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# A review on phytochemical and pharmacological properties of Holy basil (*Ocimum sanctum* L.)

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ABSTRACT

*Ocimum sanctum* Linn. commonly known as *Holy Basil* or *Tulsi* is an Ayurvedic herb of Southeast Asia with a long history of traditional use. The culinary, medicinal and industrial importance of this plant led to explore its chemical and pharmacological properties. Here, we provide a comprehensive review on scientific findings of *O. sanctum* chemical constituents and their related anticancer, antioxidant, anti-inflammatory, antistress, γ-irradiation protection, antidiabetic and antileishmanicidal activities. More than 60 chemical compounds have been reported from *O. sanctum*, including phenolics, flavonoids, phenyl propanoids, terpenoids, fatty acid derivatives, essential oil, fixed oil, and steroids. The pharmacological activities of *O. sanctum* compounds reflect their medicinal importance and in the standardization of medicinal products. This compilation will be helpful in the development of new active principle and nutraceuticals in the area of drug resistance and emerging chronic disease vectors.

## 1. Introduction

The genus *Ocimum* belongs to the family Lamiaceae, comprises about 68 species indigenous to tropical regions of Asia, Africa and, central and south America (ThePlantList, 2013). *Ocimum sanctum* Linn. (*Os*) synonym *Ocimum tenuiflorum* L. (Lamiaceae), the most prominent species of the genera is cultivated worldwide for its medicinal, perfumery, religious, ceremonial, food and essential oil importance (Nadkarni, 1976). *Os* is a short-lived perennial shrub of 30–60 cm height with hairy stems and sparsely hairy leaves, which distributed in the Himalayas up to an altitude of 6000 feet (Watt, 1972). This aromatic shrub is commonly known as *Holy Basil* or *Tulsi* and identified as two common cultivars, *Rama Tulsi* with green leaves and *Krishna Tulsi* with purple leaves (Vani et al., 2009; Darrah, 1974). *Os* have been reported for antidiabetic, wound healing, antioxidant, radiation protective, immunomodulatory, antifertility, anti-inflammatory, antimicrobial, antistress and anticancer activities (Gholap and Kar, 2004; Vats et al., 2004; Udupa et al., 2006; Trevisan et al., 2006; Gupta et al., 2006; Geetha and Vasudevan, 2004; Yanpallewar et al., 2004; Bhartiya et al., 2006; Subramanian et al., 2005; Mukherjee et al., 2005; Godhwani et al., 1988; Ahmed et al., 2002; Kelm et al., 2000; Karthikeyan et al., 1999; Prashar et al., 1998; Prakash and Gupta, 2000; Singh et al., 2005). The toxicity studies suggest that *Os* is a nontoxic herb and safe to human use (Gautum and Goel, 2014; Sadashiv, 2010). The essential oil is one of the chemosystematic features of *Os* and a good natural source of eugenol. *Os* essential oil has commercial importance in various industries including pharmaceutical, cosmetics and food as an antiallergic and antimicrobial agent (Kumar et al., 2010).

The present review on *Os* aims to provide a comprehensive study on its traditional uses, chemical constituents, nutritional values and pharmacological activities of *Os* secondary metabolites. So far, no such systematic study has been carried out on the commercially and medicinally important herb *Os*. Hence, this study on phytochemical constituents and their reported pharmacological activities will serve as a chemical database for future research as well as enable to understand the research gap and outlook for future prospects.

## 2. Traditional uses and ayurvedic recommendations

*Os*, known as *Tulsi* (the incomparable one, Hindi) has been described as *Rasayana* drug in the ancient texts of Ayurveda including Charak Samhita, Susrut Samhita and Rigveda (3500–1600 BCE) to treat cough, respiratory disorders, poisoning, impotence and arthritis (Bano et al., 2017). It is considered as one of the sacred plants in India. *Os* is used as a nervine tonic, adaptogen, improving health during cancer and has beneficial effects in stress release (Chulet and Pradhan, 2009; Balachandran and Govindarajan, 2005). The therapeutic potential of *Os* has been well documented in Ayurveda and Siddha for healing properties as well as in Greek, Roman and Unani system of medicines for the treatment of skin diseases, common cold, headaches, coughs, malarial

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**Table 1**
Traditional uses of *O. sanctum*.

| Plant parts (preparation used) | Ethnomedicinal uses | Region/Country | References |
|---|---|---|---|
| Fresh leaf with water | Enhancing mental power | Himachal Pradesh (India) | Vidyarthi et al. (2013) |
| Leaves with *Bruguiera gymnorrhiza* and coconut oil (pounded and rubbed on body) | Renovating from tiredness | Nicobar Island (India) | Dagar (1989) |
| Leaves pounded with onion bulbs (juice taken orally) | Cough, cold and headache | Tamil Nadu (India) | Muthu et al. (2006) |
| Leaves | Cough, cold, leg swelling and fever | Bangladesh | Chowdhury and Koike (2010) |
| Leaves (juice) | Cough, cold, bronchitis and gastric disorders | Bangladesh | Sharkar et al. (2013) |
| Whole plant | Cough, cold, headache, nausea, fever and skin diseases | Chuadanga, (Bangladesh) | Rahman et al. (2013) |
| Leaves pounded with garlic, leaves of *Achyranthes aspera* and pepper | Typhoid fever | Andhra Pradesh, India | Reddy et al. (1988) |
| Leaves pounded with fruits of *Tricosanthes diocia*, flowers of *Leucas indica* and leaves of *Aristolochia bracteata* | Typhoid fever | Andhra Pradesh (India) | Reddy et al. (1989) |
| Leaf decoction with flower heads of *Leucas cephalotes* | Fever | Makawanpur (Nepal) | Bhattarai (1991) |
| Leaf decoction with *Piper nigrum* and palmgur | Fever | India | Nazar et al. (2008) |
| Leaves paste with black pepper | Diarrhea and fever | Central Himalaya (India) | Kandari et al. (2012) |
| Leaves (juice) | Diarrhoea and dysentery | Tripura (India) | Sen et al. (2011) |
| Dried leaves with ghee | Dysentery, colic and piles | Central Himalaya (India) | Kandari et al. (2012) |
| Leaves (paste and decoction) | Stomach disorder, inflammations and wound cuts | Arunachal Pradesh (India) | Namsa et al. (2011) |
| Leaves (crushed and filtered extract) | Stomach ache and head ache | Assam (India) | Sajem and Gosai (2006) |
| Flowers juice with honey, ginger and onion juice | Bronchitis | India | Watt (1972) |
| Leaves (juice) | Bronchitis and catarrh | India | Watt (1972); Sharkar et al. (2013) |
| Dried leaves (vegetable) | Blood purification | Central Himalaya (India) | Kandari et al. (2012) |
| Leaves (juice) | To treat ringworm | Uttar Pradesh (India) | Siddiqui et al. (1989) |
| Leaves crushed in goat's urine and mixed with coconut oil | Skin allergy | Karnataka (India) | Shivanna and Rajakumar (2011) |
| Plant (paste) | Skin infection | Tripura (India) | Sen et al. (2011) |
| Leaves pounded with *Catharanthus roseus* leaves and mild heated | Ear boils | Karnataka (India) | Shivanna and Rajakumar (2011) |
| Leaves powder with honey | Diabetes | Assam (India) | Chakravarty and Kalita (2012) |
| Leave, flower top and roots (juice) | Antidote in snake poisoning | India | Watt (1972) |
| Leaves paste | Antidote for scorpion bite | Andhra Pradesh (India) | Reddy et al. (1988) |

fever, diarrhoea, constipation and as an antidote for snake bite (Mondal et al., 2009; Uma Devi, 2001; Javanmardi et al., 2002). *Os* leaves have expectorant, carminative, refrigerant, febrifuge, laxative properties and their infusion is used as a stomachic in gastric disorders of children (Watt, 1972). Juice of fresh *Os* leaves is used as the first-aid remedy for earache. *Os* seeds are mucilaginous and demulcent and useful in the treatment of genitor-urinary disorders (Watt, 1972).

*Os* can be consumed as herbal tea, decoction (leaves and roots) to treat cough, cold and malarial fever (Prakash and Gupta, 2005). The paste of green or dried powdered leaves is used to treat ring-worm, skin diseases and vitalizing effect, whereas essential oil as larvicidal (Nadkarni, 1976). In Ayurvedic Pharmacopeia of India (1999), *Os* is recommended to treat pratishyaya (common cold), hikka (respiratory disorders), kasa (cough), aruci (loss of taste), kustha (skin disorders), krimiroga (treatment of worms) and parsva sula (chest pain) with a therapeutic dose of 2–3 g leaf powder. The available literature on the traditional uses of *Os* is limited to Asian countries, which compiled in Table 1.

## 3. Nutraceutical value (minerals, pigments and mucilage)

Minerals in routine intake of diet play an important role in food and nutraceutical industry. Herb *Os* has been used to add distinctive flavor in food and as a home remedy in various health conditions. The recent growing interest on the nutraceutical values of *Os* revealed that it is a rich source of vitamins, minerals, fat, protein, polysaccharide, fiber, pigments and mucilage (Pattanayak et al., 2010; Koche et al., 2011; Vidhani et al., 2016; Gowrishankar et al., 2010; Pachkore and Dhale, 2012). The macro and micro contents of *Os* are compiled in Table 2. The elemental analysis on macro and micro contents of *Os* leaves using Laser Induced Breakdown Spectroscopy (LIBS) and Inductively Coupled Argon Plasma Atomic Spectroscopy (ICAP-AES) techniques revealed the presence of almost all nutritionally important elements and interestingly high concentration of potassium (10521.477 ± 391.7 mg/kg leaves) (Tripathi et al., 2015). Presence of high concentration of potassium and lighter elements like C, H, O and N suggest the application of *Os* in maintaining electrolytic balance and source of organic compounds, respectively.

*Os* contains vitamin A, vitamin C, β-carotene, chlorophyll, insoluble oxalates, protein (30 Kcal), fat (0.5 g), carbohydrate (2.3 g), minerals and other phytonutrients. Each 100 g of leaf contain vitamin C (83 μg), carotene (2.5 μg), Ca (3.15%), P (0.34%), Cr (2.9 μg), Cu (0.4 μg), Zn (0.15 μg), V (0.54 μg), Fe (2.32 μg) and Ni (0.73 μg) (Pattanayak et al., 2010). Bhattacharya et al. (2014) analyzed the antioxidant contents in *Os* leaves and found the total carotenoid content (19.77 ± 0.01 g/100 g), total phenolic content (2.09 ± 0.10 g/100 g) and total flavonoid content (1.87 ± 0.02 g/100 g) of dry weights. The presence of ascorbic acid (8.21 mg/100 g), riboflavin (0.06 mg/100 g) and thiamine (0.3 mg/100 g) contents further suggest that *Os* leaves can intake as a dietary supplement, an alternative economic source of vitamins and natural antioxidant.

Basil seed gum or mucilage is composed of two major components (i) an acid stable core gluco mannan and (ii) α-linked xylan including acidic side chains at C-2 and C-3 of xylosyl residues in acid-soluble portion (Naji-Tabasi and Razavi, 2017). The seed mucilage of *Os* (yield ~30%), is a natural polymer that contains hexouronic acid (27.25%), pentoses (38.9%) and ash (0.2%) (Khare, 2016). *Os* seed mucilage has shown protein and amino acids on phytochemical evaluation, and possess swelling index 20 ml (water) with low ash value (Kadam et al., 2012). These physicochemical properties of mucilage direct towards its pharmaceutical excipient potential.

## 4. Chemical constituents of *Os*

*Os* leaves are rich in volatile oil (0.7%), phenolics, flavonoids, neolignans, terpenoids and fatty acid derivatives. *Os* seeds contain fixed

**Table 2**
Nutritional contents of *O. sanctum*.

| Plant part | Nutritional composition (% of dry weight) | | | | | | Mineral contents (ppm) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Protein | Lipid | Carbohydrate | Fiber | Moisture | Ash | Ca | P | Zn |
| Leaves | – | – | – | – | – | 10.13 | – | – | 0.15 |
| | 12.30 | 3.00 | 77.70 | 7.00 | 83.55 | 18.35 | – | – | – |
| | 4.93 ± 0.03 | 3.12 ± 0.28 | 27.23 ± 1.92 | 16.81 ± 1.25 | 31.35 ± 1.04 | 14.21 ± 1.50 | 18900 | – | 36 |
| | 0.80 ± 0.001 | 0.90 ± 0.45 | 2.10 ± 2.80 | 9.80 ± 0.80 | 5.30 ± 0.3 | 2.50 ± 0.08 | 450 | – | 713 |
| | 20.84 | – | – | 10.0 | 11.44 | – | 1.8%* | 1.0%* | 0.49 |
| | 20.64 ± 1.47 | 3.60 ± 0.08 | 39.58 ± 2.09 | – | – | – | – | – | 32.38 ± 1.42 |
| | 2.38% | 3.20 | 66.35 | 11.45 | 6.20 | 10.42% | 41204.1 | – | 800 |
| Stem | 9.25 | 2.75 | 68.05 | 18.30 | 88.30 | 20.15 | – | – | – |
| | 1.10 ± 0.001 | 1.10 ± 0.60 | 2.20 ± 2.30 | 12.20 ± 1.40 | 6.60 ± 0.30 | 2.60 ± 0.10 | 450 | – | 747 |
| Seed | – | – | – | – | – | – | – | – | – |
| Whole plant | – | – | – | – | – | – | 4031 ± 102 | – | – |
| | – | – | – | – | – | – | 35.9–19502 | – | 1.0–0.8 |

| Mineral contents (ppm) | | Water soluble vitamins (ppm) | | | | Calorefic value (kcal/100 g) | References |
|---|---|---|---|---|---|---|---|
| Fe | Cu | Vit B1 | Vit B12 | Niacin | Vit C | | |
| 2.32 | 0.4 | – | – | – | – | – | Narendhirakannan et al. (2005) |
| – | – | – | – | – | – | – | Koche et al. (2011) |
| 546 | 47850 | – | – | – | 24.1 | – | Kashif and Ullah (2013) |
| 354 | 31 | – | – | – | 310 | – | Shafqatullah et al. (2013) |
| 189 | 0.01 | – | – | – | – | 262.84 | Barua et al. (2015) |
| – | 14.48 ± 0.72 | – | – | – | 65.41 ± 0.76 | – | Vidhani et al. (2016) |
| 2830 | 400 | – | – | – | – | – | Wisdom et al. (2016) |
| – | – | – | – | – | – | – | Koche et al. (2011) |
| 366 | 30 | – | – | – | 450 | – | Shafqatullah et al. (2013) |
| – | – | 4.8 | 2.4 | 2.7 | – | – | Pachkore and Dhale (2012) |
| 372.0 ± 7.8 | 28.8 ± 4.0 | – | – | – | – | – | Gowrishankar et al. (2010) |
| 161.8–2.2 | – | – | – | – | – | – | Pachkore and Dhale (2012) |

High concentration of calcium and phosphorus is presented in%.

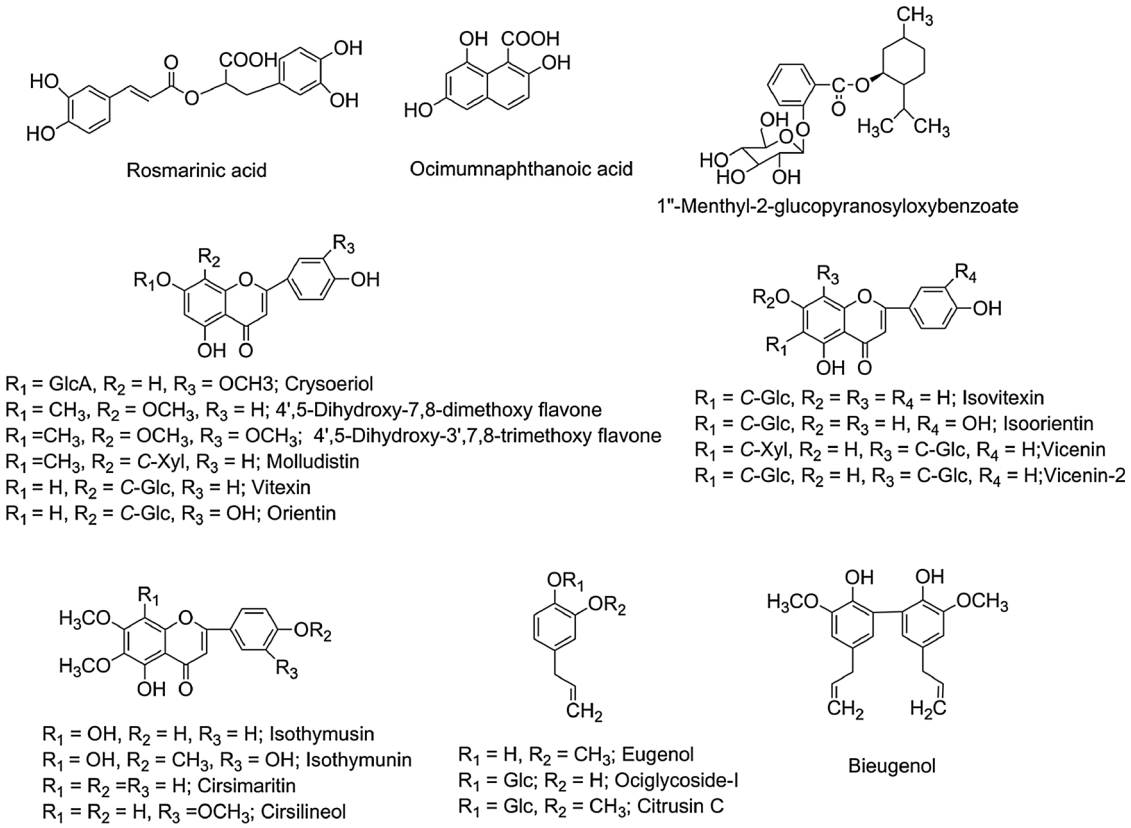


**Fig. 1.** Structure of major compounds from *O. sanctum*.

oil (18–22%), mucilage, polysaccharides and *β*-sitosterol in the unsaponifiable matter. *Os* seed oil is rich in triglycerides (94–98%) in which linolenic acid (43.8%) is the main content (Naji-Tabasi and Razavi, 2017). The structure of major secondary metabolites of *Os* is presented in Fig. 1. Table 3 summarizes the list of chemical constituents reported from *Os* and their related biological activities.

### 4.1. Phenolics

The total phenolic content in *Os* leaves has been found 4.07 ± 0.11 g gallic acid equivalent/100 g dry weight (Koroch et al., 2010). Caffeic acid, chlorogenic acid, vanillic acid, ocimumnaphthanoic acid and menthylsalicylic glucoside were isolated from the aerial parts of *Os* (Skaltsa et al., 1999; Ali and Ali, 2012; Ahmad et al., 2012a). The presence of commonly occurring phenolic compounds gallic acid, gallic acid methyl ester, gallic acid ethyl ester, protocatechuic acid, 4-hydroxybenzoic acid, vanillin and 4-hydroxybezaldehyde were confirmed by HPLC using authentic samples (Norr and Wagner, 1992). Rosmarinic acid, an ester of caffeic acid is quantified as 0.27% *w/w* in *Os* leaves using APCI mass spectrometry technique (Sundaram et al., 2012).

### 4.2. Flavonoids

Flavonoids are the major class including methoxy flavonoids and their glycosides (luteolin, isothymusin, cirsimartin), *C*-glycosides flavonoids (orientin, isoorientin, isovitexin and vicenin) from *Os* (Kelm et al., 2000, Norr and Wagner, 1992; Skaltsa et al., 1999; Uma Devi and Satyamitra, 2004) (Table 3). Grayer et al., 2001 studied the distribution of 8-oxygenated flavones on *Os* leaf surface using atmospheric pressure chemical ionization mass spectrometry (APCI-MS) and identified apigenin, cirsimaritin, salvigenin, crisilineol, eupatorin, isothymusin and gardenin. The analysis shows that flavone-7-*O*-glycosides are the characteristics of *Os*, whereas luteolin-5-*O*-glucoside considered as the marker compound in all nine species of *Ocimum*, including *Os* (Grayer et al., 2002). The flavones apigenin, isothymusin, cirsimaritin and crisilineol were isolated from the aerial parts of *Os* (Kelm et al., 2000; Suzuki et al., 2009).

### 4.3. Phenyl propanoids

Eugenol is one of the most distributed phenyl propanoid in the essential oil of *Os* leaves. Other phenyl propane derivatives such as ociglycoside or eugenyl-*β*-D-glucoside, citrusin C, ferulaldehyde, bieugenol and dehydrodieugenol were isolated from the leaves of *Os* (Kelm et al., 2000; Suzuki et al., 2009).

### 4.4. Neolignans

The methanol extract of *Os* leaves revealed seven novel neolignans named as Tulsinol A to Tulsinol G (Suzuki et al., 2009). These neolignans are formed by the polymerization of eugenol.

### 4.5. Coumarins

Three coumarins named ocimarin, aeculetin and aesculin were reported from *Os*. (Skaltsa et al., 1999; Gupta et al., 2007).

### 4.6. Terpenoids

Different terpenoids like sesquiterpenoids (β-caryophyllene and 4,5-epoxy-caryophyllene), abietane diterpenoid (carnosic acid), oleane triterpenoids (oleanolic acid, β-Amyrin-glucopyranoside) and ursane triterpenoids (ursolic acid, urs-12-en-3β,6β,20β-triol-28-oic acid) have been reported from *Os* (Suzuki et al., 2009; Baliga et al., 2013). The quantification studies revealed ursolic acid as the most abundant constituent in *Os* with 0.252%–0.478% *w/w* and 0.62–19.10 mg/g using

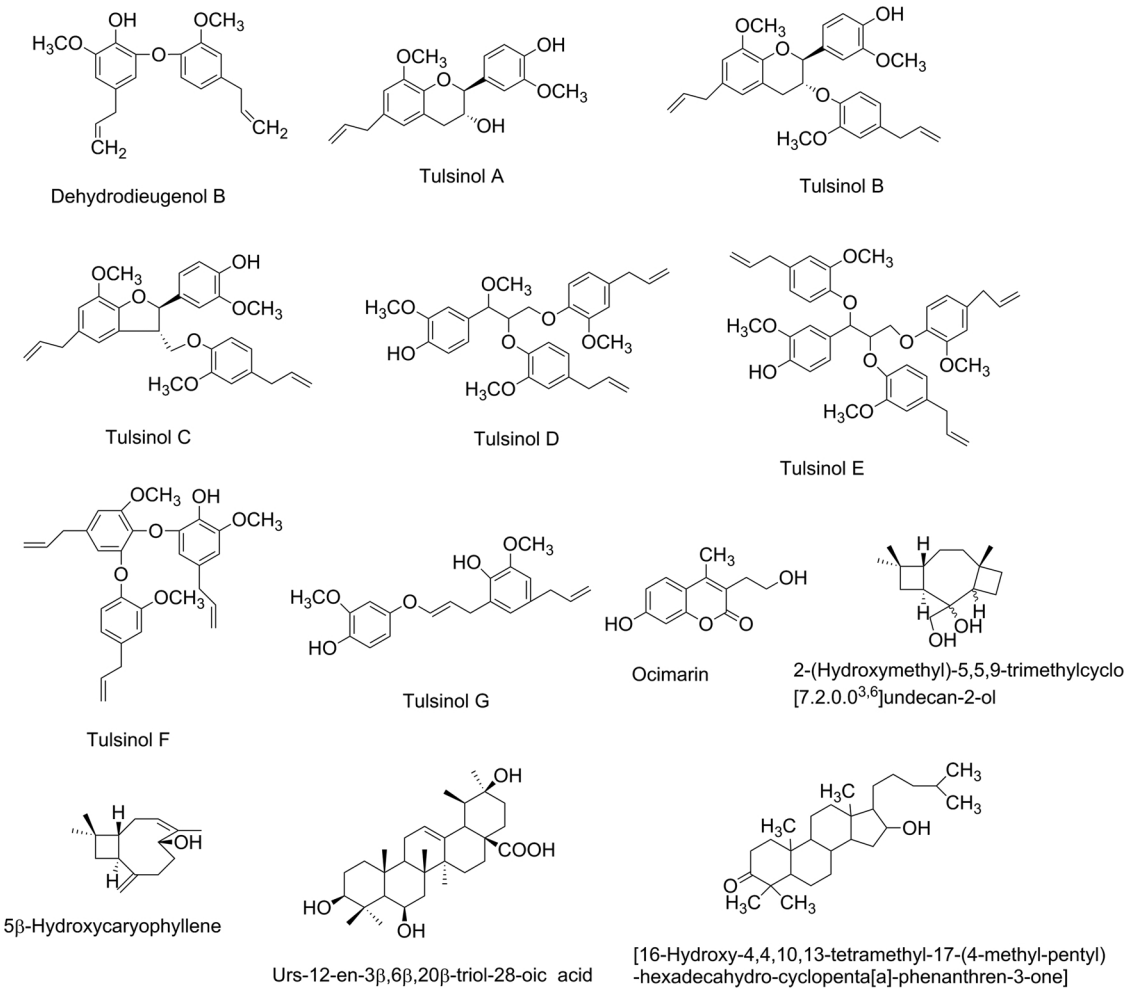


**Fig. 1.** (*continued*)

HPTLC and UPLC-ESI–MS/MS, respectively (Anandjiwala et al., 2006; Pandey et al., 2014). Two separate antidiabetic activity-guided isolation on *Os* roots and aerial parts provided two novel triterpenoids named urs-12-en-3*β*,6*β*,20*β*-triol-28oic acid and 16-hydroxy-4,4,10,13-tetramethyl-17-(4-methyl-pentyl)-hexadecahydrocyclopenta [α] phenanthren-3-one, respectively (Ahmad et al., 2012a; Patil et al., 2011). Further, a new tricyclic sesquiterpenoid 2-(hydroxymethyl)-5,5,9-trimethylcyclo[7.2.0.0$^{3,6}$]undecan-2-ol along with *β*-caryophyllene, elemene, α-humulene, α-caryophyllene, germacrene-A, trans-α-bergamotene and 5*β*-hydroxycaryophyllene were isolated from *Os* leaves (Singh et al., 2014). The novel tricyclic sesquiterpenoid was biosynthetically derived from *β*-caryophyllene (Singh et al., 2014).

### 4.7. Steroids

Four commonly occurring phytosterols *β*-sitosterol, *β*-sitosterol-3-*O*-*β*-D-glucopyranoside, stigmasterol and campesterol were isolated from leaves and stems of *Os* (Joshi and Karna, 2013; Suzuki et al., 2009; Baliga et al., 2013).

### 4.8. Essential oil

*Os* essential oil (yield 0.3-4.1%) is mainly composed of terpenoids including acyclic monoterpenoids, monocyclic terpenoids, bicyclic terpenoids, aliphatic aldehydes, phenolic acids, esters and sesquiterpenoids. The composition and yield of *Os* essential oil are differed with harvesting at different localities, cultivars (green and purple), collection periods, stages of harvesting and climatic conditions (Saharkhiz et al., 2015; Padalia and Verma, 2011). Eugenol or methyl eugenol and/or methyl chavicol were found as the major constituents of *Os* essential oil by considering the different harvesting stages and cultivars (Mondello et al., 2002; Brophy et al., 1993). The major diversities in *Ocimum species* were found in Africa followed by South America (Brazil) and Asia (India) (Verma et al., 2015). Eugenol (27–83%) was found as the main component of oils from USA, India, Germany, Thailand, Cuba and Brazil, whereas oil from plants grown in Australia contain mainly methyl chavicol (87%) (Brophy et al., 1993; Kicel et al., 2005; Kothari et al., 2005; Vani et al., 2009; Kelm and Nair, 1998). More interestingly, the decreasing concentration of eugenol and methyleugenol contents in *Os* essential oil in matured leaves might be their involvement in polymerization and synthesis of neolignans (Suzuki et al., 2009), and/or further oxidation of phenolic compounds catalyzed by the increase of polyphenoxidase and peroxidase activity (Dey and Choudhuri, 1983). The aroma compounds of *Os* essential oil (methyl eugenol chemotype, 56.18%) were identified by solid phase microextraction (SPME)/GC–MS/flame ionization detection (FID) and olfactoric evaluations. The spicy-green-notes of *Os* essential oil is due to methyl eugenol, *β*-caryophyllene, *β*-caryophyllene oxide and germacrene D, while spicy-peppery-notes corresponds to germacrene D (Jirovetz et al., 2003). Moreover, the major pharmacological activities of *Os* essential oil such as mosquitocidal, antimicrobial and anthelmintic were found due to its marker constituent eugenol (Kelm and Nair, 1998; Kumar et al., 2010; Asha et al., 2001). *Os* essential oil (40 µg/ml) was found to be non-toxic to the mammalian kidney fibroblast (VERO) and kidney epithelial cells (LLC-PK11) using Neutral Red assay (Zheliazkov et al., 2008).

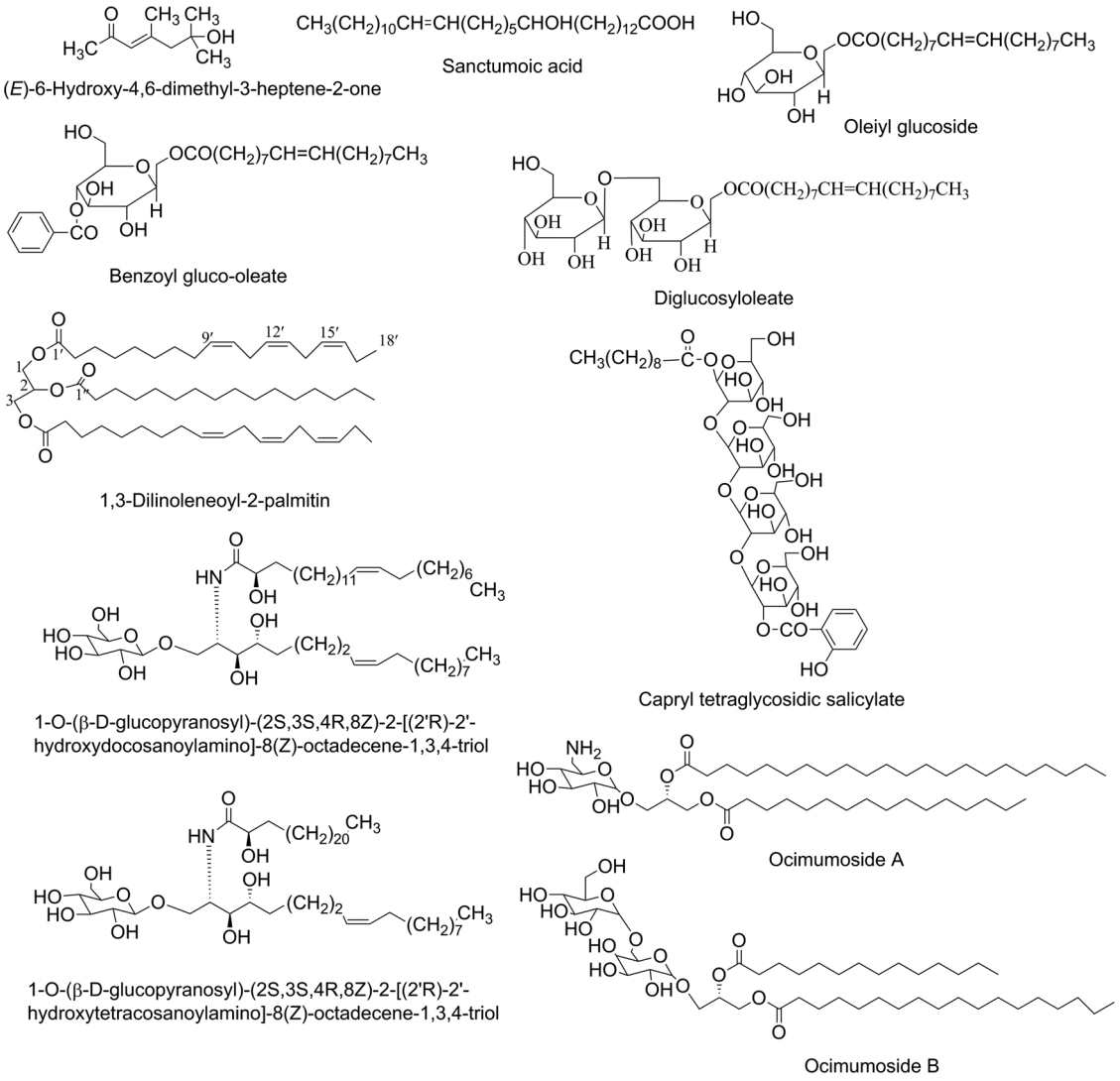


**Fig. 1.** (*continued*)

Effect of different cultivars, plant parts, collection period and geographical distribution on the yield of major components of *Os* essential oil obtained by hydrodistillation are discussed in Table 4.

### 4.9. Fixed oil (non-volatile oil)

The fixed oil content in *Os* seeds was found ~18-22% and, composed of mainly linoleic acid (66.1%), α-linolenic acid (15.7%), oleic acid (9.0%), palmitic acid (6.94%) and stearic acid (2.1%) (Gupta et al., 2002; Angers et al., 1996; Mondal et al., 2009). The major components of fixed oil, linoleic acid and linolenic acid (an ω-3 fatty acid, *cis*-9,12,15-octadecatrienoic acid) were supposed to be responsible for its anti-inflammatory, anticoagulant, hypotensive, chemopreventive, antihypercholesterolaemic and immunomodulatory activities (Singh et al., 2007). Fixed oil of *Os* is reported for anti-inflammatory, anti-arthritic, antimicrobial and antiulcer properties (Singh and Majumdar, 1997; Singh and Majumdar, 1999a; Singh and Majumdar, 1999b; Singh et al., 2001a, 2001b). The anti-inflammatory activity of fixed oil is due to the dual inhibition of arachidonate metabolism and antihistaminic activity (Singh et al., 2007). Only one report is available on the isolation of fixed oil (yield 1.046%) from *Os* leaves, along with the anti-diabetic and antioxidant potential. The fixed oil extracted from *Os* leaves was rich in α-linolenic acid (60.60%), linoleic acid (17.86%) and palmitic acid (15.65%) (Suanarunsawat et al., 2016).

### 4.10. Fatty acid derivatives

Fatty acid derivatives were isolated from the leaves and roots of *Os*, including four cerebrosides (Gupta et al., 2007; Ahmad et al., 2012a). Fatty acid derivatives like palmityl glucoside and sanctumoic acid were exhibited mosquitocidal activity, while cerebrosides showed antistress activity (Kelm and Nair, 1998).

### 4.11. Polysaccharide

A polysaccharide (~$10^6$ Da) isolated from *Os* leaves contain monosaccharide compositions rhamnose (23.3%), xylose (19.2%), arabinose (42.2%), glucose (10.3%) and galactose (5.0%) (Subramanian et al., 2005).

### 4.12. Other secondary metabolites

An acetone oligomer named (*E*)-6-hydroxy-4,6-dimethyl-3-heptene-2-one was isolated as a colorless oil from the aerial parts of *Os* (Kelm and Nair, 1998).

## 5. Pharmacological activities of *Os* secondary metabolites

The chemical constituents of *Os* are mainly studied for its

**Table 3**
Pharmacological activities of secondary metabolites from *O. sanctum*.

| Pharmacological activities | Compounds/plant part | Class of compound | Dose(s) | Results | Reference |
|---|---|---|---|---|---|
| Anticancer | 2-(Hydroxymethyl)-5,5,9-trimethylcyclo [7.2.0.0$^{3,6}$] undecan-2-ol (Lf) | Sesquiterpenoid | – | $IC_{50}$ 30 ± 0.5 μM against MCF-7 cell line | Singh et al. (2014) |
| | Luteolin (Lf and Ap) | Flavonoid | 50 μM | $IC_{50}$ (78 ± 6 μM) for androgen-independent carcinoma of prostate (LNCaP) and $IC_{50}$ (53 ± 4 μM) for androgen-dependent carcinoma of prostate (PC-3 and DU-145) cells at 72 h. | Nagaprashantha et al. (2011) |
| | Orientin (Lf and Ap) | Flavonoid | 50 μM | $IC_{50}$ (124 ± 7 μM) for androgen-independent carcinoma of prostate (LNCaP) and $IC_{50}$ (104 ± 7 μM) for androgen-dependent carcinoma of prostate (PC-3 and DU-145) cells at 72 h. | Nagaprashantha et al. (2011) |
| | Vicenin-2 (Apigenin 6,8-diglucoside (Lf and Ap) | Flavonoid | – | $IC_{50}$ (44 ± 3 μM) for androgen-independent carcinoma of prostate (LNCaP) and $IC_{50}$ (25 ± 3 μM) for androgen-dependent (PC-3, DU-145) cells at 72 h. | Nagaprashantha et al. (2011); Nair et al. (1982); Skaltsa et al. (1999) |
| Antioxidant | Rosmarinic acid (Lf and St) | Phenolic acid | 10 μM | Better antioxidant than vitamin E in liposome oxidation model. | Kelm et al. (2000) |
| | Isothymusin (Lf and St) | Flavonoid | 10 μm | Strong antioxidant activity (50% more active than positive control TBHQ and BHT) in liposome oxidation model. | Kelm et al. (2000) |
| | Isothymonin (Lf and St) | Flavonoid | 10 μm | Better antioxidant activity than positive control TBHQ and BHT using liposome oxidation model. | Kelm et al. (2000) |
| | Cirsimaritin (Lf and St) | Flavonoid | 10 μm | Poor antioxidant using liposome oxidation model. | Kelm et al. (2000) |
| | Cirsilineol (Lf and St) | Flavonoid | 10 μm | Good antioxidant activity using liposome oxidation model. | Kelm et al. (2000) |
| | Eugenol (Lf) | Phenyl propanoid | 10 μm | Better antioxidant activity than positive controls TBHQ and BHT using liposome oxidation model. | Kelm et al. (2000) |
| Anti-inflammatory | Apigenin (Lf, Ap and St) | Flavonoid | 1000 μM | Showed 65% COX-1 enzyme inhibition activity, compared to ibuprofen, naproxen and aspirin with 33%, 58% and 46% at 10, 10 and 1000 μM concentration, respectively. | Kelm et al. (2000) |
| | Rosmarinic acid (Lf and St) | Phenolic acid | 1000 μM | Showed 58% COX-1 enzyme inhibition activity, compared to ibuprofen, naproxen and aspirin with 33%, 58% and 46% at 10, 10 and 1000 μM concentration, respectively. | Kelm et al. (2000) |
| | Isothymusin (Lf and St) | Flavonoid | 1000 μM | Inactive against COX-1 and COX-2 enzyme | Kelm et al. (2000) |
| | Isothymonin (Lf and St) | Flavonoid | 1000 μM | Showed 37% COX-1 enzyme inhibition activity, compared to standard ibuprofen, naproxen and aspirin with 33%, 58% and 46% COX-1 inhibitory activity at 10, 10 and 1000 μM concentration, respectively. | Kelm et al. (2000) |
| | Cirsimaritin (Lf and St) | Flavonoid | 1000 μM | Showed 50% COX-1 enzyme inhibition activity, compared to standard ibuprofen, naproxen and aspirin with 33%, 58% and 46% at 10, 10 and 1000 μM concentration, respectively. | Kelm et al. (2000) |
| | Cirsilineol (Lf and St) | Flavonoid | 1000 μM | Showed 37% COX-1 enzyme inhibition activity, compared to standard ibuprofen, naproxen and aspirin with 33%, 58% and 46% at 10, 10 and 1000 μM concentration, respectively. | Kelm et al. (2000) |
| | 4′,5-Dihydroxy-3′,7,8-trimethoxy flavone (Lf) | Flavonoid | 1000 μM | Showed 37% COX-1 enzyme inhibition activity, compared to standard ibuprofen, naproxen and aspirin with 33%, 58% and 46% at 10, 10 and 1000 μM concentration, respectively. | Kelm et al. (2000) |
| | Eugenol (Lf) | Phenyl propanoid | 1000 μM | Showed 97% COX-1 enzyme inhibition compared to ibuprofen, naproxen and aspirin with 33%, 58% and 46% at 10, 10 and 1000 μM, respectively. | Kelm et al. (2000) |

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**Table 3** (*continued*)

| Pharmacological activities | Compounds/plant part | Class of compound | Dose(s) | Results | Reference |
|---|---|---|---|---|---|
| Radiation protection | Orientin (Lf and Ap) | Flavonoid | 50 µg/kg (i.p.) | Pre-treatment of vicenin protected foetal against irradiation induced genomic damage, and reduced the delayed chromosomal abnormalities and tumorigenesis in pregnant Swiss albino mice. | Uma Devi and Satyamitra (2004) |
| | | | 6.5, 12.5, 15, 17.5 and 20 µM | Pre-treatment significantly ($p < 0.05$–$0.001$) reduced the micronucleus counts to 51–67% of RT with dose modification factor (DMF) of 2.62 at 17.5 µM in cultured human peripheral lymphocytes using micronucleus test. | Vrinda and Uma Devi (2001) |
| | | | 50 µg/kg (*i.p.*) | Pre-treatment showed maximum survival 60% from the 30 days of administration with DMF of 1.30. | Uma Devi et al. (1999) |
| | Vicenin (Lf) | Flavonoid | 50 µg/kg (*i.p.*) | Pre-treatment of vicenin protected foetal against irradiation induced genomic damage, and reduced the delayed chromosomal abnormalities and tumorigenesis in pregnant Swiss albino mice. | Uma Devi and Satyamitra (2004) |
| | | | 6.5, 12.5, 15, 17.5 and 20 µM | Pre-treatment significantly ($p < 0.05$–$0.001$) reduced the micronucleus counts to 51–67% of RT with DMF of 2.48 at 17.5 µM in cultured human peripheral lymphocytes using micronucleus test. | Vrinda and Uma Devi (2001) |
| | | | 50 µg/kg (i.p.) | Pre-treatment showed significant ($p < 0.05$) protection to bone marrow chromosomes. Showed maximum survival 67% from the 30 days of administration with DMF of 1.37. | Uma Devi et al. (1999) |
| Antidiabetic | 16-Hydroxy-4,4,10,13-tetramethyl-17-(4-methyl-pentyl)-hexadecahydro-cyclopenta[a]-phenanthren-3-one (Ap) | Triterpenoid | – | Isolated from antidiabetic activity-guided fraction | Patil et al. (2011) |
| Antistress | Apigenin-7-*O*-*β*-D-glucuronic acid (Lf) | Flavonoid | 40 mg/kg body weight | Ineffective in Sprague-Dawley rats | Gupta et al. (2007); Norr and Wagner (1992) |
| | Ociglycoside-I (4-Allyl-1-*O*-*β*-D-glucopyranosyl-2-hydroxybenzene/or hydroxychavicol glucoside) (Lf) | Phenyl propanoid | 40 mg/kg body weight | Pre-treatment significantly ($p < 0.05$) reduced the increase in corticosterone levels and ($p < 0.01$) reduced the acute stress-induced increase in CK-levels. Less effective on plasma glucose level in acute stress induced Sprague-Dawley rats. | Gupta et al. (2007); Norr and Wagner (1992); Richard et al. (2016) |
| | 7-Hydroxy-3-(2-hydroxyethyl)-4-methyl-*2H*-1-benzopyran-2-one (ocimarin) (Lf) | Coumarin | 40 mg/kg body weight | Ineffective in acute stress male Sprague-Dawley rats | Gupta et al. (2007) |
| | 1-*O*-(*β*-D-glucopyranosyl)-(2*S*,3*S*,4*R*,8*Z*)-2-[(2′*R*)-2′-hydroxydocosanoylamino]-8(*Z*)-octadecene-1,3,4-triol (Lf) | Cerebroside | 40 mg/kg body weight | Inactive | Gupta et al. (2007) |
| | 1-*O*-(*β*-D-glucopyranosyl)-(2*S*,3*S*,4*R*,8*Z*)-2-[(2′*R*)-2′-hydroxytetracosanoylamino]-8(*Z*)-octadecene-1,3,4-triol (Lf) | Cerebroside | 40 mg/kg body weight | Pretreatment showed antistress activity and reduced significantly (ρ < /xps:span > 0.05) the increase corticosterone levels and effective (ρ < /xps:span > 0.01) reducing the creatine kinase levels in Sprague-Dawley rats. | Gupta et al. (2007) |
| | (2*S*)-1-*O*-hexadecanoyl-2-*O*-docosanoyl)-3-*O*-[6-deoxy-6-amino-*α*-D-glucopyranoside]glycerol (Ocimumoside A) (Lf) | Cerebroside | 40 mg/kg body weight | Pretreatment showed significant ($\rho < 0.05$) antistress effects by normalizing hyperglycemia, plasma corticosterone, plasma creatine kinase and adrenal hypertrophy in Sprague-Dawley rats. Ginseng crude powder root, *Panax quinquifolium* (100 mg/kg body weight) was used as standard. | Gupta et al. (2007) |
| | (2*S*)-1-*O*-Octadecanoyl-2-*O*-tetradecanoyl)-3-*O*-[*α*-D-galactopyranosyl-(1″ → 6′)-*O*-*β*-D-galactopyranosyl] glycerol (Ocimumoside B) (Lf) | Cerebroside | 40 mg/kg body weight | Pretreatment showed antistress activity and reduced significantly ($\rho < 0.05$) the increase corticosterone levels without affecting plasma glucose level in Sprague-Dawley rats. | Gupta et al. (2007) |
| Lieshmanicidal | Apigenin ((Lf, Ap and St) | Flavonoid | – | $IC_{50}$ 358.7 µg/ml against *L. major* | |

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**Table 3** (*continued*)

| Pharmacological activities | Compounds/plant part | Class of compound | Dose(s) | Results | Reference |
|---|---|---|---|---|---|
| | | | | | Suzuki et al. (2009) |
| | Luteolin (Lf and Ap) | Flavonoid | – | $IC_{50}$ 73.9 µg/ml against *L. major* | Suzuki et al. (2009); Skaltsa et al. (1999) |
| | 4′,5-Dihydroxy-7,8-dimethoxy flavone (Lf) | Flavonoid | – | $IC_{50}$ > 25 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 4′,5-Dihydroxy-3′,7,8-trimethoxy flavone (Lf) | Flavonoid | – | $IC_{50}$ > 25 µg/ml against *L. major* | Suzuki et al. (2009) |
| | Ferulaldehyde (Lf) | Phenyl propanoid | – | $IC_{50}$ 0.9 µg/ml against *L. major* | Suzuki et al., (2009) |
| | Bieugenol (Lf) | Phenyl propanoid | – | $IC_{50}$ 13.6 µg/ml against *L. major* | Suzuki et al. (2009) |
| | Dehydrodieugenol B (Lf) | Phenyl propanoid | – | $IC_{50}$ 16.9 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 6-Allyl-3′,8-dimethoxy-flavon-3,4′-diol (tulsinol A) (Lf) | Neolignan | – | $IC_{50}$ > 25 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 6-Allyl-3-(4-allyl-2-methoxyphenoxy)-3′,8-dimethoxyflavan-4′-ol (tulsinol B) (Lf) | Neolignan | – | $IC_{50}$ 43.9 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 5-Allyl-3-(4-allyl-2-methoxyphenoxymethyl)-2-(4-hydroxy-3-methoxyphenyl)-7-methoxy-2,3-dihydrobenzofuran (tulsinol C) (Lf) | Neolignan | – | $IC_{50}$ 9.1 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 1,2-Bis(4-allyl-2-methoxyphenoxy)-3-(4-hydroxy-3-methoxyphenyl)-3-methoxypropane (tulsinol D) (Lf) | Neolignan | – | Not done | Suzuki et al. (2009) |
| | 1-(4-Hydroxy-3-methoxyphenyl)-1,2,3-tris (4-allyl-2-methoxyphenoxy)propane (tulsinol E) (Lf) | Neolignan | – | $IC_{50}$ 47.1 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 1-Allyl-4-(5-allyl-2-hydroxy-3-methoxyphenoxy)-3-(4-allyl-2-methoxyphenoxy)-5-methoxybenzene (tulsinol F) (Lf) | Neolignan | – | $IC_{50}$ 23.8 µg/ml against *L. major* | Suzuki et al. (2009) |
| | 3-(5-Allyl-2-hydroxy-3-methoxyphenyl)-1-(4-hydroxy-3-methoxyphenoxy)-prop-1-ene (tulsinol G) (Lf) | Neolignan | – | $IC_{50}$ 89.7 µg/ml against *L. major* | Suzuki et al. (2009) |
| | β-Caryophyllene epoxide (Lf) | Sesquiterpenoid | – | $IC_{50}$ > 25 µg/ml against *L. major* | Suzuki et al. (2009) |
| | Ursolic acid | Triterpenoid | – | $IC_{50}$ 2.2 µg/ml against *L. major* using amphotericin B ($IC_{50}$ 0.04 µg/ml) as positive control | Suzuki et al. (2009) |
| | Oleanolic acid | Triterpenoid | – | $IC_{50}$ 17.1 µg/ml against *L. major* | Suzuki et al. (2009) |
| | β-Sitosterol glucopyranoside (Lf) | Steroid | – | – | Suzuki et al. (2009); Ali and Ali (2012) |
| | Stigmasterol (Lf) | Steroid | – | $IC_{50}$ > 25 µg/ml against *L. major* | Suzuki et al. 2009 |
| Antimicrobial | Orientin (Lf and Ap) | Flavonoid | 400 mg/ml | Active against *S. aureus*, *S. cohni* and *K. pneumonia* with maximum zone inhibition (18.04, 17.13 and 16.11 mm). | Ali and Dixit (2012); Skaltsa et al. (1999) |
| | Vicenin (Lf) | Flavonoid | 400 mg/ml | Effective against *E. coli* and *Proteus* with maximum ZOI (18.84 and 17.16 mm). | Ali and Dixit (2012) |

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**Table 3** (*continued*)

| Pharmacological activities | Compounds/plant part | Class of compound | Dose(s) | Results | Reference |
|---|---|---|---|---|---|
| Mosquitocidal | Eugenol (Lf) | Phenyl propanoid | 100–250 ppm | $LD_{100}$ (200 μg/ml) against fourth-instar *Aedes aegyptii* larvae. | Kelm and Nair (1998) |
| | (*E*)-6-Hydroxy-4,6-dimethyl-3-heptene-2-one (Lf and St) | Acetone oligomer | – | $LD_{100}$ (6.25 μg/ml) against fourth-instar *Aedes aegyptii* larvae in 24 h. | Kelm and Nair (1998) |
| | 1,3-dilinoleneoyl-2-palmitin (Lf and St) | Triglyceride | – | Inactive against *Aedes aegyptii* larvae. | Kelm and Nair (1998) |

Abbreviations: Ap (areal part); Lf (leaf); Rt (root); St (stem); Tw (twig).

therapeutic potential like anticancer, antioxidant, anti-inflammatory, leishmanicidal, radiation protective, mosquitocidal, antimicrobial and antistress activity. The pharmacological activities of *Os* secondary metabolites are discussed here and summarized in Table 3.

### 5.1. Anticancer activity

A tricyclic sesquiterpenoids 2-(hydroxymethyl)-5,5,9-trimethylcyclo[7.2.0.$0^{3,6}$]undecan-2-ol isolated from the oil of *Os* leaves showed antiproliferative activity against MCF-7 cell line ($IC_{50}$ 30 ± 0.5 μM) using doxorubicin as standard ($IC_{50}$ 9.7 μg/ml) (Singh and Chaudhuri, 2013; Singh et al., 2014). In continuation of antiproliferative screening against MCF-7 cell line, sesquiterpenes *β*-caryophyllene, 4,5-epoxycaryophyllene and 5*β*-hydroxycaryophyllene showed $IC_{50}$ values 73.0, 7.0 and 4.8 μg/ml, respectively. Compounds rosmarinic acid, apigenin, luteolin, orientin, vicenin-2, ursolic acid and oleanolic acid are well studied for their anticancer potential (Nagaprashantha et al., 2011). The terpenoids and flavonoids are the major class of compounds responsible for the anticancer activity of *Os*.

### 5.2. Antioxidant activity

Phenolics/flavonoids of *Os* were investigated for their free radical scavenging activity (Kelm et al., 2000). The antioxidant activity guided isolation of *Os* leaves and stems in liposome oxidation model yielded six flavonoids including apigenin, rosmarinic acid, isothymusin, isothymonin, cirsimaritin, cirsilineol along with eugenol (Kelm et al., 2000). Compounds isothymusin, isothymonin and eugenol showed good antioxidant activity at 10 μM, compared to standards TBHQ (terbutyl hydroquinone) and BHT (butylated hydroxyl toluene). Koroch et al. (2010) found rosmarinic acid as the main constituent responsible for the antioxidant activity of *Os* due to its rapid scavenging effect of free radicals. A polysaccharide (constituted of 23.3% rhamnose; 19.2% xylose; 42.2% arabinose; 10.3% glucose and 5% galactose) isolated from *Os* leaves demonstrated antioxidant activity in DPPH free radical scavenging, anti-lipid peroxidation, hydrogen peroxide scavenging and superoxide radical scavenging assays (Subramanian et al., 2005). The polysaccharide showed potent DPPH free radicals scavenging activity with $IC_{0.2}$ value of 5.61 ± 0.17 μg/ml, compared to α-tocopherol ($IC_{0.2}$ = 11.9 ± 0.2 mM) and BHA ($IC_{0.2}$ = 14.5 ± 2.5 mM). Also, *Os* polysaccharide scavenged ~ 54% and ~79% of superoxide free radicals at 10 and 50 μg/ml, respectively. The antioxidant results showed that *Os* polysaccharide possesses reactive oxygen species scavenging and iron chelating properties. The pretreatment of *Os* polysaccharide at 100 μg/ml protects 30 ± 3.2% mouse splenocytes against γ-ray irradiation. The antioxidant potential of *Os* polysaccharide against oxidative damage to lipid, DNA and splenocytes warrants its application in radiation protection.

### 5.3. Anti-inflammatory activity

*Os* leaves and seeds are reported for reducing the level of uric acid, the causing factor of arthritis and joint inflammation (Sarkar et al., 1990). The anti-inflammatory activity of compounds isolated from *Os* aerial parts named rosmarinic acid, apigenin, isothymusin, isothymonin, cirsimaritin, cirsineol and eugenol has been evaluated for cyclooxygenase-1 (COX-1) and COX-2 inhibitory activities in human prostaglandin H synthase isozymes (hPGHS-1). Eugenol was the most active compound and showed 97% of COX-1 inhibition at 1000 μM, compared to ibuprofen, naproxen and aspirin with 33%, 58% and 46% COX-1 inhibition at 10, 10 and 1000 μM, respectively (Kelm et al., 2000). Moreover, cirsineol, cirsimaritin, isothymonin and apigenin showed 37%, 50%, 37% and 65% COX-1 enzyme inhibition, respectively. Singh et al. (1996) were found that *Os* volatile oil inhibits arachidonic and leukotriene induced inflammation via cyclooxygenase inhibition and lipo-oxygenase pathways in arachidonic acid metabolism. Whereas, *Os* fixed oil exhibited anti-inflammatory effect due to the dual inhibition of arachidonate metabolism and antihistaminic activity (Singh et al., 2007).

### 5.4. Radiation protective activity

The radioprotective effect of *Os* was first investigated in the aqueous extract of leaves by Uma Devi and Ganasoundari (1995). The optimum dose of extract for radiation protection was found to be 50 mg/kg b.w. (*i.p.*), with $LD_{50}$ 6.0 g/kg body weight. Further, chemical investigation on *Os* aqueous extract gave two water soluble flavonoids orientin (8-*C*-*β*-D-glucopyranosyl-luteolin) and vicenin (8-*C*-*β*-D-xylopyranosyl-8-*C*-*β*-D-glucopyranosyl apigenin). Both, the flavonoids exhibited protective effect against radiation-induced chromosomal damage in mice due to their free radical scavenging and metal chelating effects (Uma Devi et al., 2000). The iron chelating effect of flavonoids inhibited the formation of thiobarbituric acid reactive substances (TBRAS) that protects lipid peroxidation initiated by iron ion bound to the lipid membrane (Uma Devi et al., 2000). Subsequently, Nayak and Uma Devi (2005) have investigated the optimum dose for flavonoids orientin and vicenin i.e. 50 μg/kg body (in vivo) in radiation protection against bone marrow damage. Both flavonoids showed similar protective effects at doses 50 μg/kg, whereas vicenin at a higher dose (150 mg/kg) showed better bone marrow protection (Uma Devi et al., 1998). Additionally, vicenin showed better survival effects than orientin at 30 days with long lasted protective effects (Uma Devi et al., 1999). Also, the dose modification factor was found to be higher for vicenin ($LD_{50}$ = 1.37) than orientin ($LD_{50}$ = 1.30) and exerted equal protective effects against γ-ray-induced lipid peroxidation in mouse liver. The low and non-toxic concentration of orientin and vicenin showed significant radiation protection to human peripheral lymphocytes (in vitro), suggest their clinical application in cancer radiotherapy as normal tissues protector (Vrinda and Uma Devi, 2001).

### 5.5. Antihyperlipidaemic and antidiabetic activity

*Os* leaves have been studied for serum lipid lowering activity in both normal albino rabbits and diabetic rats, the antihyperlipidaemic effect

**Table 4**
Variation studies of major chemical components of essential oil from *O. sanctum*.

| Compounds | Content (%), cultivar (collection stage) | Plant parts | Country of origin | References |
|---|---|---|---|---|
| Monoterpenoids | | | | |
| (E)-β-Ocimene | 5.8 (green cultivar) and 0.9 (purple cultivar) | Fresh aerial parts | India | Padalia and Verma (2011) |
| | 4.2 (vegetative), 10.6 (full bloom) and 6.6 (seed setting) in green cultivar; 5.0 (vegetative), 6.4 (full bloom) and 4.2 (seed setting) in purple cultivar | Fresh aerial parts (leaves, stem and inflorescence) | India | Padalia et al. (2013) |
| D-Limonene | 3.8 (green cultivar) and 0.6 (purple cultivar) | Leaves and inflorescence | Bangladesh | Mondello et al. (2002) |
| | 4.39 | Dried leaves and stem | India | Khan et al. (2010a, 2010b) |
| Linalool | 21.84 | Dried leaves and stem | India | Khan et al. (2010a, 2010b) |
| 1,8-Cineole (eucalyptol) | 0.7 | Fresh leaves | India | Raju et al. (1999) |
| | 8.9 (pre-flowering), 33.0 (flowering), 32.2 (end of flowering) and 15.3 (fruit bearing) | Dried aerial parts | Poland | Kicel et al. (2005) |
| | 20.78 (vegetative), 19.41 (floral budding) and 20.45 (flowering) | Aerial parts | Iran | Saharkhiz et al. (2015) |
| Sesquiterpenoids | | | | |
| α-Caryophyllene (α-humulene) | 2.0 | Dried stalks and leaves | Cuba | Pino et al. (1998) |
| | 1.7 | Fresh leaves | India | Raju et al. (1999) |
| | 5.3 (green cultivar) and 2.1 (purple cultivar) | Leaves and inflorescence | Bangladesh | Mondello et al. (2002) |
| | 8.1 (flowering) | Fresh leaves | Brazil | Trevisan et al. (2006) |
| | 3.3 | Dried leaves and stem | India | Khan et al. (2010a, 2010b) |
| β-Caryophyllene | 5.33 | Dried leaves | Northern Australia | Brophy et al. (1993) |
| | 23.1 | Dried stalks and leaves | Cuba | Pino et al. (1998) |
| | 9.8 | Leaves | Brazil | Machado et al. (1999) |
| | 40.7 | Inflorescence | Brazil | Machado et al. (1999) |
| | 31.7 | Fresh leaves | India | Raju et al. (1999) |
| | 24.4 (green cultivar) and 10.7 (purple cultivar) | Leaves and inflorescence | Bangladesh | Mondello et al. (2002) |
| | 16.60 | Fresh leaves | Southern India | Jirovetz et al. (2003) |
| | 4.37(full bloom) | Main branch | Southern India | Kothari et al. (2004) |
| | 7.97 (full bloom) | Shoot biomass cut at 30 cm above ground | Southern India | Kothari et al. (2004) |
| | 6.37 (full bloom) | Stem | India | Kothari et al. (2005) |
| | 11.97 | Inflorescence | India | Kothari et al. (2005) |
| | 29.4 (flowering) | Fresh leaves | Brazil | Trevisan et al. (2006) |
| | 27.6 (green cultivar) and 17.1 (purple cultivar) | Fresh plant | Northern India | Awasthi and Dixit (2007) |
| | 33 | – | India | Dohi et al. (2009) |
| | 11.89 | Fresh leaves | India | Kumar et al. (2010) |
| | 7.3 (green cultivar) and 8.4 (purple cultivar) | Fresh aerial parts | India | Padalia and Verma (2011) |
| | 12.6 (vegetative), 9.2 (full bloom), 6.6 (seed setting) in green cultivar; 15.6 (vegetative), 8.0 (full bloom) 7.8 (seed setting) in purple cultivar | Fresh aerial parts (leaves, stem and inflorescence) | India | Padalia et al. (2013) |
| | 5.5 | Dried aerial parts | India | Verma et al. (2015) |
| β-Caryophyllene oxide | 3.8 | Dried stalks and leaves | Cuba | Pino et al. (1998) |
| | 0.0 | Leaves and inflorescence | Brazil | Machado et al. (1999) |
| | 18.5 | Leaves and inflorescence | Brazil | Machado et al. (1999) |
| | 1.5 | Fresh leaves | India | Raju et al. (1999) |
| | 5.1 (green cultivar) and 1.1 (purple cultivar) | Leaves and inflorescence | Bangladesh | Mondello et al. (2002) |
| | 1.10 | Fresh leaves | Southern India | Jirovetz et al. (2003) |
| | 0.75 (full bloom) | Leaves | India | Kothari et al. (2005) |
| | 1.70 | Stem | India | Kothari et al. (2005) |
| | 3.02 | Inflorescence | India | Kothari et al. (2005) |
| | 2.7 | Fresh aerial parts | Nigeria | Gbolade and Lockwood (2008) |
| | 7.5 | Dried aerial parts | India | Verma et al. (2015) |
| β-Bisabolene | 15.4 (pre-flowering), 14.4 (flowering), 13.0 (end of flowering) and 20.4 (fruit bearing) | Dried leaves | Poland | Kicel et al. (2005) |
| | 20.99 (vegetative), 13.29 (floral budding) and 18.76 (flowering) | Aerial parts | Iran | Saharkhiz et al. (2015) |
| | 4.3 | Dried aerial parts | India | Verma et al. (2015) |
| γ-Elemene | 10.47 (vegetative), 7.7 (floral budding) and 7.8 (flowering) | Aerial parts | Iran | Saharkhiz et al. (2015) |

(*continued on next page*)

**Table 4** (*continued*)

| Compounds | Content (%), cultivar (collection stage) | Plant parts | Country of origin | References |
|---|---|---|---|---|
| *β*-Elemene | 18.0 | Dried stalks and leaves | Cuba | Pino et al. (1998) |
| | 5.0 | Leaves | Brazil | Machado et al. (1999) |
| | 5.8 | Inflorescence | Brazil | Machado et al. (1999) |
| | 6.2 | Fresh leaves | India | Raju et al. (1999) |
| | 1.73 | Fresh leaves | Southern India | Jirovetz et al. (2003) |
| | 2.54 (full bloom) | Main branch | Southern India | Kothari et al. (2004) |
| | 16.3 (green cultivar) and 4.3 (purple cultivar) | Fresh plant | Northern India | Awasthi and Dixit (2007) |
| | 8.8 | Fresh aerial parts | Nigeria | Gbolade and Lockwood (2008) |
| | 11.0 (green cultivar) and 10.9 (purple cultivar) (full bloom) | Fresh aerial parts | India | Padalia and Verma (2011) |
| | 10.8 (vegetative), 9.8 (full bloom) and 11.3 (seed setting) in green cultivar; 17.7 (vegetative), 14.3 (full bloom) and 9.6 (seed setting) in purple cultivar | Fresh aerial parts (leaves, stem and inflorescence) | India | Padalia et al. (2013) |
| Germacrene D | 5.10 | Fresh leaves | Southern India | Jirovetz et al. (2003) |
| | 9.14 | Fresh leaves | India | Kumar et al. (2010) |
| | 0.1 (green cultivar) and 1.4 (purple cultivar) | Fresh plant | Northern India | Awasthi and Dixit (2007) |
| | 2.4 (green cultivar) and 2.2 (purple cultivar) (blooming) | Fresh aerial parts | India | Padalia and Verma (2011) |
| 9-*epi*-(E)-Caryophyllene | 23.68 | Fresh leaves | Thailand | Suanarunsawat et al. (2016) |
| Selinene | < 0.1 (vegetative), 0.1 (full bloom) and 0.1 (seed setting) in green cultivar; 12.3 (vegetative), 9.4 (full bloom) and 9.7 (seed setting) in purple cultivar | Fresh aerial parts (leaves, stem and inflorescence) | India | Padalia et al. (2013) |
| Phenylpropanoids | | | | |
| Eugenol | 34.3 | Dried stalks and leaves | Nigeria | Pino et al. (1998) |
| | 79 | Leaves | Brazil | Machado et al. (1999) |
| | 17.6 | Inflorescence | Brazil | Machado et al. (1999) |
| | 53.4 | Fresh leaves | India | Raju et al. (1999) |
| | 41.7 (green cultivar) and 77.5 (purple cultivar) | Leaves and inflorescence | Bangladesh | Mondello et al. (2002) |
| | 1.66 | Fresh leaves | Southern India | Jirovetz et al. (2003) |
| | 7.28 (main branch), 8.48 (secondary branch) and 4.36 (shoot biomass cut at 30 cm above ground) (full bloom) | Branches | India | Kothari et al. (2004) |
| | 0.79 (full bloom) | Leaves | India | Kothari et al. (2005) |
| | 8.9 (pre-flowering), 4.1 (flowering), 4.6 (end of flowering) and 7.1 (fruit bearing) | Dried aerial parts | Poland | Kicel et al. (2005) |
| | 59.4 (flowering) | Fresh leaves | Brazil | Trevisan et al. (2006) |
| | 46.2 (green cultivar) and 0.6 (purple cultivar) | Fresh plant | Northern India | Awasthi and Dixit (2007) |
| | 0.4 | Fresh aerial parts | Nigeria | Gbolade and Lockwood (2008) |
| | 59 | – | India | Dohi et al. (2009) |
| | 61.30 | Fresh leaves | India | Kumar et al. (2010) |
| | 18.25 | Fresh leaves | Thailand | Suanarunsawat et al. (2016) |
| | 67.4 (green cultivar) and 72.8 (purple cultivar) (full bloom) | Fresh aerial parts | India | Padalia and Verma (2011) |
| | 62.2 (vegetative), 64.1 (full bloom) and 58.3 (seed setting) in green cultivar;35.3 (vegetative), 45.7 (full bloom) and 44.9 (seed setting) in purple cultivar | Fresh aerial parts (leaves, stem and inflorescence) | India | Padalia et al. (2013) |
| | 10.66 | – | Thailand | Sutaphanit and Chitprasert, (2014) |
| | 41.70 (green cultivar) and 77.50 (purple cultivar) | – | India | Pandey et al. (2014) |
| | 15.70 (vegetative), 37.15 (floral budding) and 24.63 (flowering) | Aerial parts | Iran | Saharkhiz et al. (2015) |
| Methyl eugenol | traces (green cultivar) and 0.1 (purple cultivar) | Leaves and inflorescence | Bangladesh | Mondello et al. 2002 |
| | 56.18 | Fresh leaves | Southern India | Jirovetz et al. (2003) |
| | 75.03 (full bloom) | Main branch | Southern India | Kothari et al. (2004) |
| | 75.25 (full bloom) | Leaves | India | Kothari et al. (2005) |
| | 83.70 | Stem | | |
| | 65.20 (full bloom) | Inflorescences | India | Kothari et al. (2005) |
| | Not detected (green cultivar) and 67.8 (purple cultivar) | Fresh plant | Northern India | Awasthi and Dixit (2007) |
| | 44.7 | Fresh aerial parts | Nigeria | Gbolade and Lockwood (2008) |
| | 47.06 | Fresh leaves | Thailand | Suanarunsawat et al. (2016) |
| | 42.58 | – | Thailand | Sutaphanit and Chitprasert (2014) |
| | 50.9 | Dried aerial parts | India | Verma et al. (2015) |
| Methyl chavicol (estragole) | 44.63 | Dried leaves and stem | India | Khan et al. (2010a, 2010b) |
| | 11.49 (vegetative), 10.61 (floral budding) and 11.40 (flowering) | Aerial parts | Iran | Saharkhiz et al. (2015) |
| | 1.9 (pre-flowering), 12.5 (flowering), 10.2 (end of flowering) and 5.2 (fruit bearing) | Dried leaves | Poland | Kicel et al. (2005) |
| | 87.69 | Dried leaves | Northern Australia | Brophy et al. (1993) |

is mainly due to its essential oil content (Suanarunsawat et al., 2016). *Os* essential oil riched with eugenol (18.25%), methyl eugenol (47.06%) and *β*-caryophyllene (23.68%) has been reported to suppress the serum total cholesterol (93.62 ± 3.29) mg/dl and triglycerides (36.29 ± 3.33 mg/dl) in hypercholesterolaemic rats, compared to the negative control i.e. high cholesterol treated rats (total cholesterol, 138.12 ± 10.21 mg/dl and triglyceride, 50.79 ± 2.86 mg/dl). *Os* essential oil also showed antihyperlipidaemic effects comparable with standard drug simvastatin (total cholesterol, 90.35 ± 5.70 mg/dl and triglyceride, 48.50 ± 4.35 mg/dl). The antihyperlipidaemic activity of *Os* essential oil was due to the suppression of liver lipid synthesis, and the presence of phenylpropanoid constituents. These antihyperlipidaemic results suggest that *Os* essential oil is potentially beneficial in the prevention and treatment of diseases like atherosclerosis and cardiovascular disorders (Suanarunsawat et al., 2009).

The fixed oil obtained from fresh *Os* leaves constitute of mainly α-linolenic acid (60.60%), which significantly lower the diabetically-elevated blood glucose levels and serum lipid profile with an increase in serum insulin levels in streptozotocin-induced type 1 diabetes mellitus rats within three weeks (Suanarunsawat et al., 2016). Fixed oil prevents renal injury caused by diabetes mellitus and significantly increases the serum insulin (4.50 ± 0.24 μU/ml) and lower the serum lipid profile (total cholesterol, 70.0 ± 4 mg/dl; triglyceride 45.0 ± 8), compared to the untreated group (serum insulin, 3.22 ± 0.18 μU/ml; total cholesterol, 93.0 ± 7 mg/dl and triglyceride 92.0 ± 7). Fixed oil decreases the levels of creatinine and blood urea nitrogen ($p < 0.001$) 1.27 ± 0.18 mg/dl and 20.2 ± 0.7 mg/dl, respectively. Additionally, fixed oil suppresses the elevated TBRAS level and increases the activity of antioxidative enzymes in the liver and cardiac tissues. Further, exploration of fixed oil potential in the type 2 diabetes mellitus is recommended.

A tetracyclic triterpenoid, 16-hydroxy-4,4,10,13-tetramethyl-17-(4-methyl-pentyl)-hexadecahydro-cyclopenta [a]-phenanthren-3-one isolated from the antidiabetic activity guided fraction of hydro alcoholic extract of *Os* aerial parts (Patil et al., 2011). The bioactive fraction (20 mg/kg) exhibited significant ($p < 0.001$) anti-diabetic activity and decreases the level of serum glucose, triglycerides, LDL cholesterol and total cholesterol in alloxan induced diabetic rats.

### 5.6. Antistress activity

*Os* is well known for its adaptogenic and immunomodulatory properties since ancient time and these potentials credited to its antistress activity. The designed extract named *OciBest* obtained by the blending of water and methanol extracts of *Os* whole plant with the required level of active constituents, ociglycoside-I (> 0.1%w/w), rosmarinic acid (> 0.2%w/w), oleanolic acid and ursolic acid (> 2.5%), which was found to be effective against chronic variable stress (Richard et al., 2016). The antistress effects were studied on cortisol release and CHHR1 receptor activity using cell-based assay, while 11*β*-hydroxysteroid dehydrogenase type-1 (11β-HSD1) and catechol-*O*-methyltransferase (COMT) for cell-free assays. Further, *OciBest* showed inhibitory activity on COMT ($IC_{50}$ = 11.65 μg/ml) and 11*β*-HSD1 (99.96% at 200 μg/ml) compared to the standard 3,5-dinitrocatechol ($IC_{50}$ = 24.91 nM) and carbenoxolone (61.44% at 600 nM). Moreover, *Os* (6.25–100 μg/ml) also inhibits cortisol release in forskolin-induced human adreno-carcinoma cells (NCI-H295R) and this effect might be attributed by ursolic acid (625 μM to 10 μM) (Sembulingam et al., 1997; Richard et al., 2016). Thus, inhibition of cortisol release, blocking the CRHR1 receptor, inhibition of 11β-HSD1 and COMT effects are found to be responsible for the antistress activity of *Os* (Richard et al., 2016).

In a separate study, the ethanol extract of *Os* leaves and its *n*-butanol fraction significantly ($p < 0.05$) normalize the acute stress and chronic unpredictable stress at a dose of 200 mg/kg body weight, compared to the standard drug *Panax quinquifolium* at a dose of 100 mg/kg body weight (Gupta et al., 2007). Further, the antistress activity guided isolation of *n*-butanol fraction led to yield three compounds ociglycoside-I, ocimumoside A and ocimumoside B (Gupta et al., 2007). The prior treatment of all three compounds significantly reduces the increased cortisone levels ($p < 0.05$) and creatine kinase levels ($p < 0.01$) at a dose of 40 mg/kg body weight in acute stress induced rats, compared to the normal group. Among all these compounds, ocimumoside A showed potent antistress effects by normalizing hyperglycemia, plasma corticosterone, plasma creatine kinase and adrenal hypertrophy (Gupta et al., 2007). Ociglycoside-I and ocimumoside B were found to be effective against normalizing stress parameters, while ineffective on plasma glucose levels.

The extended study on antistress potential of ocimumoside A and B on chronic unpredictable stress (CUS) normalizes the stress-induced responses like alterations in monoaminergic (nor-adrenalin, dopamine, serotonin and their metabolites like dihydroxyphenylacetic acid, homovanillic acid and 5-hydroxyindole acetic acid), antioxidant systems and changes in plasma corticosterone levels (Ahmad et al., 2012b). Pretreatment of ocimumoside A and B (40 mg/kg body weight *p.o.*) in CUS-induced animals significantly reduce the plasma corticosterone levels 211.50 ± 13.67 and 225.13 ± 13.28, respectively. These results were efficacious with standard antioxidant, melatonin (211.57 ± 117.73 at 20 mg/kg *i.p.*) and CUS-induced animals as control (308.11 ± 24.59). Additionally, ocimumoside A and B normalizes the CUS-induced perturbations of enzymatic activities such as glutathione level and lipid peroxidation in seven days. Interestingly, these compounds do not show any alterations in the baseline values of stress related parameters, when administered alone. Further, the researchers warrant establishing the pathway to modulate the neurotransmitter levels by these compounds. These findings suggest the traditional application of *Os* in adaptogen with modern pharmacological activities. The antistress potential of *Os* compounds shows its application in the treatment of stress-induced neurological disorders and suggest for future studies.

### 5.7. Lieshmanicidal activity

The essential oil from *Os* exhibited leishmanicidal activity against *Leishmania donovani* (Zheliazkov et al., 2008). The essential oil demonstrated leishmanicidal activity with $IC_{50}$ = 37.3 ± 4.6 μg/ml and $IC_{90}$ = 90.0 ± 4.6 μg/ml, using pentamidine ($IC_{50}$ = 1.46 ± 0.51 μg/ml and $IC_{90}$ = 4.98 ± 1.1 μg/ml) and amphotericin B ($IC_{50}$ = 0.09 ± 0.01 μg/ml and $IC_{90}$ = 0.35 ± 0.12 μg/ml) as positive controls. Interestingly, the major contents of essential oil eugenol and methyl chavicol did not possess leishmanicidal activity, while minor constituent (+)-δ-cadinene (yield 0.168 ± 0.0194%) showed potent leishmanicidal activity ($IC_{50}$ = 4.0 μg/ml and $IC_{90}$ = 7.0 μg/ml).

The hydroalcoholic extract of *Os* leaves inhibits the growth of promastigotes of *L. amazonensis* by 8.8 ± 1.2% at 50 μg/ml and 10.3 ± 1.3% at 100 μg/ml, compared to pentamidine 96.9 ± 0.2% and 99.2 ± 0.3% at 50 and 100 μg/ml (Garcia et al., 2010). The leishmanicidal activity guided isolation of *Os* ethyl acetate fraction resulted ferulaldehyde and ursolic acid with $IC_{50}$ values 0.9 μg/ml and 2.2 μg/ml, respectively against promastigotes of *L. major* compared to positive control amphotericin B ($IC_{50}$ = 0.04 μg/ml) (Suzuki et al., 2009). Eugenol and caryophyllene oxide showed $IC_{50}$ values > 25 μg/ml against *L. major*, while eugenol dimers bieugenol ($IC_{50}$ = 13.6 μg/ml) and dehydrodieugenol ($IC_{50}$ = 16.9 μg/ml) were found better leishmanicidal components. Also, a novel neolignan tulsinol C (5-Allyl-3-(4-allyl-2-methoxyphenoxymethyl)-2-(4-hydroxy-3-methoxyphenyl)-7-methoxy-2,3-dihydrobenzofuran) exhibited potent leishmanicidal activity with $IC_{50}$ value 9.1 μg/ml against *L. major* (Suzuki et al., 2009).

### 5.8. Antimicrobial activity

*Os* flavonoids orientin and vicenin were screened against bacterial

strains causing urinary tract infection in human e.g. *Staphylococcus aureus*, *Staphylococcus cohni* (gram positive), and *Escherichia coli*, *Proteus* and *Klebsialla pneumonia* (gram negative) using disc diffusion method (Ali and Dixit, 2012). Orientin (400 mg/ml) showed antibacterial activity against *S. aureus, S. cohni* and *K. pneumonia* with maximum zone inhibition (ZOI) of 18.04, 17.13 and 16.11 mm, respectively. While, vicenin at 400 mg/ml was found to be active against *E. coli* (ZOI, 18.84 mm) and *Proteus* (ZOI, 17.16 mm). Moreover, the synergistic effect of orientin and vicenin (in a ratio of 1:1) on antibacterial activity showed better results in all the strains than individual flavonoids with maximum ZOI of 20.12, 20.75, 20.95 and 20.31 mm at 400 mg/ml concentrations against *E. coli*, *Proteus*, *S. aureus, S. cohni* and *K. pneumonia*, respectively. The antibacterial activity results were concluded that the potent synergistic effect of flavonoids orientin and vicenin can be used as a new choice for the treatment of bacterial infected UTI infections. Further, the use of positive control during antibacterial screening is recommended to validate these results.

### 5.9. Mosquitocidal activity

The mosquitocidal activity against *Aedes aegyptii* larvae guided fraction of *Os* yielded two compounds eugenol and (*E*)-6-hydroxy-4,6-dimethyl-3-heptene-2-one (Kelm and Nair, 1998). Eugenol and (*E*)-6-hydroxy-4,6-dimethyl-3-heptene-2-one demonstrated mosquitocidal activity with $LD_{100}$ values 200 µg/ml and 6.25 µg/ml, respectively in 24 h, while there was no mortality for control larvae. Further, the researchers suggest to investigate the different *Os* extract in search of novel mosquitocidal compounds.

## 6. Conclusions and future prospects

*Os* has been studied for the bio-assay guided isolation of chemical constituents as well as in search of novel molecules from different extracts. Several chemical class of compounds including phenolics, flavonoids, phenylpropanoids, neolignans, terpenoids, coumarins, fatty acid derivatives, essential oil and fixed oil have been reported from this herb. The essential oil of *Os* is a good source of natural eugenol and well explored in analytical, chemical and biological aspects due to its high commercial importance in pharmaceutical, cosmetics and food industry. Fixed oil of the seeds is rich in ω-3 fatty acids and is the recent interest of the research, due to its wide range of pharmacological properties especially in cardioprotection. Flavonoids are the major class of compounds isolated from *Os* and have been found as the main active constituents. The water-soluble flavonoids, orientin and vicenin have been well explored in terms of their radiation protective effects at lower and higher doses. The hydrophilic character of both the flavonoids makes them useful for their antioxidant effect in detoxification as well as radiation protector in cancer therapy. The traditional importance of *Os* as immunomodulator herb was further supported by the isolation of antistress molecules, ocimumoside A and ocimumoside B, which suggest the application of *Os* in the treatment of neurological disorders. Further studies are needed to explore the molecular and cellular mechanism of antistress activity of ocimumoside A and ocimumoside B. So far, the research work carried out on *Os* is mainly focused on the biological activity of its different extracts and essential oil. The literature study showed that eugenol, ursolic acid, rosmarinic acid, orientin, vicenin, ocimumoside A and ocimumoside B are the main active chemical constituents of *Os* and suggesting an opportunity of finding new bioactive molecules.

However, there are several aspects that needed to explore and investigate further: (1) work on the large scale of plant material to isolate sufficient amount of major as well as minor chemical constituents to explore their pharmacological activities and mechanism for therapeutic potential; (2) explore pre-clinically studied compounds for clinical practices, especially in antistress and radiation protection; (3) limited work has been carried out for the isolation and biological studies on the root extracts of *Os*; (4) more than 60 compounds have been isolated, whereas only a few have been explored for pharmacological activities and pre-clinical studies. Overall, there is a need to further research on the chemical aspects of *Os* to get novel molecules with new pharmacological potential. The long history of traditional uses, wide spectrum of pharmacological properties and toxicity studies suggested *Os* as a safe valuable herb for clinical applications. The present compilation of chemical constituents along with their pharmacological properties will be helpful in future studies on *Os* plant as well as in search of new leads for drug discovery.

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