of linoleic acid in *O. sanctum* L. fixed oil could contribute towards its antibacterial activity. The oil show good antibacterial activity against *Staphylococcus aureus, Bacillus pumius* and *Pseudomonas aeruginosa*, where *S. aureus* was the most sensitive organism [53]. The aqueous extract of *O. sanctum* L. (60 mg/kg) show wide zones of inhibition compared to alcoholic extract against *Klebsiella, E. coli, Proteus, S. aureus* and *Candida albicans* when studied by agar diffusion method. Alcoholic extract showed wider zone for *Vibrio cholera* [54].

4.4 Anti-Convulsant Activity

Different extractives of stem, leaf and stem callus of *O. sanctum* were tested for anticonvulsant activity against standard drug phenytoin using maximal electroshock (MES) model. Ethanol and chloroform extractives of stem, leaf and stem calli were effective in preventing tonic convulsions induced by transcorneal electroshock [55]. Ethanolic extract of leaves of *O. sanctum* L. prolonged the time of lost reflex in mice due to pentobarbital, decreased the recovery time and severity of electroshock and pentylenetetrazole-induced convulsions and decreased apomorphine-induced fighting time and ambulation in 'open field' studies [56].

4.5 Anti- Fertility Activity

Treatment of albino rats with a benzene extract of *Ocimum sanctum* leaves (250 mg/kg body weight) for 48 days decreased total sperm count, sperm motility, and forward velocity. The results suggest that such effects are due to androgen deprivation, caused by the anti-androgenic property of *Ocimum* leaves. The effect was reversible because all parameters returned to normal 2 week after the withdrawal of treatment [2]. A significant decrease was noted in the sperm count in rabbits. Serum testosterone levels showed marked increase

while FSH and LH levels were significantly reduced in *Ocimum* -treated rabbits (2 g fresh leaves/rabbit for 30 days). Hence tulsi may prove to be a promising antifertility agent devoid of side effects.

4.6 Anti-Diabetic Properties

The antidiabetic effects [57] of Ethyl acetate (Et-Ac), Petroleum-ether (Pet-ether), and Chloroform fractions from ethanolic extract of the leaves of *Ocimum sanctum* were investigated in normal and alloxan induced diabetic rats (AIDRs). The effect of these fractions on fasting blood glucose (FBG), total cholesterol (TC), triglyceride (TG), serum glutamate oxaloacetate transaminases (SGOT), serum glutamate pyruvate transaminases (SGPT) level, and liver glycogen content were investigated in AIDRs and found significant effects. Administration of these fractions to the AIDRs resulted in the significant elevation of liver glycogen content. In diabetic rats, SGOT and SGPT levels were significantly elevated that were further reduced after intraperitoneal administration of these fractions. These results indicate that different fractions of *O. sanctum* have favourable effects in bringing down the severity of diabetes together with hepatoprotectivity.

4.7 Radioprotective Properties

In a study [58], the radioprotective effect of *Ocimum sanctum* on the salivary gland of rats administered radioiodine (131) I and compared its efficacy with a known radioprotectant, amifostine were conducted. Before exposing the rats to radiation it was orally administered with *O. sanctum* (40 mg/kg for 5 days) and amifostine (200 mg/kg, s.c). *O. sanctum* and amifostine pre- supplemented and subsequently exposed to (131)I rats at 3 and 6 months duration exhibited comparable histopathology with controls. Our study indicates possible radioprotective effect of *O. sanctum* and amifostine against high-dose (131) I exposure. In another study [59], the radiopreotective effect of two flavonoids: orientin and Vicenin obtained from the leaf of *O. sanctum* and synthetic compounds WR- 2721 and 2-mercaptopropionyl glycine (MPG) have been compared by examining chromosome aberration in cells of bone marrow in irradiated mice. Vicenin produced the maximum reduction in percentage aberrant cells while MPG was the least effective. However WR-2721 was the most effective against reduction of complex aberrations followed by Vicenin. Considering the low dose needed for protection and the high margin between the effective and toxic doses, the *Ocimum* flavonoids may be promising for human radiation protection.

4.8 Anti- Inflammatory Properties

Inflammation is a protective attempt by the organism to remove the injurious stimuli and to initiate the healing process. It was found that the pale yellow colored fixed oil and linolenic acid found to possess significant anti- inflammatory activity against PGE2, leukotriene and arachidonic acid-induced paw edema [60]. The anti- inflammatory activity of fixed oil is due to dual inhibition of arachidonate metabolism supplemented by antihistaminic activity. The antiinflammatory activity is not dependent on the pituitary adrenal axis [61]. Moreover it is also shown that linolenic acid percent in the fixed oils of different species of *Ocimum (O. basilicum* and *O. americanum*) has the capacity to block both the cyclooxygenase and lipoxygenase pathways of arachidonate metabolism and could be responsible for the antiinflammatory activity. A methanol extract and an aqueous suspension of *Ocimum*

sanctum inhibited acute as well as chronic inflammation in rats as tested by carrageenan-induced pedal edema and croton oil-induced granuloma and exudate, respectively. In both test procedures, the anti-inflammatory response of 500 mg/kg of methanol extract and aqueous suspension was comparable to the response observed with 300 mg/kg of sodium salicylate [62]. Compounds isolated from *O. sanctum* L. extract, Civsilineol, Civsimavatine , Isothymonin, Apigenin, Rosavinic acid and Eugenol when observed for their anti-inflammatory activity [63] or cyclooxygenase inhibitory activity shows the following results. Eugenol demonstrated 97% cyclooxygenase-1 inhibitory activity when assayed at 1000 µM concentration (pn). Civsilineol, Civsimavitin, Isothymonin, Apigenin and Rosavinic acid displayed 37%, 50%, 37%, 65% and 58% cyclooxygenase-1 inhibitory activity, respectively, when assayed at 1000 µM concentrations.

4.9 Cardioprotective Properties

Effect of pre- and co-treatment of hydroalcoholic extract of *Ocimum sanctum* at different doses was investigated against isoproterenol induced myocardial infarction in rats. Myocardial infarction (MI) was produced in rats with 85, 200 and 300 mg/kg of isoproterenol administered subcutaneously twice at an interval of 24 h. Shift in antioxidant parameters, lactate dehydrogenase (LDH) together with morphological and histopathological changes were investigated. The study [64] concluded by further confirmation by histopathological findings that *O. sanctum* at the dose of 50 mg/kg was found to demonstrate maximum cardioprotective effect and hence *Ocimum* may be of therapeutic and prophylactic value in the treatment of Myocardial infarction. In a study [65] investigating the cardioprotective activity of a combined treatment of Ginkgo biloba phytosomes (GBP) and *Ocimum sanctum* extract in isoproterenol (ISO)- induced myocardial necrosis in rats shows the combined treatment demonstrates significant cardiac protection.

4.10 Immunomodulatory Effect

Tulsi is considered as a sacred herb and traditionally it is believed that consumption of Tulsi leaf on empty stomach increases immunity. Experimental studies have shown that alcoholic extract of Tulsi modulates immunity. Three hundred milligrams capsules of ethanolic extracts of leaves of Tulsi or placebo were administered to 24 healthy volunteers on empty stomach and the results of 22 subjects who completed the study were analysed [66]. The immunological parameters such as the levels of Th1 and Th2 cytokines (interferon-γ and interleukin-4) during both pre and post intervention period in blood culture supernatants following stimulation with lipopolysaccharide and phytohaemagglutinin, T-helper and T-cytotoxic cells, B-cells and NK-cells also were analysed using Flow cytometry. Statistically significant result from the study proves the immunomodulatory role of Tulsi in healthy volunteers. Feeding of *Ocimum sanctum* to wistar albino rats enhanced both types of immune responses: antibody titre against *Salmonella Typhimurium* '0' antigen and cell mediated immune response using DNCB as an antigen [67]. Daily uptake of 250mg/kg for 20 days by rats did not show any sign of toxicity. Spleen cells harvested from the treated rats in the presence of ConA showed increased proliferation. Wistar albino rats' spleen cells treated *in vitro* with different concentrations (25–500µg/ml) of *Ocimum sanctum* in presence of ConA also exhibited increase in their proliferation.

4.11 Hepatoprotective Properties

Hepatoprotective activity of *Ocimum sanctum* leaves extract was evaluated in experimentally induced chronic lead toxicity in male wistar rats. The TSP, antioxidant enzymes and histopathological examination, immunohistochemical staining and ultrastructural examination of liver were estimated. Lead residue in liver was also measured. The *Ocimum sanctum* significantly increased the levels of total serum proteins, CAT, SOD and GPx in liver and lead residues were significantly reduced in *Ocimum* treated groups. Tulsi significantly minimized the gross and histopathological changes and also reduces the apoptosis in hepatocytes. By the end of experiment in *Ocimum* treated animals the liver almost coming to its normal appearance. This experiment suggests that the *Ocimum santum* exhibited significant hepatoprotective effect on lead induced hepatic damage in rats [68]. In another study [69], Wistar strains of Albino rats were induced using lead for hepatotoxicity and the aqueous extract of *O. sanctum were* administered to the animals orally for a period of 21 days. All the parameters considered for the study were restored to near normal when treated with the aqueous extract of *O. sanctum* with a statistically significant value, $p<0.05$ depicting the hepatoprotective nature.

4.12 Anti-Carcinogenic Properties

In a study [70] conducted to determine the efficacy of novel flavonoid vicenin-2 (VCN-2), an active constituent of the medicinal herb *Ocimum Sanctum* Linn. in combination with docetaxel (DTL) in carcinoma of prostate shows VCN-2 effectively induced anti-proliferative, anti-angiogenic and pro-apoptotic effect in CaP cells (PC-3, DU-145 and LNCaP) irrespective of their androgen responsiveness or p53 status. It was observed in a study [71] that the treatment of a squamous cervical cancer cell line, SiHa with the ethanolic extracts of leaves of *Ocimum sanctum* at IC50 values for 48 h resulted in formation of internucleosomal fragments of DNA. Because of its anti- inflammatory, anti- proliferative and anti- angiogenic agent O sanctum proves to be effective in cancer treatment.

4.13 Mosquito Repellent Property

Essential oil of Tulsi has been reported to be possessing 100% larvicidal activity against Culex mosquitoes. Its extracts have marked insecticidal activity against mosquitoes. It is repellant action lasts for about two hours. The *O sanctum* leaf extract was tested [72] at various concentrations ranging from 150- 900ml volume against three species: Anopheles, Culex and Ades adult mosquitoes (3-5 days old) in small net, large net and large room conditions The results suggested that at high concentration of *O. sanctum* leaf extract show greater repellent activity in all net containing mosquitoes. However, low concentration of extract show greater activity in small net but poor in large net. From the above it can be concluded that high concentration of *O. sanctum* leaf extract can be used for preparation of mosquito's repellent formulation without side effects.

4.14 Analgesic Activity

The analgesic activity of fixed oil from the seeds of *Ocimum sanctum* was studied after intraperitoneal injection in mice and rats using the tail flick, tail clip, tail immersion and acetic acid-induced writhing methods; results were compared with morphine and aspirin. It was found that O. sanctum failed to raise the pain threshold which indicated that analgesic activity is not centrally mediated. Using the acetic acid-induced writhing method, the oil

showed significant inhibition in a dose-dependent manner suggesting its possible mechanism related to the peripheral system [73].

4.15 Other Properties

O. sanctum is a rich source of secondary metabolites and has an outstanding role in medicine. These metabolites are not essential for its survival but they are of considerable importance to human. Secondary metabolites carry out a number of protective functions in the human body. Plant secondary metabolites can boost the immune system; protect the body from free radicals, kills pathogenic germs and much more. It was shown that the transdermal drug delivery using a combination of Tulsi oil and terpentine oil, demonstrated significantly higher drug delivery than the synthetic combination such as isopropylene and propylene glycol when subjected to abdominal skin of rat [74]. To improve shelf life of a soybean product called 'Tofu', aqueous extract of Tulsi was added to it. The shelf life of 'Tofu' increased from normal 3-4 days to 7-8 days [75].Since Ocimum is a potential source of various phytochemicals, it possess numerous other Ethanopharmacological properties including anti- toxic activity, Ethanoveterinary activity and a brief description of other pharmacological property is listed in Table 3.

Table 3. Other Ethanopharmacological property of *O. sanctum*

Ethanopharmacological Properties	Plant Parts and Type of Extract
Anti- ulcer Activity	Fixed oil from seeds [76]
Anti- arthritic Activity	Fixed oil from seeds [61]
Anti- asthmatic Activity	Hydro- alcoholic Extract from leaves [77]
Anti- cataleptic Activity	Alcoholic Extract from leaves [78]
Anti- cataract Activity	Aqueous Extract from leaves [79]
Anti- coagulant Activity	Fixed oil/ methanol extract from leaf [80]
Anti- emetic Activity	Leaf extract [81]
Anti- helminthic Activity	Essential oil from leaves [82]
Anti- hyperlipidemic Activity	Fixed/ essential oil from seeds/leaves [61]
Anti- oxidant Activity	Alcoholic extract from whole plant [83]
Anti- plasmodial Activity	Alcoholic extract from leaves [84]
Anti- pyretic Activity	Fixed oil from seeds [62]

Anti-spasmodic Activity	Leaf Infusion [85]
Anti- stress Activity	Alcoholic extract from whole plant [86]
Anti- thyroidic Activity	Leaf extract [87]
Anti- tussive Activity	Aqueous / Alcholic extract from aerial parts [88]
Anti- anxiety Activity	Alcoholic extract from leaves [89]
Anti- depressant Activity	Alcoholic extract from leaves [89]
Chemopreventive Activity	Fixed oil from seeds [90]
Genoprotective Activity	Hydroalcholic extract from leaves [91]
Larvicidal Property	Chloroform and methanol extract leaf [92]

5. CONCLUSION

Medicinal plants are rich source of secondary metabolites which has powerful physiological effects in humans and are used as medicines. The significance of *Ocimum sanctum* and its extracts as source of medicines dates back to centuries and hence it is mentioned in age old art of medicine the “Ayurveda”. It is remarkably evident that the Tulsi leaves and its juice effectively reduces many diseases including the digestive disorders, respiratory disorders, kidney related problems, Cardiovascular disorders, Cancer, mosquito repellent and all. Proper conservation and sustainable use of such plant resources may enhance the longevity of human life as well as contribute considerably against the drug resistant microorganisms. In the developing countries, increased cost of medication and their side effects are of great concern to general public hence opening new channels of pharmacological investigations focusing on natural medication and diverting human trends toward natural cure. *Ocimum sanctum* is present in almost every parts of the Indian subcontinent and its immunomodulatory properties may be explored to provide additional immunity to mankind at very low cost due to its easy availability. Future studies in detail in Tulsi regarding neuro-protective and regenerative properties will open new vista to understand its role in Parkinson’s and Alzheimer’s diseases. Further studies on holy basil should focuss more on unknown aroma impact compounds. More advanced studies should be conducted in exploring the biopesticidal effect of *O sanctum* as well as developing new tulsi based drugs. The various phytochemicals found in this plant act as a vitalizer if intake daily and hence Tulsi is obviously the “Elixir of Life”.

CONSENT

Not applicable.

ETHICAL APPROVAL

Not applicable.

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COMPETING INTEREST

Authors have declared that no competing interests exist.

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