Chemical Constituents and Pharmacological Activities of *Marrubium vulgare* L., an Important Medicinal Herb

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Abstract

Marrubium vulgare L. (Lamiaceae), popularly called pahari gandana (Hindi) and truppad (Kashmiri), is a herb indigenous to Asia, Europe, and the Mediterranean region. In India, this species is mainly found in Kashmir at an altitude of 1524–2438 meters. In traditional medicine, *M. vulgare* is used in Europe, Tunisia, Brazil, and Pakistan to cure ailments associated with respiration such as asthma and cough. The phytochemical investigation showed the availability of flavonoids, phenylpropanoid esters, steroids, tannins, saponins, and terpenoids as major metabolites. Its aerial parts mainly contain marrubiin, a furan labdane diterpenoid, considered an important marker compound of *Marrubium* genus. Pharmacological studies have shown that *M. vulgare* exhibits antispasmodic, antinociceptive, antihypertensive, antidiabetic, gastroprotective, antioxidant, anti-inflammatory, and hepatoprotective properties. The present chapter summarizes all the scientific researches so far being done on the plant to make an attempt to unveil its secondary metabolites so that their therapeutic properties could be assessed.

Keywords

Angiosperms · *Marrubium vulgare* · Marrubiin · Flavonoids · Phenylpropanoid · Antidiabetic

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12.1 Introduction

Plants have been a very important part of human civilization since the ancient times when man was learning to survive. Humans utilized plants and plant products to fulfill all the most important necessities for their nourishment, and their existence is extremely reliant on primary producers, mostly plants. Near about 5000 plant species have been widely utilized by humans as food. Besides fulfilling primary needs, the plant species have been utilized as medicines for various human illnesses (Sullivan and Shealy 1997). Such plants are called traditional medicinal plants and have played the most important roles in our ancient system of medicine. Those plants have been provided hypothesis and become primary source of modern drug discovery by the pharmaceutical industry. The World Health Organization has prepared a list of over 21,000 species of plants that are in use as traditional medicines around the world, with more than 80% of the world's population believing on plant-based medicines for their primary health care (Gurib-Fakim 2006).

Among a total of 21,000 medicinal plant species, the family Lamiaceae (also mint family) is known as the biggest family explored for the discovery of bioactive secondary metabolites (Wink 2003). This family is represented by 7200 species belonging to 236 genera (Brauchler et al. 2010). Many species belonging to this family are greatly aromatic and mainly produce oil that evaporates in low temperature due to the existence of peripheral glandular structures (Giuliani and Bini 2008). Among them, the genus *Marrubium* L. has attracted most of the researchers around the world, due to the presence of active chemical and pharmacological properties. This genus normally consists of approximately 40 species. Among these, *Marrubium vulgare* L. is a perennial herb found along road sides, and in waste areas.

12.2 Methodology

Data related to *Marrubium vulgare* were collected from published articles using Google Scholar, SciFinder, PubMed, and Science Direct. Books and journals available in library were also consulted while preparing this manuscript. More than 160 articles were studied of which 105 important references are included in this chapter. The present chapter is formulated in such a way so that it becomes easy for researchers to get up to date information on this particular plant, whether related to its phytochemistry or pharmacology.

12.3 Morphological Description

Marrrubium vulgare (Fig. 12.1) height varies from 25 to 45 cm. Leaves of the plants vary from 2.0 to 5.0 cm in length along with very dense surface, mostly sheltered in downy hairs. They also possess blunt and rounded tips. The stems mostly contain branched and woody base. These woody stems are quadrangular and very tightly

Fig. 12.1 Habit and morphology of *Marrubium vulgare* (left side: habit of whole plant; middle: root parts used as medicine; right side: leaves)

covered with hairs. Fruits or nutlets are enclosed by calyx. Seeds are brown or black and vary from 1 to 2.5 mm in length (Halvorson 2003).

12.4 Traditional Uses of *Marrubium vulgare*

Marrubium vulgare is utilized as traditional medicine to treat different types of ailments. Conventionally, the leaves of the plants have been used to get relief from joint pain, chest infection, inflammation, inflamed eyes, cough and cold, and night-time blindness. This plant is also used as a purgative agent and widely used as a bitter tonic and appetizer, and is also helpful in expulsion of fetus (Kirtikar and Basu 1996). It is also used as a herbal tea due to its stimulating and antispasmodic properties (Yamaguchi et al. 2006). It is also used in curing head-pain, and the chemicals in it contain diterpene labdane and marrubiin (Piccoli and Bottini 2008; Karioti et al. 2003). It is also used to treat liver problems and flu (Balme 1982; Grieve 1984; Chevallier 1996; Lorenzi and Matos 2002; Sahpaz et al. 2002a). The leaves and stems are used as antiseptic, cholagogue, antispasmodic, and as stimulant (Grieve 1984; Chiej 1984; Launert 1981; Lust 1983; Mills 1985; Bown 1995). It is often made into a syrup or candy, though it can be used as a tea (Grieve 1984). Due to bitter nature, this plant species is also helpful in digestive functioning of the stomach (Bown 1995; Chevallier 1996). Traditionally, this herb has been considered helpful for the treatment of persistent fever and cholera (Anonymous 2005). *M. vulgare* extracts are also used as flavoring agents in food industries in the USA (Bradley 1992; Vincenzi et al. 1995). The volatile oil produced by *M. vulgare* has a folk status for its use in calming nervous heart. The tiny amount of marrubiin, a labdane diterpene, has the potential to normalize heartbeats. The hot water-extract is used as sweat-inducer, whereas the cold infusion of bitter taste is used as tonic for digestion. The extract of *M. vulgare* is also useful in curing malaria and reducing fever (McIntyre et al. 1988).

The dry flowering stem is used in treating menstrual irregularities and pain. It is also useful in treating sore wounds. The presence of highly volatile constituents makes it the best stimulant and antihelmintic, as well as useful in amenorrhea, chronic rheumatism, dyspepsia, and hepatitis (Singh and Panda 2005; Haq et al. 2011). *M. vulgare* extract also showed appetite stimulant effects through bitter receptors (Janssen et al. 2011). This plant species is also used by local people for candy in the province of United Kingdom. The Egyptians and Romans used plant extracts as the antidote for snake bites. The extract assists to destroy cankerworms when sprayed on fruiting trees. It is also used to ease heartburn, in digestion, and to raze worms in human intestine.

Traditionally, people slowly chewed fresh chopped leaves of *M. vulgare* along with honey to treat cold and sore throat. A candy formulation that contains fresh leaves (four ounces), crushed seeds of cardamom, one teaspoon anise seed, and 250 mL water is used as a standard care for cough in children aged between 5 and 12 years (Barrett 2009). In the Latin American country Brazil, the plant is employed in treating gastrointestinal disorders, inflammation, and respiratory ailments (Meyre Silva et al. 2005; Culpeper 2006). An infusion of leaves is given as an insecticidal (Benedum et al. 2006).

12.5 Phytochemistry

A large number of chemical studies have been carried out on this plant around the world and have confirmed the occurrence of different classes of molecules in *M. vulgare*, which belongs to diterpenes, sesquiterpenes, flavonoids, phenylpropanoid esters, and essential oils (EOs).

12.5.1 Diterpenoids

Various terpenoids (Fig. 12.2) are characterized from aerial parts of *Marrubium vulgare*. The first diterpenoid to be isolated is marrubiin, which was first isolated in 1842; since then, a number of research work has been published regarding isolation, structure elucidation, chemical reaction, stereochemistry, and synthesis of this bioactive compounds (Fulkke et al. 1968; Knoss et al. 1997).

In a separate research, the low-pressure-based chromatographic method was developed for the marrubiin isolation (Knoss et al. 1997). In the year 1992, Rey and co-workers established stereochemistry of marrubiin by X-ray crystallography (Rey et al. 1992). It has been reported that marrubiin has two rings. Some other diterpenoids like marrubenol, marrubiol, peregrinin, premarrubiin, marrubinone B, and vulgarol have also been isolated from aerial parts of *Marrubium vulgare* (El Bardai et al. 2003b; Popa et al. 1968; Sahpaz et al. 2002b; Masoodi et al. 2015; Henderson and McCrindle 1969; Popa and Pasechnik 1975). A novel compound, 11-oxomarrubiin, was identified from methanol extract (ME) of whole plant of *M. vulgare* (Shaheen et al. 2014). Additionally, two new labdane diterpenoids,

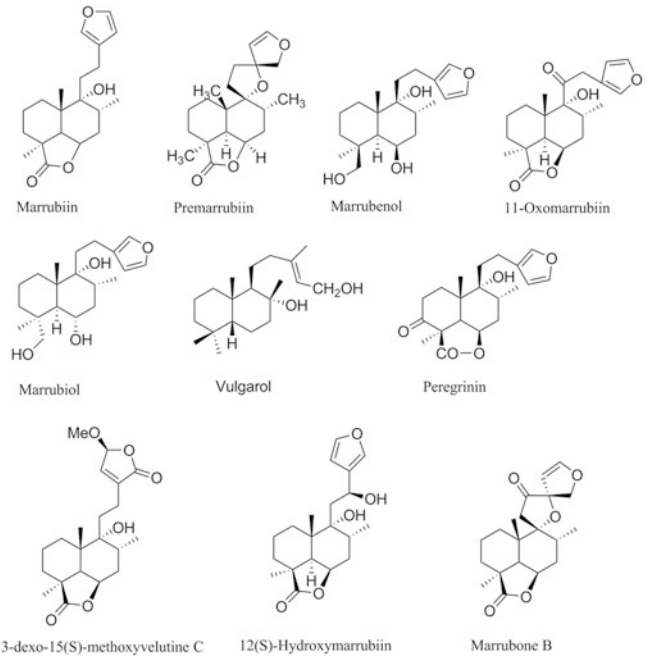

Fig. 12.2 Diterpenoids of *Marrubium vulgare*

3-deoxo-15-methoxyvelutine C and 12(S)-hydroxymarrubiin, were isolated and identified from the extract of methanol from the collection from Srinagar, Jammu and Kashmir (Masoodi et al. 2015).

12.5.2 Flavonoids

Various flavonoids (Fig. 12.3) including aglycones and their glycosides were reported from different plant parts of *M. vulgare*. These include chrysoeriol, vitexin, isoquercitrin, quercetin 3-O-rhamnosyl-glucoside, apigenin 7-lactate, luteolin, apigenin and its 7-(6″-p-coumaroyl)- glucoside, apigenin-7-O-glucoside, luteolin-7-O-β-D-glucoside, and luteolin 7-lactate (Nawwar et al. 1989; Kowalewski and Matlawska 1978; Rahman 2005).

In the year 2014, a flavone derivative 3-hydroxyapigenin-4′-O-(6″-O-p-coumaroyl)-β-D-glucopyranoside was isolated from alcoholic extract of *M. vulgare* (Shaheen et al. 2014). Alkhatib et al. (2010) have reported Ladanein from dichloromethane extract of *M. vulgare*. In another study, a flavone 7-O-β-glucuronyl luteolin was reported for the first time in aerial part of *M. vulgare* along with some well-characterized compounds such as 5,6-dihydroxy-7,4′-dimethoxyflavone (ladanein) and 7-O-β-glucopyranosyl luteolin (Pukalskas et al. 2012).

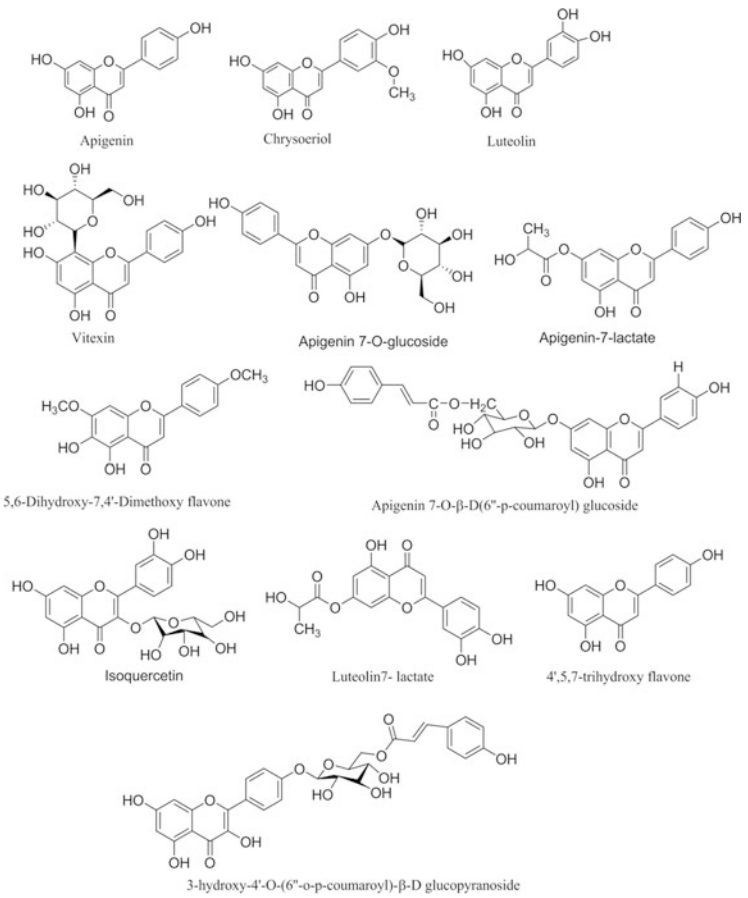

Fig. 12.3 Flavonoids from *Marrubium vulgare*

12.5.3 Phenylpropanoid and Phenylethanoid Glycosides

Several phenylpropanoids (Fig. 12.4) like forsythoside B, (+) (E)-caffeoyl-L-malic acid, ballotetroside, acteoside, and arenarioside were isolated from flowering portion of this species (Sahpaz et al. 2002a).

Two compounds, verbascoside and forsythoside B, were isolated by Pukalskas et al. (2012) from the above-ground portion of this species using gradient solvent of methanol:water:acetic acid (79:20:1). Vulgarcoside A, a diglycoside derivative of the diterpene peregrinol, was isolated from this plant (Pukalskas et al. 2012). Sahpaz and co-workers have reported few phenylethanoid glycosides such as marruboside and acetyl marruboside (Sahpaz et al. 2002a).

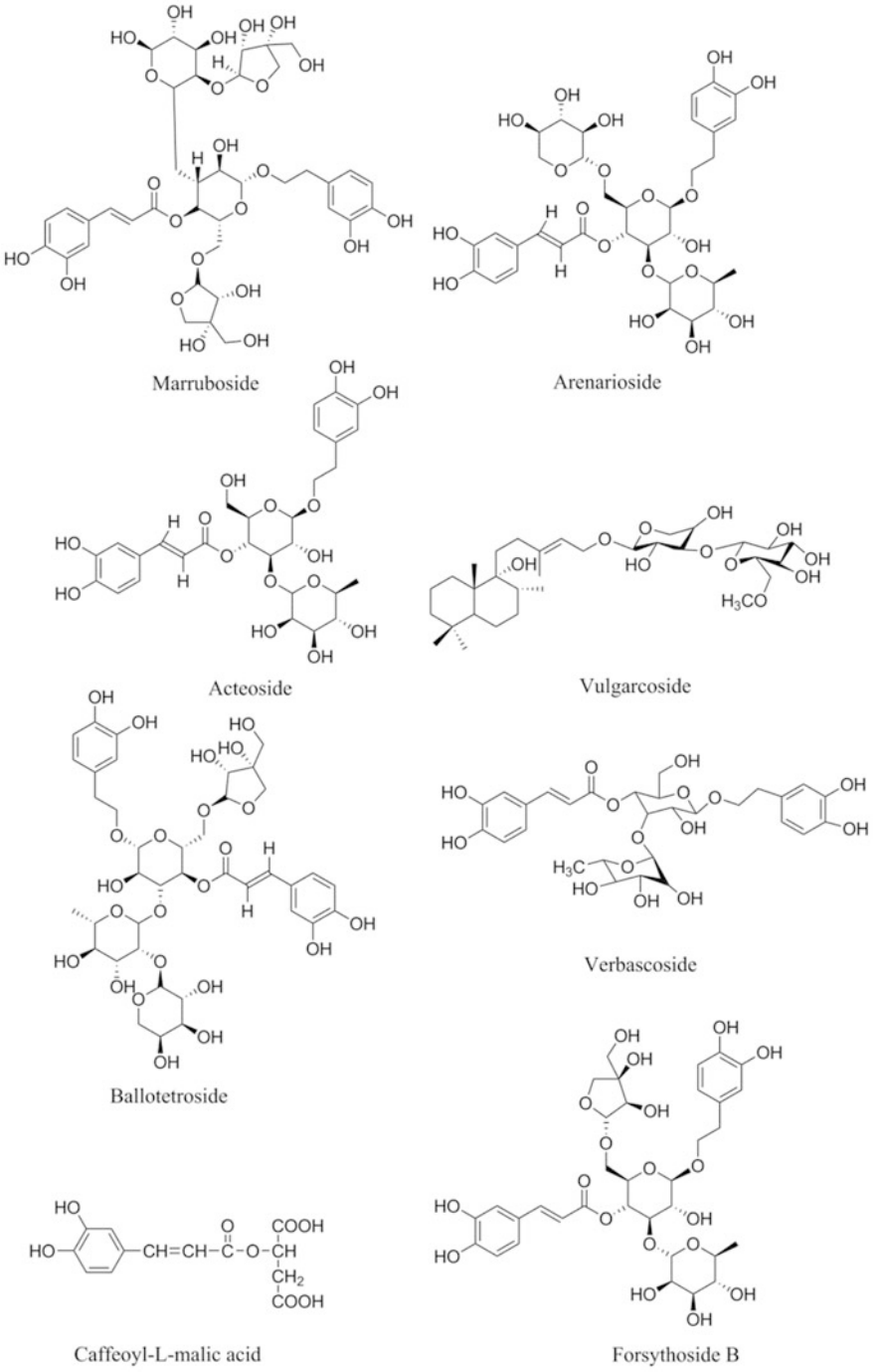

Fig. 12.4 Phenylpropanoid and phenylethanoid glycosides isolated from *Marrubium vulgare*

12.5.4 Active Compounds in Essential Oil of *Marrubium vulgare*

To date, several essential oils present in *M. vulgare* have been studied by different workers across the globe. They include monoterpenes, sesquiterpenes, and esters.

Monoterpenes: Monoterpenes (Fig. 12.5) isolated from *Marrubium vulgare* includes: pinene, tricyclene, thymol, carvacrol, α-pinene, piperitone, 1,8-cineole, sabinene, limonene, p-cymene, α-terpinolene, camphene, p-fenchene, geranial, a-thujone, citronellyl acetate, β-citronellol, and p-menthane-5,6-dihydroxy-3-carboxylic acid, commonly known as marrubic acid (Saleh and Glombitza 1989, Ahmad et al. 2010, Salama et al. 2012, Abadi and Hassani 2013, Zawislak 2012; Kadri et al. 2011).

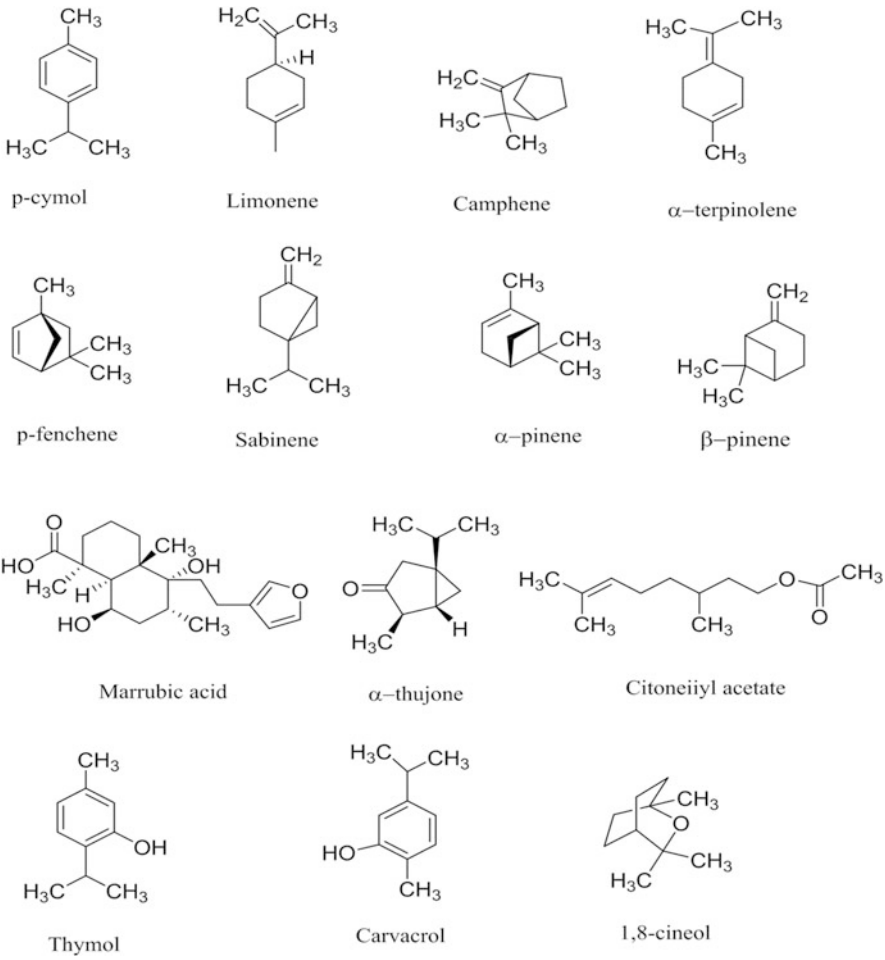

Fig. 12.5 Active constituents under the class monoterpenes of *Marrubium vulgare*

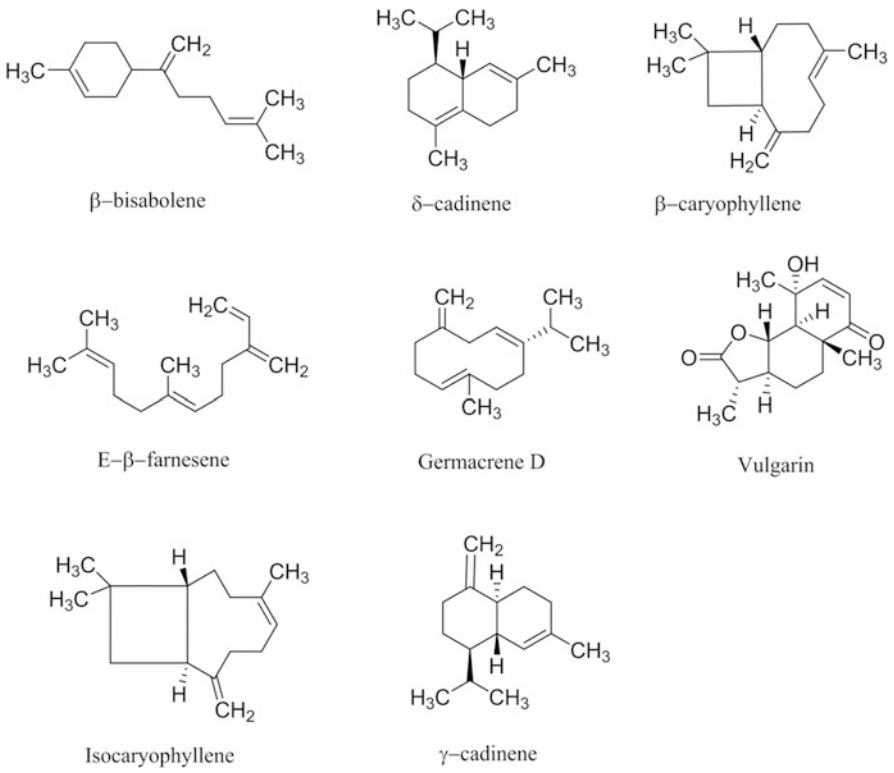

Fig. 12.6 Sesquiterpene in essential oil of *Marrubium vulgare*

Sesquiterpene: From *Marrubium vulgare*, following sesquiterpenes (Fig. 12.6) have been isolated: β-bisabolene, δ-cadinene, isocaryophyllene, γ-cadinene, E-β-farnesene, β-caryophyllene, germacrene D-4-ol, Germacrene D, Vulgarin, γ-eudesmol, ledene, and transcaryophyllene (Salama et al. 2012; Zawislak 2012; Kadri et al. 2011; Al Ahl et al. 2015; Amer 1993).

12.5.5 Other Chemical Constituents

Besides pentacyclic triterpene ursolic acid, two steroids including β-sitosterol and stigmasterol, and phenolic acids such as gallic and caffeic acids, have also been isolated from this species (Nawwar et al. 1989; Laonigro et al. 1979). Small amounts of alkaloids, betonicine and turicine have also been isolated in separate studies (Baxter et al. 1998; Daniel 2006). Other compounds such as 2-(omega-1)-dimethylalkanes, 3-methylalkanes, 2-methylalkanes, and 3-(omega-9)-dimethylalkanes have been reported from the above-ground parts of the plant (Meyresilva and Cechinelfilho 2010).

12.6 Pharmacological Activities

12.6.1 Anti-inflammatory Activity

Genus *Marrubium vulgare* is regularly found in European and Mediterranean countries, hence several researches took place in these regions. In folklore, inflammation and neuro-sedative disorders are treated. The hydro-alcoholic extract (HAE) and methanol extract (ME) were studied separately, which exhibited significant anti-inflammatory activities. The ME at a dosage of 200 mg/kg proved anti-inflammatory function against prostaglandin E_2 and carrageenan-induced inflammation in Swiss mice (Kanyonga et al. 2011). The HAE of *M. vulgare* showed both the in vivo and in vitro anti-inflammatory properties (Abbouyi et al. 2013). The study was assessed by investigating its effect on O_2 consumption and O_2 generation by rat pleural polymorphonuclear leukocytes (PMNs) stimulated with O_2. Five major phenylpropanoid esters, (+) (E)-caffeoyl L-malicacid, acteoside, arenarioside, forsythoside B, and ballotetroside, were isolated, characterized, and their anti-inflammatory activity was studied by Sahpaz et al. (2002a). In another research, ME was assessed on isoproterenol (100 mg/kg/day) induced myocardial infarction (MI) in rat model (Yousefi et al. 2014). It was observed that extract administered orally with dosages of 10, 20, and 40 mg/kg/12 h significantly reduces levels of tumor necrosis factor-α (TNF-α) and peripheral neutrophil count. It was also revealed that the anti-inflammatory activity might be attributed to the presence of marrubiin and other glycosidic phenyl propanoid esters that were isolated from methanolic extract.

12.6.2 Analgesic and Antinociceptive Activities

Marrubium vulgare being utilized in traditional medicine to cure a number of diseases, its hydro-alcoholic extract (aerial parts) showed significant analgesic activity (deSouza et al. 1998). The study clearly proves the analgesic strength with inhibitory dose 50% (ID_{50}) at 22.2 and 272.2 mg/kg for the i.p. and p.o., respectively. It was also discovered that these effects may be due to steroids and terpenes. Meyre-Silva et al. (2005) also recorded that *M. vulgare* had shown strong analgesic properties, which was accredited to marrubiin, a furan labdane-type diterpene. Marrubiinic acid, which was synthesized from marrubiin, and two of its ester derivatives, marrubiinic acid benzyl and methyl esters, showed antinociceptive activity to acetic acid-induced abdominal writhing in mice at doses of 10 mg/kg i.p. and 50 mg/kg orally. Marrubiin was also described to have antinociceptive effect in nociception mice models by DeJesus et al. (2000). In this study, marrubiin was shown to have exhibited dose-dependent antinociceptive effects with 90–900 μmol/kg by p.o. route or 3–90 μmol/kg by i.p. route. The study was completed to assess the antinociceptive effects due to acetic acid-induced, formalin-induced, and capsaicin-induced pains. The results clearly showed that marrubiin is very effective in inhibiting writhing responses (acetic acid-induced) in mice with an ID_{50} value of

2.2–90 μmol/kg, i.p. For formalin-induced responses, the ID_{50} values for i.p. and oral routes were found to be 6.6 μmol/kg and 126 μmol/kg, respectively. For capsaicin-induced inflammation, the ID_{50} value was found to be 28.8 μmol/kg. In 2011, Kanyonga et al. (2011) proved that ME at a dose of 200 mg/kg of *M. vulgare* exhibited the analgesic activity analogous to the acetylsalicylic acid.

12.6.3 Antiodematogenic Activity

Marrubiin being the main constituent of *Marrubium vulgare*, the researchers always try to discover its biological importance. In a study done by Stulzer and co-workers on mice model, marrubiin obtained from this species showed considerable and dose-dependent antiodematogenic effects on micro-vascular leakage in mice ears for different phlogistic agents (Stulzer et al. 2006). The percentage inhibition for histamine at 13.84 mg/kg was found to be 73.7%, for carrageenan at 13.61 mg/kg it was 63.0%, and for bradykinin it was 70% at 18.82 mg/kg. However, dextran yields slight inhibition of 32% as a phlogistic agent.

12.6.4 Antispasmodic Activity

Since *Marrubium vulgare* is used in folk medicine in numerous countries against several diseases, including gastrointestinal disorders, so to prove its biological feature research is being conducted in most of the countries where this species grows naturally. One such study was done in Brazil, where in vitro studies were conducted on aerial parts and roots of *M. vulgare* for antispasmodic effects on numerous smooth muscles, and it was observed that HAE (50% ethanol) exhibits antispasmodic properties in considerable range (Schlemper et al. 1996). Antispasmodic activity of plant extract may be due to the inhibition of neurotransmitters with a reasonable selectivity toward cholinergic contraction. A considerable difference was observed for bradykinin on guinea-pig ileum at 1 mg/mL of extract.

12.6.5 Gastroprotective Activity

In Brazil, *M. vulgare* is used traditionally for the management of respiratory and gastrointestinal infections; so on scientific basis, Paula de Oliveira et al. (2011) evaluated a diterpene marrubiin isolated and characterized from ME of this species for gastroprotective activity. It was observed that ME produced a considerable ulcer-protective effect on alcohol-induced mice model at a dose of 50 and 100 mg/kg, and the result was equivalent to the standard drug omeprazole at 30 mg/kg. In case of indomethacin-induced ulcers, methanolic extract exhibited potential activity at a dose of 50 mg/kg and cimetidine showed similar efficacy at a dose of 100 mg/kg. In both ulcer models, marrubiin (at 25 mg/kg) has shown considerable reduction in gastric parameters.

12.6.6 Antihypertensive Properties

Water extract of *M. vulgare* is extensively used as antihypertensive drug in folk medicine in Morocco. In order to establish its scientific basis, aqueous extract of *M. vulgare* was evaluated in normotensive Wistar-Kyoto rats/spontaneously hypertensive rats (SHRs) (El Bardai et al. 2001). Oral administration of aqueous extract considerably lowered the observed systolic blood pressure (SBP) in SHRs and inhibited the noradrenaline and potassium chloride (100 mM) induced contractile responses of rat aorta in an in vitro study. These results clearly indicate that hypotensive activity exhibited vascular relaxant activity. Vasorelaxant activities of two secondary metabolites marrubenol and marrubiin were reported in a study from aqueous extract and its cyclohexane fraction (El Bardai et al. 2003a). Marrubenol and marrubiin have inhibited contraction in dose-dependent manner in rat aorta. Marrubenol (inhibitory concentration 50% [IC_{50}] values 7.7 ± 1.9 μM) was found somewhat more effective than marrubiin (IC_{50} values 24 ± 2.3 μM). In Mexico, for ethnomedical practices, *M. vulgare* is used as an antihypertensive drug (Jorge et al. 2013).

12.6.7 Antidiabetic and Antihyperlipidemic Activities

Knowing that *Marrubium vulgare* is traditionally used for management of diabetes, a scientific study was conducted to investigate the hypoglycemic effect on alloxan-induced diabetic rats, and results clearly demonstrated that the ethanol extract (EE) of aerial parts of *M. vulgare* showed hypoglycemic effect at a dose of 300 mg/kg with percentage inhibition of 30.3% (Novaes et al. 2001). A similar study was done on alloxan albino Wistar rats for the evaluation of antidiabetic and antihyperlipidemic effects of aqueous extract of *M. vulgare* (Boudjelal et al. 2012). A 50% reduced blood glucose level was observed at a dose of 100 mg/kg, and at 200 and 300 mg/kg, >60% reduction in blood glucose level was observed. In addition, a considerable lowering in total cholesterol, triglyceride, and lipid levels was also observed due to the same extract. These results were found comparable to the market drug glibenclamide.

In the year 2015, a study was conducted in streptozotocin-induced diabetic model to evaluate antidiabetic and antidyslipidemic effects of *M. vulgare* (Elberry et al. 2015). It was observed that methanolic extract of *M. vulgare* considerably lowered the glucose level in blood after treatment for 14 days at a single dose of 500 mg/kg/day. Furthermore, methanolic extract also showed a significant increase in tissue glycone and plasma insulin. A rare fatty acid, 6-octadecynoic acid (6-ODA) was identified in methanol extract of *M. vulgare* and reported to function as PPARγ α agonist (Ohtera et al. 2013). In another study, effectiveness of plant extract of *M. vulgare* was investigated in cyclosporine A and streptozotocin-induced diabetes mellitus-type 1 in mice (Maraia 2014). Thus, this species was recommended as useful in curing auto-immune diabetes.

12.6.8 Antihepatotoxic Activity

Horehound is used in herbal medicine of Saudi Arabia for the treatment of gastroenteric, respiratory, and inflammatory disorders. In Saudi Arabia, a study was performed to investigate the antihepatotoxic activity of methanolic extract against carbon tetrachloride (CCl_4) induced hepatic damage in rats (Elberry et al. 2010). The observation exposed that methanolic extract (500 mg/kg/day) significantly reduced the levels of lactate dehydrogenase (LDH), aspartate transaminase (AST), and alanine transaminase (ALT), suggesting antihepatotoxic effect of *M. vulgare*. This effect may be attributed partially to the antioxidant activity of the extract. CCl_4-induced hepatotoxicity in albino mice was also studied by Ibrahim et al. (2014). The effects of aqueous, ethanol, and petroleum ether extracts from aerial parts of *M. vulgare* were also evaluated. The results showed that ethanol extract exhibited strong protection against the damage caused by carbon tetrachloride (CCl_4).

In another study, first time 12 active compounds isolated from *M. vulgare* were evaluated for their drug-likeness and hepatoprotective activity in a computer-based in-silico model (Verma et al. 2012). Among the 12 compounds, vulgarin exhibited significant antihepatotoxic activity against CCl_4-induced toxicity in Wistar rat model. Akther et al. (2013) also reported hepatoprotective activity of methanolic extract of whole plant of *M. vulgare* against paracetamol-induced toxicity in Wistar rats (Akther et al. 2013).

12.6.9 Antioxidant Activity

Majority of the Mexican people make teas from leaflets, while roots of some medicinal plants are used to treat various ailments including infections, arthritis, heart disorders, headaches, fever, asthma, and menstrual pain. So, in a study, over 30 medicinal plants were investigated for evaluation of antioxidants (VanderJagt et al. 2002). Among them, it was reported that aqueous extract of leaves of *M. vulgare* contained about 560 μmol/g Trolox equivalent/g dry weight. In another study methanol extract of leaves showed strong antioxidant activity against 2,2-diphenyl-1-picrylhydrazyl (DPPH; IC_{50} = 35 μg/mL) radical scavenging (Chedia et al. 2014). Antioxidant activities of acetone extract (AE), deodorized acetone extract (DAE), and deodorized water extract (DWE) from leaves of *M. vulgare* were tested at 80 °C in rapeseed (*Brassica napus*) oil (Weel et al. 1999) and found that acetone extract was better as compared with water extract.

Essential oils (EOs) are considered to be naturally occurring antioxidants, so various researches were performed on the essential oils obtained from *M. vulgare*. In one such research done in Tunisia, it was reported that the antioxidant effects of aerial parts of *M. vulgare* were produced through their essential oils probably due to the hydroxylated and oxygen-containing compounds (Kadri et al. 2011). Based on the results obtained from DPPH, β-carotene bleaching test, and reducing power assay, it was concluded that EOs obtained from *M. vulgare* can be used as a natural

food preservative and to improve human health as a natural antioxidant. Comparing the antioxidant activity with synthetic butylated hydroxyanisole (BHT), it was revealed that EOs exhibited an IC_{50} value of 153.84 μg/mL, which was two times greater than BHT (Abadi and Abdellatif 2013). In a study, the antioxidant activities of five compounds (5–6-dihydroxy,7–4′-dimethoxy flavone, 7-O-β-glucopyranosyl luteolin, 7-O-β-glucuronyl luteolin, verbascoside, and forsythoside B) isolated from aerial parts of *M. vulgare* were examined using DPPH and $ABTS^+$ free radical scavenging assays, and compared with rosmarinic acid (Pukalskas et al. 2012). The effects of forsythoside B and verbascoside were found comparable to that of natural rosmarinic acid in $ABTS^+$ assay. Among the fractions, the *n*-butanol fraction from which four compounds, except 5–6-dihydroxy,7–4′-dimethoxy flavone, were isolated exhibited highest antioxidant activity in the β-carotene bleaching assay and linolenic acid model. There was another study in which antioxidant capacity was again found in methanol leaf extract of *Marrubium vulgare* (Amessis-Ouchemoukh et al. 2014). In this study, antioxidant capacities of methanol and acetone extracts were evaluated using DPPH, H_2O_2, total antioxidant capacity, and iron-reducing power assays. The percentage of DPPH radical scavenging activity of methanol extract in the range of 51.90–97.15% suggests its importance as a natural preservative and in the prevention of oxidative stress-related disorders.

12.6.10 Antimicrobial Activity

Various studies have been done for the evaluation of antimicrobial activities of *Maruubium vulgare*, the most widely used herb as medicine in Arab counties. Ethanol extracts (EEs) from leaves and flowers of *M. vulgare* were investigated for their antimicrobial potential using rapid colorimetry (XTT) and viable count method (Al-Bakri and Afifi 2007). The promising antimicrobial activity shown by EE of *M. vulgare* was against *Bacillus subtilis* and *Staphylococcus aureus*, while against *Escherichia coli* and *Pseudomonas aeruginosa* it was very weak. Promising activity was also exhibited against *S. aureus*, *Staphylococcus epidermidis*, and *B. subtilis* when methanolic extract of *M. vulgare* whole plant was evaluated for its antimicrobial activity (Masoodi et al. 2008). With regard to other two strains, that is, *P. vulgaris* and *E. coli*, there was a moderate effect. In this study, six bacterial organisms were used and ciprofloxacin was used as a standard drug. In another study, the *M. vulgare* roots were extracted with ethanol and inhibition assay of biofilm formation was done in methicillin-resistant *S. aureus* (MRSA; Quave and Smeltzer 2009). The minimum inhibitory concentration (MIC) was determined after 18 h growth using broth microtiter dilution method. A significant inhibition of biofilm formation (IC_{50} = 32 μg/mL) and adherence (IC_{50} = 8 μg/mL) was established in extract of roots at dose-dependent manner. A remarkable antimicrobial activity against *Staphylococcus aureus* and *S. epidermidis* was also shown by EE of *M. vulgare* and that study was done in Turkey in which 22 plant species were investigated (Kunduhoglu et al. 2011). The study was done on ethanol, acetone, and ether extracts obtained from leaves, flowers, and stems of 22 plant species.

Helicobacter pylori bacterium is considered the major cause of infection in gastric disorders; a study was undertaken in which anti-*Helicobacter pylori* activity was investigated (Robles-Zepeda et al. 2011). In the study, methanol extract at a concentration of 10 mg/mL of *M. vulgare* was evaluated using broth micro-dilution method and MIC was observed at 800 μg/mL. In another study, essential oil of *M. vulgare* was evaluated for antimicrobial activity and MIC was determined. (Zarai et al. 2011). In yet another study, essential oil was investigated against series of bacterial and fungal strains by agar disk diffusion method. The results showed a considerable antimicrobial activity against Gram positive bacteria with MIC values in the range of 1.12–2.6 mg/mL by zone inhibition assay. However, Gram negative bacteria exhibited resistance against tested sample. MIC was found in the range of 12.5–25 mg/mL in a study where methanolic extract of *M. vulgare* was evaluated for antibacterial activity against seven pathogenic bacteria (Chedia et al. 2014). In this study, total phenolic content was also determined and it was concluded that antibacterial activity was due to high content of phenolic compounds in the extract. Tuberculosis, one of the serious health problems, has developed resistance to some of the antibiotics, which has enforced researchers to create or discover new drugs.

EOs are considered naturally occurring antioxidants and were evaluated for antibacterial activity against different Gram positive and Gram negative pathogens. In one study, EOs showed varied antibacterial activity against *Listeria monocytogenes*, *P. aeruginosa*, *Agrobacterium tumefaciens*, and *Salmonella enterica* (Abadi and Abdellatif 2013). By Mueller-Hinton broth dilution method, the MIC values were obtained, which were in the range of 0.1–15 μg/mL. In another study, the essential oils and ethanol extract prepared from the leaves of *M. vulgare* were studied for antimicrobial activity against 17 clinical strains of *Staphylococcus aureus* (Bokaeian et al. 2014). Minimum MIC value for the extract was found to be 2.5 mg/mL and maximum MIC value for essential oils was found to be 2.5 mg/mL. This study confirmed the use of essential oils and ethanol extract of *M. vulgare* as antibacterial agents.

12.6.11 Anticancer Activity

In recent years, the search for naturally occurring anticancer chemotherapeutic drugs continued at a great speed and research focus on *M. vulgare* in this field is no way behind. Yamaguchi and co-workers (2006) reported antiproliferative effect on colon cancer cells with leaf extract. In this study, methanol extracts upregulated gene (NAG-1) through a trans-activation of the NAG-1 promoter at a dose of 250 μg/mL. This was for the first time that a major compound ladanein isolated from *M. vulgare* was studied for its cytotoxic activity against drug dasatinib-resistant murine leukemia cell line (DA1-3b/M2$^{BCR\text{-}ABL}$). In the study, ladanein showed nonsignificant (20–40 μM) activity against series of cancer cell lines, including K562, K562R, and 697 human leukemia cell lines, but was found inactive to MOLM13 and human peripheral blood mononuclear cells (Alkhatib et al. 2010). In vitro cytotoxicity of *M. vulgare* against cancer cell lines was studied in Morocco,

Tunisia, and Dubai. In one of the studies, essential oils obtained from *M. vulgare* were examined using a modified MTT assay for their cytotoxic activity against HeLa cell lines (Zarai et al. 2011). The results depicted that essential oils destroyed HeLa cells by 27% in a concentration of 0.25 mg/mL and 100% cells died at >0.5 mg/mL concentration. A considerable cytotoxic effect with IC_{50} at 258 mg/mL against tumor cells was also observed. In another study, alcoholic leaves extract and isolated phenolic compounds, viz., acacetin, apigenin, and its glycoside, were tested against U251 (Ehrlich tumor cell lines) and breast cancer line (MCF7). Alcoholic extract and its secondary metabolites showed significant anticancer activity against U251 with effective dose for 50% (ED_{50}) < 20 μg/mL whereas extract and acacetin showed moderate activity against MCF7 with ED_{50} > 20 μg/mL (Nawal and Atta 2013). Different extracts including methanol, hexane, dichloromethane, and ethyl acetate obtained from aerial parts of *M. vulgare* were evaluated for cytotoxic activity against tumor cell lines (Belayachi et al. 2013). The results showed that dichloromethane extract of *M. vulgare* was effective against colorectal cancer cell lines (SW620 cells), acute T-cell leukemia, and mantle cell lymphoma cell line with IC_{50} at 30 μg/mL.

12.6.12 Antiprotozoal, Molluscicidal, and Mosquitocidal Activities

Marrubium vulgare is also used for treating intestinal disorders. In two separate studies, *M. vulgare* was reported for antiprotozoal property. In one study, acetone and methanolic extracts of *M. vulgare* were found to have activity against *Entamoeba histolytica* and *Giardia lamblia* with growth inhibition $IC_{50} = 7$ at a dose of 12 μg/mL and $IC_{50} = 90$ at 34 μg/mL (Ramos-Guerra et al. 2007). Methanolic extracts of this plant species were reported to be effective against *Trypanosoma cruzi* with percentage growth inhibition between 88% and 100% at a concentration of 150 μg/mL (Molina-Garza et al. 2014).

Regarding molluscicidal and mosquitocidal activities, the volatile oils of *M. vulgare* were used to evaluate their activities on eggs of *Biomphalaria alexandrina* and *Culex pipiens* (Salama et al. 2012). The result clearly showed 100% ovicidal activity at 200 ppm/24 h. Another study stated that methanol extract of *M. vulgare* leaves are very effective against fourth instar larvae of the mosquito *C. pipiens* L. (Amel and Selima 2015).

12.7 Conclusion and Future Perspectives

The traditional uses, phytochemistry, and pharmacology of *Marrubium vulgare* are presented here in this chapter. Regarding chemical constituents, diterpenes, flavonoids, and phenylpropanoids are the major ones identified. Marrubiin is one of the major labdane diterpenes and exists in higher concentration. The pharmacological activities of *M. vulgare* proved its potential for the development of new efficacious botanicals in future. It is evident from the literature that mostly methanolic extracts of leaves and aerial parts have been studied in detail; however,

no substantial work has been carried out, so there is a need to work over other plant parts (root, stem, flowers, seeds) along with different extracts and their fractions. Though the plant contains different classes of secondary metabolites, but only marrubiin, marrubenol, and ladanein were well studied and hence other compounds need to be investigated. Clinical trials should be initiated for this plant, which is crucial for the diagnosis of herbal toxicity and development of plant-based new drugs.

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References

Abadi A, Abdellatif F (2013) Antibacterial and antioxidant activities of *Marrubium vulgare* essential oil cultivated in Eastern Algeria. Int J Chem Stud 1:32–38

Abadi A, Hassani A (2013) Chemical composition of *Marrubium vulgare* L. essential oil from Algeria. Int Lett Chem Phys Astron 13:210–214

Abbouyi AE, Khyari S, Eddoha R, Filali Ansari N (2013) Anti-inflammatory effect of hydromethanolic extract from *Marrubium vulgare* lamiaceae on leukocytes oxidative metabolism: an in vitro and in vivo studies. Int J Green Pharm 7:224–229

Ahmad B, Masoodi MH, Siddique AH, Khan SA (2010) A new monoterpene acid from *Marrubium vulgare* with potential antihepatotoxic activity. Nat Prod Res 24:1671–1680

Akther N, Shawla AS, Sultanab S, Chandanc BK, Akhter M (2013) Hepatoprotective activity of *Marrubium vulgare* against paracetamol induced toxicity. J Pharm Res 7:565–570

Al Ahl HA, Gendy ASH, Mahmoud AA, Mohamed HF (2015) Essential oil composition of *Marrubium vulgare* L. cultivated in Egypt. Int J Plant Sci Ecol 4:138–141

Al-Bakri AG, Afifi FU (2007) Evaluation of antimicrobial activity of selected plant extracts by rapid XTT colorimetry and bacterial enumeration. J Microbiol Methods 68:19–25

Alkhatib R, Joha S, Cheok M, Roumy V, Idziorek T, Preudhomme C, Quesnel B, Sahpaz S, Bailleul F, Hennebelle T (2010) Activity of ladanein on leukemia cell lines and its occurrence in *Marrubium vulgare*. Planta Med 76:86–87

Amel A, Selima B (2015) Larvicidal effect of *Marrubium vulgare* on Culex pipiens in eastern Algeria. Energy Procedia 74:1026–1031

Amer MMA (1993) Constituents of the aerial parts of *Marrubium vulgare* L. Mansoura J Pharm Sci 9:92–98

Amessis-Ouchemoukh N, Abu-Reidah IM, Quirantes-Pine R, Madani K, Segura-Carretero A (2014) Phytochemical profiling, in vitro evaluation of total phenolic contents and antioxidant properties of *Marrubium vulgare* (horehound) leaves of plant growing in Algeria. Ind Crop Prod 61:120–129

Anonymous (2005) The wealth of India, a dictionary of Indian raw materials and industrial product. Publication and Information Directorate, New Delhi

Balme F (1982) Plantas Medicinais. Hemus Sao, Paulo

Barrett J (2009) What can I do with my herbs: how to grow, use, and enjoy these versatile plants. A&M University Press, College Station

Baxter H, Harborne JB, Moss GP (1998) Phytochemical dictionary: a handbook of bioactive compounds from plants. CRC Press, London, p 79

Belayachi L, Aceves-Luquero C, Merghoub N, Bakri Y, Fernandez de Mattos S, Amzazi S, Villalonga P (2013) Screening of North African medicinal plant extracts for cytotoxic activity against tumor cell lines. Eur J Med Plants 3:310–332

Benedum J, Loew D, Schilcher H (2006) Medicinal plants in traditional medicine. Kooperation Phytopharmaka, Bonn, p 136

Bokaeian M, Saboori E, Saeidi S, Niazi AA, Amini N, Khaje H, Bazi S (2014) Phytochemical analysis, antibacterial activity of *Marrubium vulgare* L. against *Staphylococcus aureus* in vitro. Zahedan J Res Med Sci 16:60–64

Boudjelal A, Henchiri C, Siracusa L, Sari M, Ruberto G (2012) Compositional analysis and in vivo anti-diabetic activity of wild Algerian Marrubium vulgare L. infusion. Fitoterapia 83:286–292

Bown D (1995) Encyclopaedia of herbs and their uses. Dorling Kindersley, London. ISBN 0-7513-020-31

Bradley PR (1992) British herbal compendium, White Horehound-Marrubii Herba, vol 1. British Herbal Medicine Association, Bournemouth

Brauchler C, Meimberg H, Heubl G (2010) Molecular phylogeny of Menthinae (Lamiaceae, Nepetoideae, Mentheae) taxonomy, biogeography and conflicts. Mol Phylogenet Evol 55:501–523

Chedia A, Ghazghazi H, Brahim H, Abderrazak M (2014) Total phenolic content, antioxidant and antibacterial activities of *Marrubium vulgare* methanolic extract. Tunis J Med Plant Nat Prod 11:1–8

Chevallier A (1996) The encyclopaedia of medicinal plants. Dorling Kindersley, London. ISBN 9-780751-303148

Chiej R (1984) Encyclopaedia of medicinal plants. MacDonald ISBN 0-356-10541-5

Culpeper N (2006) Culpeper's complete herbal and English physician, illustrated reprint. Published by Apple Wood Books, London, pp 96–97

Daniel M (2006) Medicinal plants: chemistry and properties. CRC Press, Boca Raton, p 67

DeJesus RA, Cechinel Filho V, Oliveira AE, Schlemper V (2000) Analysis of the antinociceptive properties of marrubiin isolated from *Marrubium vulgare*. Phytomedicine 7:111–115

deSouza MM, de Jesus RA, Cechinel-Filho V, Schlemper V (1998) Analgesic profile of hydroalcoholic extract obtained from Marrubium vulgare. Phytomedicine 5:103–107

El Bardai S, Lyoussi B, Wibo M, Morel N (2001) Pharmacological evidence of hypotensive activity of *Marrubium vulgare* and *Foeniculum vulgare* in spontaneously hypertensive rat. Clin Exp Hypertens 23:329–343

El Bardai S, Morel N, Wibo M, Fabre N, Llabres G, Lyoussi B, Quetin-Leclercq Q (2003a) The vasorelaxant activity of marrubenol and marrubiin from *Marrubium vulgare*. Planta Med 69:75–77

El Bardai S, Wibo M, Hamaide MC, Lyoussi B, Quetin Leclercq J, Morel N (2003b) Characterization of marrubenol, a diterpene extracted *from Marrubium vulgare*, as an L-type calcium channel blocker. Br J Pharmacol 140:1211–1216

Elberry AA, Harraz FM, Ghareib SA, Nagy AA, Gabr SA, Suliaman MI, Sattar EA (2010) Antihepatotoxic effect of *Marrubium vulgare* and *Withania somnifera* extracts on carbon tetrachloride-induced hepatotoxicity in rats. J Basic Clin Pharm 1:247–254

Elberry AA, Harraz FM, Ghareib SA, Nagy AA, Sattar EA (2015) Methanolic extract of *Marrubium vulgare* ameliorates hyperglycemia and dyslipidemia in streptozotocin-induced diabetic rats. Int J Diabetes Mellit 3:37–44

Fulkke JWB, Henderson MS, McCrindle R (1968) Some reactions of the diterpene marrubiin and its congeners. J Chem Soc C:807–810

Giuliani C, Bini ML (2008) Insight into the structure and chemistry of glandular trichomes of Labiatae, with emphasis on subfamily Lamioideae. Plant Syst Evol 276:199–208

Grieve A (1984) Modern herbal. Penguin ISBN 0-14-046-440-9

Gurib-Fakim A (2006) Medicinal plants: traditions of yesterday and drugs of tomorrow. Mol Aspects Med 27:1–93

Halvorson WLGP (2003) USGS weeds in the west project: status of introduced plants in southern Arizona Parks, Factsheet for *Marrubium vulgare* L., U.S.G.S.N.P. Service., US

Haq F, Ahmad H, Alam M (2011) Traditional uses of medicinal plants of Nandiar Khuwarr catchment. J Med Plant Res 5:39–48

Henderson MS, McCrindle R (1969) Premarrubiin: a diterpenoid from *Marrubium vulgare* L. J Chem Soc C Org 15:2014–2015

Ibrahim FM, Ibrahim AY, Omer EA (2014) Potential effect of *Marrubium vulgare* L. extracts on CCL4 model induced hepatotoxicity in Albino mice. World J Pharma Sci 2:1664–1670

Janssen S, Laermans J, Verhulst PJ, Thijs T, Tack J, Depoortere I (2011) Bitter taste receptors and gustducin regulate the secretion of ghrelin with functional effects on food intake and gastric emptying. Proc Natl Acad Sci U S A 108:2094–2099

Jorge VG, Melina HG, Joaquin HC, Patricia CE, Emmanuel RM, Marisa EC, Samuel ES, Angel SO, Emmanuel HN (2013) Vasorelaxant effect of ethanolic extracts from *M. vulgare*: Mexican medicinal plant as potential source for bioactive molecules isolation. Indo Global J Pharm Sci 3:1–5

Kadri A, Zarai Z, Bekira A, Gharsallah N, Damak M, Gdoura R (2011) Chemical composition and antioxidant activity of *Marrubium vulgare* L. essential oil from Tunisia. Afr J Biotechnol 10:3908–3914

Kanyonga PM, Faouzi MA, Meddah B, Mpona M, Essassi EM, Cherrah Y (2011) Assessment of methanolic extract of *Marrubium vulgare* for anti-inflammatory, analgesic and anti-microbiologic activities. J Chem Pharm Res 3:199–204

Karioti A, Skaltsa H, Heilmann J, Sticher O (2003) Acylated flavonoid and phenylethanoid glycosides from *Marrubium velutim*. Phytochemistry 64:655–660

Kirtikar KR, Basu BD (1996) Indian medicinal plants. International Book Distributors, Dehradun

Knoss W, Reuter B, Zapp J (1997) Biosynthesis of the labdane diterpene marrubiin in Marrubium vulgare *via* a non-mevalonate pathway. Biochem J 326:449–454

Kowalewski Z, Matlawska I (1978) Flavonoid compounds in the herb *Marrubium vulgare*. Herba Polonica 24:183–186

Kunduhoglu B, Pilatin S, Caliskan F (2011) Antimicrobial screening of some medicinal plants collected from Eskisehir, Turkey. Fresen Environ Bull 20:945–952

Laonigro G, Lanzetta R, Parrilli M, Adinolfi M, Mangoni L (1979) The configuration of the diterpene spiro ethers from *Marrubium vulgare* and from *Leonotis leonurus*. Gazzetta Chimica Italiana 109:145–150

Launert E (1981) Edible and medicinal plants. Hamlyn ISBN 0-600-37216-2

Lorenzi H, Matos FJA (2002) Plantas medicinais do Brasil: nativas e exoticas. Instituto Plantarum, Nova Odessa

Lust J (1983) The herb book. Bentam books ISBN 0-553-23827-2

Maraia FE (2014) Hypoglycemic effects of Marrubium vulgare (Rubia) in experimentally induced autoimmune diabetes mellitus. Int Res J Biochem Bioinform 4:42–54

Masoodi MH, Ahmed B, Zargar IM, Khan SA, Khan S, Singh P (2008) Antibacterial activity of whole plant extract of *Marrubium vulgare*. Afr J Biotechnol 7:86–87

Masoodi M, Ali Z, Liang S, Yin H, Wang W, Khan IA (2015) Labdane diterpenoids from Marrubium vulgare. Phytochem Lett 13:175–279

McIntyre A, Mabey R, McIntyre M (1988) The new age herbalist: how to use herbs for healing, nutrition, body care, and relaxation. Simon and Schuster, New York

Meyre Silva C, Yunes RA, Schlemper V, Campos Buzzi F, Cechinel Filho V (2005) Analgesic potential of marrubiin derivatives, a bioactive diterpene present in *Marrubium vulgare* (Lamiaceae). Farmacoterapia 60:321–326

Meyre Silva C, Cechinel Filho V (2010) A review of the chemical and pharmacological aspects of the genus Marrubium. Curr Pharm Des 16:3503–3518

Mills SY (1985) The dictionary of modern herbalism. Simon Mills

Molina-Garza ZJ, Bazaldua-Rodríguez AF, Quintanilla-Licea R, Galaviz-Silva L (2014) Anti-Trypanosoma cruzi activity of 10 medicinal plants used in Northeast Mexico. Acta Trop 136:14–18

Nawal HM, Atta EM (2013) Cytotoxic and antioxidant activity of *Marrubium vulgare* and its flavonoid constituents, In: 2nd International Conference on Chemical, Environmental and Biological Sciences (ICCEBS'2013). UAE, Dubai, pp 40–42

Nawwar MA, El Mousallamy AM, Barakat HH, Buddrus J, Linscheid M (1989) Flavonoid lactates from leaves of Marrubium vulgare. Phytochemistry 28:3201–3206

Novaes AP, Rossi C, Poffo C, Pretti Júnior E, Oliveira AE, Schlemper V, Niero R, Cechinel-Filho V, Burger C (2001) Preliminary evaluation of the hypoglycemic effect of some Brazilian medicinal plants. Therapie 56:427–430

Ohtera A, Miyamae Y, Nakai N, Kawachi A, Kawada K, Han J, Isoda H, Neffati M, Akita T, Maejima K, Masuda D, Kambe T, Mori N, Irie K, Nagao M (2013) Identification of 6-octadecynoic acid from a methanol extract of *Marrubium vulgare* L. as a peroxisome proliferator-activated receptor agonist. Biochem Biophys Res Commun 440:204–209

Paula de Oliveira A, Santin JR, Lemos M, Klein Júnior LC, Couto AG, Meyre da Silva Bittencourt C, Cechinel Filho V, Falonide de Andrade S (2011) Gastroprotective activity of methanol extract and marrubiin obtained from leaves of *Marrubium vulgare* L. (Lamiaceae). J Pharm Pharmacol 63:1230–1237

Piccoli PN, Bottini R (2008) Accumulation of the labdane diterpene marrubiin in glandular trichome cells along the ontogeny of *Marrubium vulgare* plants. Plant Growth Regul 56:71–76

Popa DP, Pasechnik GS (1975) Structure of vulgarol-new diterpenoid from *Marrubium vulgare*. Chem Nat Compd 11:752–756

Popa DP, Pasechnik GS, Thuc Anh P (1968) Marrubiol, a new diterpenoid from *Marrubium vulgare*. Chem Nat Compd 4:291–293

Pukalskas A, Venskutonis PR, Salido S, Waard P, Beek TA (2012) Isolation, identification and activity of natural antioxidants from horehound (*Marrubium vulgare* L.) cultivated in Lithuania. Food Chem 130:695–701

Quave CL, Smeltzer M (2009) Anti-biofilm activity of *Marrubium vulgare* L. (Lamiaceae) extract on MRSA. Planta Med 75:96

Rahman A (2005) Studies in natural products chemistry. Bioactive Nat Prod:266. Elsevier Pakistan

Ramos-Guerra MC, Mata-Cárdenas BD, Vargas-Villarreal J, Sampayo-Reyes A, González-Salazar F, Morales-Vallarta M, Said-Fernandez S (2007) In vitro activity of organic leaf/stem extracts from *Marrubium vulgare* and *Mentha spicata* against *Entamoeba histolytica* and *Giardia lamblia*. Pharmacologyonline 1:108–112

Rey JP, Levesque J, Pousset JL (1992) Extraction and high-performance liquid chromatographic methods for the lactones parthenolide (*Chrysanthemum parthenium* Bernh.), marrubiin (*Marrubium vulgare* L.) and artemisinin (*Artemisia annua* L.). J Chromatogr 605:124–128

Robles-Zepeda RE, Velázquez-Contreras CA, Garibay-Escobar A, Gálvez-Ruiz JC, Ruiz-Bustos E (2011) Antimicrobial activity of Northwestern Mexican plants against *Helicobacter pylori*. J Med Food 14:1280–1283

Sahpaz S, Garbacki N, Tits M, Bailleul F (2002a) Isolation and pharmacological activity of phenylpropanoid esters from *Marrubium vulgare*. J Ethnopharmacol 79:389–392

Sahpaz S, Hennebelle T, Bailleul F (2002b) Marruboside, a new phenylethanoid glycoside from *Marrubium vulgare* L. Nat Prod Lett 16:195–199

Salama MM, Taher EE, El Bahy MM (2012) Molluscicidal and Mosquitocidal activities of the essential oils of *Thymus capitatus* Hoff. et Link. and *Marrubium vulgare* L. Revista do Instituto de Medicine Tropicalde Sao Paulo 54:281–286

Saleh MM, Glombitza KW (1989) Volatile oil of *Marrubium vulgare* and its anti-schistosomal activity. Planta Med 55:105–108

Schlemper V, Ribas A, Nicolau M, Cechinel Filho V (1996) Antispasmodic effects of hydroalcoholic extract of *Marrubium vulgare* on isolated tissues. Phytomedicine 3:211–216

Shaheen F, Rasoola S, Shah ZA, Soomro S, Jabeen A, Mesaik MA, Choudhary MI (2014) Chemical constituents of *Marrubium vulgare* as potential inhibitors of nitric oxide and respiratory burst. Nat Prod Commun 9:903–906

Singh MP, Panda H (2005) Medicinal herb with their formulations. Daya Publishing House, New Delhi

Stulzer HK, Tagliari MP, Zampirolo JA, Cechinel Filho V, Schlemper V (2006) Antioedematogenic effect of marrubiin obtained from *Marrubium vulgare*. J Ethnopharmacol 108:379–384

Sullivan K, Shealy CN (1997) Complete natural home remedies. Afr J Pharm Pharmacol 3:621–625

VanderJagt TJ, Ghattas R, VanderJagt DJ, Crossey M, Glew RH (2002) Comparison of the total antioxidant content of 30 widely used medicinal plants of New Mexico. Life Sci 70:1035–1040

Verma A, Masoodi M, Ahmed B (2012) Lead finding from whole plant of *Marrubium vulgare* L. with hepatoprotective potentials through in silico methods. Asian Pac J Trop Biomed 2:1308–1311

Vincenzi M, Maialetti F, Dessi MR (1995) Monographs on botanical flavouring substances used in foods. Fitoterapia 66:203–310

Weel KG, Venskutonis PR, Pukalskas A, Gruzdiene D, Linssen JP (1999) Antioxidant activity of horehound (*Marrubium vulgare* L.) grown in Lithuania. Eur J Lipid Sci Technol 101:395–400

Wink M (2003) Evolution of secondary metabolites from an ecological and molecular phylogenetic perspective. Phytochemistry 64:3–19

Yamaguchi K, Liggett JL, Kim NC, Baek SJ (2006) Anti-proliferative effect of horehound leaf and wild cherry bark extracts on human colorectal cancer cells. Oncol Rep 15:275–281

Yousefi K, Fathiazad F, Soraya H, Rameshrad M, Maleki Dizaji N, Garjani A (2014) *Marrubium vulgare* L. methanolic extract inhibits inflammatory response and prevents cardiomyocyte fibrosis in isoproterenol- induced acute myocardial infarction in rats. Bioimpacts 4:21–27

Zarai Z, Kadri A, Ben Chobba I, Ben Mansour R, Bekir A, Mejdoub H, Gharsallah N (2011) The in-vitro evaluation of antibacterial, antifungal and cytotoxic properties of *Marrubium vulgare* L. essential oil grown in Tunisia. Lipids Health Dis 10:161

Zawislak G (2012) Chemical composition of essential oils of *Marrubium vulgare* L. and *Marrubium incanum* Desr. Grown in Poland. Chemija 23:136–140