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Essential Oils for Pest Control

Leo M.L. Nollet, Hamir Singh Rathore

Basil (Ocimum basilicum L.) Oil: As a Green Pesticide

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Basil (*Ocimum basilicum* L.) Oil: As a Green Pesticide

N.C. Basantia

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5.1 Introduction

Basil (*Ocimum basilicum*) is one of the most popular herbs grown in the world. The name *basil* is thought to be derived from the Greek words *okimon* ("smell") and *basilikon* ("royal or king") (Selvakkumar et al. 2007). It is often referred to as the king of the herbs. Basil originated in Asia and Africa. In Hindu houses, basil is used to protect the family from evil spirits. In early 1600, the English used basil in their food and doorways to ward off uninvited pests, such as flies, as well as evil spirits. Several interesting beliefs are ascribed to the historical use of basil. Some Europeans considered it to be funeral and dreamt of it as unlucky, whereas in Italy, women wear it in their hair and the youth stick a spring of it above ear when they are courting. In India, Hindus believed that a leaf of basil buried with them would serve as their passport to heaven (Sekar et al. 2009). Sweet basil has been grown and sold in New York State since the end of the eighteenth century.

O. basilicum is known by different names in different languages around the world, including the Indian subcontinent. In English, it is known as basil or sweet basil, whereas in Hindi and Bengali it is called babui tulsi. The plant is known as badroaj, hebak, or rihan in Arabic, as nasabo or sabje in Gujrati, and as jungle tulsi in Urdu.

Basil oil is the oil obtained from basil (*O. basilicum*) through different conventional (e.g., steam distillation), nonconventional, or novel techniques having major constituents such as linalool, methyl chavicol, methyl cinnamate, eugenol, and cineole. It is reported to possess antimicrobial, bactericidal, and antioxidant activity, and fungicidal, marked insecticidal, lavricidal, and insect repellant properties against mosquitoes (Jayasinghe et al. 2003; Bozin et al. 2006; Kumar et al. 2011; Nour et al. 2012; Shirazi et al. 2014). Due to these activities, the oil is potent for use in perfumery, cosmetics, food, pharmaceutical formulation, and insecticidal and insect repellent formulations.

5.2 Botany of the Plant

5.2.1 Taxonomic Classification

Kingdom: Plantae
Subkingdom: Tracheobionta
Super division: Spermatophyta
Division: Magnoliophyta
Class: Magnoliophyta
Subclass: Asteriadae
Order: Lamiales
Family: Lamiaceae
Genus: Ocimum
Species: Basilicum
Botanical name: Ocimum basilicum

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5.2.2 Habitat and Distribution

Basil originated in Asia and Africa. Sweet basil is indigenous to Persia and Sindh and the lower hills of Punjab in India. The plant is widely grown as an ornamental and field crop throughout the greater part of India, Cameroon, Ceylon, and several Mediterranean countries, including Turkey.

5.2.3 Botanical Description of the Plant

Sweet basil is an autogamous, aromatic, and herbaceous plant that is annual and perennial. It is an erect branching herb, 0.6–0.9 m high, glabrous, and more or less pubescent. The stems and branches are green and sometimes purplish.

The leaves of *O. basilicum* are simple opposite. They are around 2–2.5 cm long, ovate acute entire or more or less toothed or lobed with a cuncate and entire base. The petiole is 1.3–2.5 cm long. The leaves have numerous dot-like oil glands that secrete strongly scented volatile oil. The whorls are densely racemose, where the terminal raceme is usually much longer than the lateral ones. The bracts are stalked, shorter than the calyx, ovate, and acute. The calyx is 5 mm long, with enlarging fruit, and very shortly pedicelled. Its lower lip with two central teeth is longer than the rounded upper lip. The corolla, 8–13 mm long, is white, pink, or purplish in color, glabrous or variously pubescent. The upper filaments of slightly exerted stamen are toothed at the base. Nutlets are about 2 mm long, ellipsoid, black, and pitted. The sepals of the flower are and remain fused into a two-lipped calyx. The ovary is superior, and there are two carpellaxy, four-locular, and a four-partite fruit of achenes.

5.3 Methods of Extraction of Oil

Essential oils are complex mixtures of volatile substances generally present at low concentrations. There are several methods for extracting the essential oil from the natural sources. Different methods can be used for that purpose.

5.3.1 Conventional Methods

Distillation and Soxhlet extraction are the conventional methods.

5.3.1.1 Distillation

Distillation is an extracting oil process. It converts volatile liquid (essential oils) into a vapor state and then condenses the vapor into a liquid state. There are different distillation processes, such as water distillation, steam distillation, and hydrodiffusion (Ranjitha and Vijiyalakshmi 2014).

5.3.1.1.1 Water Distillation

In this process, the botanic material is completely immersed in water and the still is brought to boil. It is used to protect the oils to a certain degree since the surrounding

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water acts as a barrier to prevent overheating. When condensed material cools down, the water and essential oil are separated and the oil is decanted to use as an essential oil. Water distillation can also be done at reduced pressure (under vacuum) to reduce the temperature to less than 100°C. This is useful in protecting the botanical material in obtaining the essential oil.

It is a simple and easy-to-operate extraction method of oil from plant species. Due to the use of heat in this method, it may not be used on very fragile plant material. Oil components like esters are sensitive to hydrolysis, while others like cyclic monoterpene hydrocarbons, and aldehydes are susceptible to polymerization. Oxygenated compounds such as phenols have a tendency to dissolve in distilled water, so their complete removal is not possible. As water distillation tends to be a small operation, it takes a long time to accumulate much oil (Ranjitha and Vijiyalakshmi 2014).

5.3.1.1.2 Steam Distillation

In the steam distillation method, the botanical material is placed in a still and steam is forced over the material. The hot steam is used to release the aromatic molecules from the plant material. The steam forces the pockets to open, and then the molecules of these volatile oils escape from the plant material and evaporate in the steam. The steam containing the essential oil is passed through a cooling system to condense the steam, which forms a liquid form of essential oil. Then, the water is separated. The steam is produced at greater pressure than atmospheric pressure, and therefore it helps to boil at 100°C without raising the temperature beyond 100°C. This is used to remove the essential oil from the plant material (Masango 2005; Ranjitha and Vijiyalakshmi 2014).

The major advantage of steam distillation is that the temperature never goes above 100°C, so temperature-sensitive compounds can be distilled. A disadvantage is that not many compounds can be steam distilled—only the aromatic ones.

5.3.1.1.3 Hydrodiffusion

The hydrodiffusion method is similar to the steam distillation process. The main difference between these two methods is how the steam is introduced into the still. In the case of hydrodiffusion, the steam is fed into the top, onto the botanical material, instead of from bottom, as in normal steam distillation. The steam containing the essential oil is passed through a cooling system to condense steam, which forms a liquid of essential oil, and then water is separated (Ranjitha and Vijiyalakshmi 2014).

The main advantages of this method are that less steam is used, the processing time is shorter, and there is a higher yield.

5.3.1.2 Soxhlet Extraction

Soxhlet extraction is a well-established technique that surpasses other conventional extraction techniques in performance, except for limited fields of application, for example, the extraction of thermolabile compounds. Most of the solvent extraction units worldwide are based on Soxhlet principles with recycling of solvents. Basically, the equipment consists of a drug holder extractor, a solvent storage vessel, a reboiler kettle, a condenser, a breather system, and supporting structures like a boiler, a refrigerated chilling unit, and a vacuum unit (William 2007).

This technique is based on the choice of solvent coupled with heat or agitation. In this process, the circulation of solvents causes the displacement of transfer equilibrium by

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repeatedly bringing fresh solvent into contact with the solid matrix. This method maintains a relatively high extraction temperature and no filtration of extract is required (Shams et al. 2015).

However, the limitation of this technique is that there is a possibility of thermal decomposition of thermolabile targeted compounds because the extraction usually occurs at the boiling point of the solvent for a long time.

Nidia et al. (2013) compared supercritical fluid extraction (SFE) using carbon dioxide with the Soxhlet and hydrodistillation processes for extraction of basil oil and reported a higher yield of oil by Soxhlet extraction.

5.3.2 Novel Extraction Methods

5.3.2.1 Microwave Extraction Method

Solvent-free microwave extraction (SFME) is used to separate the essential oil from plant material. The method involves placing the sample in a microwave reactor without any addition of organic solvent or water. The internal heating of the water within the sample distends its cells and leads to rupture of the glands and oleiferous receptacles. This process frees essential oil, which is evaporated by the *in situ* water of plant material.

A cooling system outside the microwave oven continuously condenses the vapors, which are collected in specific glassware. The excess of water is refluxed back to the extraction vessel in order to restore the *in situ* water to the sample.

The microwave isolation offers a net advantage in terms of yield and better oil composition. Furthermore, it is environmentally friendly. In this method, low-boiling-point hydrocarbon compounds undergo decomposition (Marie et al. 2004; Ranjitha and Vijiyalakshmi 2014).

5.3.2.2 Subcritical Water Extraction

Subcritical water extraction (SWE) is the extraction using hot water under pressure. It has recently emerged as a useful tool to replace traditional extraction methods. Subcritical water extraction is an environmentally clean technique that, in addition, provides higher extraction yields to extract solid samples. Subcritical water extraction is carried out using hot water (from 100°C to 374°C, the latter being the water critical temperature under high pressure [usually up to 10 bars]), enough to maintain water in the liquid state. The most important factor to take into account in this type of extraction procedure is the dielectric constant. This parameter can be modulated easily within a wide range of values by only tuning the extraction temperature.

Water at room temperature is a very polar solvent, with a dielectric constant close to 80. However, this level can be significantly decreased to values close to 27 when water is heated up to 250°C while maintaining its liquid state applying pressure. The dielectric constant value is similar to that of ethanol, and therefore is appropriate for solubilizing less polar compounds.

Basically, the experimental setup needed to use this technique is simple. The instrumentation consists of a water reservoir coupled to a high-pressure pump to introduce the solvent into the system, an oven where the extraction cell is placed and where the extraction takes place, and a restrictor to maintain the pressure along the extraction line. Extracts are collected in the collector vial placed at the end of the extraction system (Shams et al. 2015).

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5.3.2.3 Supercritical Fluid Extraction

Supercritical fluid extraction is used for the extraction of flavors and fragrances. SFE is a separation technology that uses supercritical fluid as solvent. Every fluid is characterized by a critical point, which is defined in terms of the critical temperature and critical pressure. Fluids cannot be liquefied above the critical temperature regardless of the pressure applied, but may reach the density close to the liquid state. A substance is considered to be a supercritical fluid when it is above its critical temperature and critical pressure.

The main supercritical solvent used is carbon dioxide. Carbon dioxide (critical condition 30.9°C and 73.8 bar) is cheap, environmentally friendly, and generally recognized as safe. Supercritical carbon dioxide is also attractive because of its high diffusivity and easily tunable solvent strength. Another advantage is that carbon dioxide is gaseous at room temperature and ordinary pressure, which makes analyte recovery very simple (Taylor 1996; Ranjitha and Vijiyalakshmi 2014).

5.3.2.4 Accelerated Solvent Extraction

Accelerated solvent extraction (ASE) is sometimes called pressurized solvent extraction (PSE). It uses organic solvents at elevated pressure and temperature in order to increase the efficiency of the extraction process. Increased temperature accelerates the extraction kinetics, and elevated pressure keeps the solvent in a liquid state, thus enabling safe and rapid extraction. Furthermore, high pressure forces the solvent into the matrix pores, and hence facilitates the extraction of analyte. High temperature decreases the viscosity of the liquid solvent, allowing a better penetration of the matrix and a weakened solute matrix interaction. Elevated temperature enhances diffusivity of the solvent, resulting in increased extraction speed. The solvent is selected based on the polarity of the analyte and compatibility with postextraction processing equipment. In ASE applications, generally organic solvents are used in conventional techniques, such as methanol. The use of hot water as the extraction solvent under atmospheric or higher pressure is very efficient for extracting phytochemicals.

ASE has been reported to be more efficient than other extraction methods by consuming less solvent and allowing faster extraction. However, since the extraction is performed at elevated temperature, the thermal degradation is a cause of concern, especially for thermolabile compounds in extracts.

The efficiency of ASE is influenced by factors such as pressure, temperature, static extraction time, flush volume, and vessel void volume (Shams et al. 2015).

5.3.2.5 Ultrasound-Assisted Extraction

The mechanical effect of ultrasound accelerates the release of organic compounds within the plant body due to cell wall disruption, mass transfer intensification, and easier access of the solvent to the cell content. Ultrasound-assisted extraction (UAE) is reported to be one of the important techniques for extracting valuable compounds from the vegetable material (Vilkhu et al. 2008). General ultrasonic devices are the ultrasonic cleaning bath and ultrasonic probe system.

The efficiency of UAE depends on various factors, such as the nature of the tissue being extracted, the location of the component to be extracted, the treatment of the tissue prior to extraction, the effect of ultrasonics, the surface mass transfer, and intraparticle diffusion.

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UAE can extract analytes under a concentrated form and free from any contaminants or artifacts. It also demonstrates advantages in terms of yield, selectivity, operating time, energy input, and preservation of thermolabile compounds (Shams et al. 2015).

5.4 Composition of Oil

Essential oils extracted from the plants are a complex mixture of terpenes, sesquiterpenes, oxygenated derivatives, and other aromatic compounds. These compounds are characteristic for basil aroma, which is a precursor to the presence of 1,8-cineole, methyl cinnamate, methyl chavicol, and linalool. In general, these substances are volatile and are present at low concentrations.

Lawrence (1985) classified basil oil into three large groups, European type, exotic or Reunion type, and African type, according to their chemical composition and geographical origin. He established four essential oil chemotypes: methyl chavicol, linalool, methyl eugenol, and methyl cinnamate.

The major constituents that have been isolated from different *Ocimum* oils include 1,8-cineole, linalool, piene, eugenol, camphor, methyl chavicol, ocimene, terpinene, and limonene. Within the *Ocimum* species, there is a clear variation in their composition in terms of types of constituents. The chemical composition of sweet basil oil has been investigated, and by now, more than 200 chemical components have been reported from many regions of the world (Marwat et al. 2011).

A substantial number of studies conducted on the composition of essential oil of basil revealed a huge diversity in constituents with different chemotypes from many regions of the world. The major constituents that have been isolated from different *O. basilicum* oils include linalool, methyl chavicol, eugenol, methyl cinnamate, 1,8-cineole, and bergamotene (Pandey et al. 2014). In the Czech Republic, Guinea, and Reunion, the major compounds of basil essential oil are linalool and eugenol (Klimankova et al. 2008). Marotti et al. (1996) reported the presence of linalool, methyl chavicol, and eugenol as components of Italian basil oil. Four major compounds reported in basil essential oil from Austria are linalool, methyl chavicol, methyl cinnamate, and α-cadinol. Linalool was reported as the main component in basil oil from Romania (Benedec et al. 2009).

However, the major compound from French basil leaf essential oil is methyl cinnamate (Adam et al. 2009). Regarding Pakistan basil, four major compounds were found: linalool, epi-α-cardinol, α-bergamonotene, and γ-cadinene (Hussain et al. 2008). In the case of Egypt basil oil, linalool, 1,8-cineole, eugenol, and methyl cinnamate are the dominant components (Simon et al. 1990). Concerning basil oil from Turkey, three major compounds, methyl chavicol, limonene, and p-cymene, were cited (Chalchat and Ozcan 2008). Essential oil from Iran and Thailand is rich in methyl chavicol (Bunrathep et al. 2007). Camphor, followed by limonene and β-selenene, was the major compound in *O. basilicum* essential oil from northeast India. Egyptian basil oil is very similar to the European type characterized by linalool and methyl chavicol as major compounds (Simon et al. 1990).

The observed difference in the constituents of basil essential oil between countries may be due to environmental conditions and genetic factors, different chemotypes, and nutritional elements of the plants, as well as other factors.

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5.4.1 Effect of Extraction Method on Composition and Yield of Basil Oil

Techniques commonly employed for extracting essential oils include hydrodistillation, steam distillation, solvent extraction, and liquid carbon dioxide extraction. The composition of extracted oil may vary from one extraction method to another.

Charles and Simon (1990) compared the influences of three extraction methods on measurements of essential oil content. No significant difference was observed between steam distillation and the hydrodistillation method.

The extraction yields obtained by SFE using CO_2 were from 0.719% to 1.483% (w/w), depending on CO_2 density. The predominant compounds of the CO_2 extracts were linalool (10.14%–16.60%), eugenol (5.91%–9.78%), and ∂-cadinene (3.94%–7.21%) depending on SFE condition. The extraction yields using SFE were higher for some compounds (eugenol, α-beragmotene, and ∂-cadinene) than those obtained by hydrodistillation (Zekovic et al. 2015).

5.5 Methods of Analysis of Basil Oil

The chemical composition of basil oil is analyzed generally using gas chromatographic methods, such as by gas chromatography with a flame ionization detector (GC-FID) or gas chromatography with mass spectrometry (GC-MS).

5.5.1 Gas Chromatography with Flame Ionization Detector

Basil oil may be analyzed using GC-FID. Different types of column (nonpolar, polar, and a combination of polar and nonpolar) have been used for the analysis of essential oil. Based on the required resolution and target analyte, different types of column and types and quantities of mobile phase and temperature conditions have been used. Quantification of compound is done based on area normalization or peak area of external standards. Retention indices (RIs) using n-alkanes (octane, nonane, decane, dodecane, octadecane, eicosane, docosane, tetracosane, and hexacosane) were used as the basis.

5.5.2 Gas Chromatography with Mass Spectrometry

Today, GC-MS plays an important role in the identification of the chemical composition of basil essential oil. Various researchers worldwide have used this technique and identified a number of analytes, as shown in Table 5.1.

Nevertheless, incomplete identification was achieved by GC-MS due to the complex nature of constituents of essential oil. Comprehensive two-dimensional gas chromatography (GC×GC) is a powerful technique that has been successfully used for separation of the volatile constituents in highly complex samples. This technique is a combination of two columns with different separation mechanisms coupled via a cryogenic modulator interface. Many coeluting components on the first column are separated in the second column. The application of comprehensive two-dimensional gas chromatography coupled to time-of-flight mass spectrometry (GC×GC-TOFMS) has been employed to analyze the aromatic compounds of basil samples (Klimankova et al. 2008). The relative abundance of different constituents allowed differentiation between examined cultivars.

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TABLE 5.1

Methods Used for Analysis of Basil Oil

Serial No.	Source of Oil	Method of Extraction	Number of Compounds	Method Used	Reference
1	Leaves of *O. basilicum*	Steam distillation	4	GC-MS	Mindaryani and Rahayu (2007)
2	Leaves and flowers of *O. basilicum*	Supercritical fluid extraction	20	GC-MS and GC-FID	Zekovic et al. (2015)
3	Aerial parts of *O. basilicum*	Distillation	73	GC-MS Column: HP-5MS capillary 30 m × 0.25 mm i.d., 0.25 μ thickness Detector: MSD Temp program: 100 @ 2°C/min to 220°C Detector temp: 280°C Injector temp: 250°C	Pripdeevech et al. (2010)
4	Fresh leaves of *O. basilicum*	Hydrodistillation	16	Column: RXi-5MS capillary 30 m × 0.25 mm i.d., 0.25 μ thickness Detector: mass spectroscopy detector (MSD) Temp program: 100 @ 2°C/min to 220°C Detector temp: 280°C Injector temp: 250°C	Astuti et al. (2016)
5	Fresh aerial parts of *O. basilicum*	Hydrodistillation	20	GC-MS Column: Rtx-5 capillary 30 m × 0.25 mm i.d., 0.25 μ thickness Detector: MSD Temp program: 50 @ 5°C/min to 250°C Detector temp: 230°C Injector temp: 240°C	Ismail (2006)
6	Leaves of *O. basilicum*	Solvent-free microwave extraction and hydrodistillation	65	GC-MS and GC-FID Column: HP-1 capillary 50 m × 0.2 mm i.d., 0.25 μ thickness Detector: FID Temp program: 45 @ 2°C/min to 250°C Detector temp: 250°C Injector temp: 250°C	Chenni et al. (2016)
7	Fresh leaves of *O. basilicum*	Steam distillation	13	GC-MS Column: fused silica capillary 30 m × 0.25 mm i.d., 0.25 μ thickness Detector: MSD Temp program: 100 @ 2°C/min to 220°C Detector temp: 280°C Injector temp: 250°C	Pripduvech et al. (2010)

(*Continued*)

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TABLE 5.1 (CONTINUED)

Methods Used for Analysis of Basil Oil

Serial No.	Source of Oil	Method of Extraction	Number of Compounds	Method Used	Reference
7	Fresh leaves of *O. basilicum*	Hydrodistillation	16	Column: RXi-5MS capillary 30 m × 0.25 mm i.d., 0.25 μ thickness Detector: MSD Temp program: 70°C for 2 min, from 70°C @ 3°C/min to 230°C, @ 5°C to 240°C Detector temp: 250°C Injector temp: 250°C	Nour et al. (2012)

Note: MSD, mass spectroscopy detector; Temp, temperature.

5.6 Physicochemical Properties of Basil Oil

The physicochemical properties include appearance, color, solubility, density, specific gravity, refractive index, and optical rotation (Table 5.2). These may vary from type to type and by method of extraction of basil oil. There are several standards at the national and international level to maintain the quality of the oil.

Chenni et al. (2016) reported that there are no significant differences between the physical constants of essential oil obtained by hydrodistillation and solvent-free microwave extraction, except for differences in color and odor. The color of the essential oil obtained by SFME was lighter and the odor more pleasant.

TABLE 5.2

Physicochemical Properties of Basil Oil as per the Food Chemical Codex

Serial No.	Parameter	Basil Oil, Comoros Type	Basil Oil, European Type	Reference
1	Description	Light yellow liquid with camphoraceous odor	Pale yellow liquid with a more floral odor	Food Chemical Codex (2014)
2	Specific gravity at 20°C	0.952–0.973	0.900–0.920	Food Chemical Codex (2014)
3	Angular rotation at 20°C	–2° to +2°	–5° to +5°	Food Chemical Codex (2014)
4	Refractive index at 20°C	1.512–1.520	1.483–1.493	Food Chemical Codex (2014)
5	Acid value	1.0 max	2.5 max	Food Chemical Codex (2014)
5	Saponification value	4–10	Not specified	Food Chemical Codex (2014)
6	Ester value of acetylation	25–45	140–180	Food Chemical Codex (2014)
7	Solubility in 80% ethanol	Should pass the test	Should pass the test	Food Chemical Codex (2014)

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5.7 General Uses of Basil Oil

Sweet basil (*O. basilicum*) essential oil has been used for centuries in perfumery, cosmetics, and medicine, and has been added to food as a part of spices or herbs. The essential oil has been investigated, and now more than 200 chemical components have been reported from many regions of the world. The essential oil or the components have been shown to possess not only a broad range of antimicrobial (antibacterial and antifungal) properties (Table 5.3), but also antiproliferative or anticancer, antioxidant, and antiwormal activities, and it also affects the central nervous system (CNS) activities, as summarized in Table 5.4.

TABLE 5.3

Antimicrobial Activities of Various Constituents of Basil Oil (*Ocimum basilicum*)

Serial No.	Constituents	Biological Activity	Organism	Reference
1	Essential oil	Antibacterial	*Staphylococcus aureus*, *Salmonella enteritidis*, *Escherichia coli*	Marwat et al. (2011)
2	Essential oil	Antibacterial	*Bacillus cereus*	Budka and Khan (2010)
3	Essential oil	Antibacterial	*Haemophilus influenza*, pneumococci	Kristinsson et al. (2005)
4	Essential oil	Antibacterial	*Staphylococcus aureus*	Nguefack et al. (2004)
5	Linalool, methyl chavicol, methyl cinnamate	Antibacterial	*Staphylococcus aureus*, *Enterococcus*, *Pseudomonas*	Opalchenova and Obreshkova (2003)
6	Essential oil	Antibacterial	*Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis*, *Pasteurella multocida*	Suppakul et al. (2003)
7	Linalool	Antibacterial	*Giardia lamblia*	Almeida et al. (2007)
8	Rosamarinic acid	Antibacterial	*Pseudomonas aeruginosa*	Bais et al. (2002)
9	Essential oil	Antibacterial	*Bacillus cereus*, *Bacillus subtilis*, *Bacillus megaterium*, *Escherichia coli*, *Staphylococcus aureus*, *Listeria monocytogenes*, *Shigella boydii*, *Shigella dysenteriae*, *Vibrio mimicus*, *Vibrio parahaemolyticus*, *Salmonella typhi*	Hossain et al. (2010)
10	Essential oil	Antifungal activity	*Cadida albicans*, *Penicillium notatum*, *Microsporeum gyseum*	Marwat et al. (2011)
11	Essential oil	Antifungal activity	Yeast and mold	Suppakul et al. (2003)
12	Essential oil	Antifungal activity	*Aureobasidium pullulans*, *Debaryomyces hansenii*, *Penicillium citrinum*, *Penicillium expansum*	De Martino et al. (2009)
13	Essential oil	Antifungal activity	*Alternaria alternata*, *Fusarum solani* var. *coeruleum*	Zhang et al. (2009)
14	Linalool, methyl chavicol, cineole, eugenol	Antifungal activity	*Fusarium oxysporum* f. sp. *vasinfectum*, *Rhizopus nigricans*	Reuveni et al. (1984)
15	Linalool and eugenol	Antifungal activity	*Sclerotonia sclerotiorum*, *Rhizopus stolonifer*	Edris and Farrag (2003)

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TABLE 5.4
Pharmacological Actions of Basil Oil (*Ocimum basilicum*)

Serial No.	Biological Activity	Constituents	Observation	Potential Use	Reference
1	Antimicrobial and antibiofilm activity	Essential oil	Antibacterial activity against *Streptococcus mutans* and biofilm formation inhibition and biofilm degradation	As active ingredient in antibacterial and antibiofilm product formulation	Astuti et al. (2016)
2	Antioxidant activity	Essential oil	In hypoxanthine/xanthine oxidase assay, strong antioxidant activity was evidenced		Jayasinghe et al. (2003)
3	Anticancer activity	Essential oil	Had an IC_{50} value of 0.0362 mg/ml (12.7 times less potent than 5-Fluorouracil [5-FU]) in P388 cell lines	Potential for cancer treatment	Manosroi et al. (2006)
4	Antiviral	Apigenin, linalool, and ursolic acid	Broad spectrum of antiviral activity against herpes virus, hepatitis B, and adenoviruses		Chiang et al. (2005)
5	Dermatologic effect	Essential oil	Effect on acne vulgaris in humans		Balambal et al. (1985)
6	Antigiardial activity	Essential oil	Inhibits proteolytic activity mainly of cysteine proteases		Almeida et al. (2007)
7	Antiwormal response	Volatile oil	Two fractions showed antiwormal response under test condition		Marwat et al. (2011)
8	Spemicidal effect	Essential oil	Spermicidal action	Spermicidal purpose	

5.7.1 Food

The food industry primarily uses basil oil as flavoring. However, it also represents an interesting source of natural antimicrobials for food preservation. The addition of basil oil results in a better flavor and less microbial contamination. The flavor of sausage was better retained when basil oil was added in the form of microencapsulated oleoresin, which penetrated even in intramuscular fats in sausage (Flint and Seal 1985). A basil flavor has been developed that can be directly sprayed on foods that are particularly suited for the microwave.

Basil essential oil and its principal constituents exhibit antimicrobial activity against a wide range of gram-negative and gram-positive bacteria, yeasts, and molds. According to Budka and Khan (2010), essential oil from basil oil exhibited bactericidal properties against *Bacillus cereus* in rice-based food. The methanolic extract of *O. bacilicum* showed antibacterial activity against *Pseudomonas aeruginosa*, *Shigella* sp., *Listeria monocytogenes*, *Staphylococcus aureus*, and two different strains of *Escherichia coli*. Basil oil had the strongest antimicrobial activity against *Salmonella* sp. (*S. enteritidis* SE3). The oil at a concentration of 50 ppm reduced the number of bacteria in the food from 5 to 2 log cfu/g after storage for 3 days. At a level of 100 ppm in ham, the bacteria count was reduced without affecting the acceptance of consumers.

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5.7.2 Cosmetics

Basil essential oil is used topically and is massaged into the skin. It enhances the luster of dull-looking skin and hair. As a result, it is frequently used in many skin care supplements that claim to improve the tone of the skin. Basil oil is appreciated by the cosmetic industry because it has antimicrobial and antioxidant properties. Basil oil showed an enhancing activity for accelerating the transdermal delivery of indomethacin (Fang et al. 2004). The mechanism of action is probably due to the increased skin–vehicle partitioning by the oil.

5.7.3 Pharmaceutical

The essential oil of *O. basilicum* has been used in folk medicine to treat various diseases. It has been studied in different parts of the world, and it has been reported to exhibit various biological activities, such as antibacterial and antifungal properties (summarized in Table 5.4), antioxidant activities, anticancer activity, antiwormal response, antiviral activity, dermatological effects, spermicidal effect, and effects on the central nervous system. The constituents or parts of basil oil studied for different biological activities and their observation and potential use are described in Table 5.4.

5.7.3.1 Antibacterial and Antifungal Activities

The antibacterial and antifungal activity of basil oil leads to its use in dental care. Due to its antifungal and disinfectant action, it cleans and disinfects the mouth and relieves inflammation of the gums. The antibacterial potency of the essential oil suggests it has potential activity against *Streptococcus mucans*, which is the main cause of dental carries. The oil has been used in a microemulsion mouth wash formula and exhibited antibacterial and antibiofilm activity and was stable for 3 months at accelerated storage conditions. It was suggested as an active ingredient in an antibacterial and antibiofilm formulation (Astuti et al. 2016). As a component of mouth wash, the volatile oil completely inhibited the growth of organisms at a concentration of 0.5% (Ahonkhal et al. 2009). Volatile oil of *O. basilicum* at 0.01% in toothpaste showed antibacterial activities against most resistant organisms.

5.8 Pesticidal Uses of Basil Oil

This essential oil also possesses properties to act as an insecticide. Basil oil also has marked repellency and larvicidal properties against mosquitoes.

5.8.1 Insecticidal Activity of Basil Oil

Essential oils of plants contain a number of bioactive compounds that may exert regulatory or inhibitory influence on insect life processes, such as growth and development, reproduction, or orientation. Among the compounds present in essential oils are monoterpenes, which are consequently regarded as a candidate for insecticidal activity. These natural compounds have been proposed as lead compounds for the development of safe, effective, and fully biodegradable insecticides. Most of the monoterpenes are cytotoxic to plant and animal tissue, causing a drastic reduction in the number of mitochondria and Golgi bodies,

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impairing respiration and photosynthesis and decreasing cell membrane permeability (Tripathi et al. 2009). At the same time, they are volatile and may serve as chemical messengers for insects. The doses of essential oils needed to kill insects or pests and their mechanism of action are important for the safety of humans and other vertebrates. Therefore, the target sites and mode of action need to be understood and should be well elucidated. Although little is known about the physiological actions of essential oils on insects, treatment with various essential oils causes symptoms that suggest a neurotoxic mode of action. A monoterpenoid linalool has been demonstrated to act on the nervous system, affecting ion transport and releasing acetylcholine esterase in insects (Re et al. 2000).

Digilio et al. (2008) assayed essential oil extracted from *O. basilicum* for insecticidal activity against the aphid pests *Acyrthosiphon pisum* (Harris) and *Myzus persicae* (Sulzer). Basil essential oil resulted in high mortality (100%) against *A. pisum* and 96.15% against *M. persicae*, even applied at low doses, and activity was dose dependent. The pea aphid *A. pisum* is a phloem-feeding insect that colonizes leguminous crops where it produces direct damage in terms of nutritive subtraction and the injection of toxic saliva, besides being responsible for the transmission of more than 30 viral diseases (Blackman and Eastop 2000). The green peach aphid or peach potato aphid *M. persicae*, a major pest worldwide, has an extreme host range over 40 different plant families, where it not only produces severe direct damage but also is considered the most important aphid virus vector, being able to transmit more than 100 plant viruses. *M. persicae* has resistance to many synthetic insecticides. The insecticidal activity of basil essential oil is assumed to be due to the presence of monoterpenes (Diglio et al. 2008).

Basil oil and three major active constituents (*trans*-anethole, estragole, and linalool) were tested on three tephritid fruit fly species for insecticidal activity. All tested chemicals acted fast and showed a step dose–response relationship (Chang et al. 2009).

5.8.2 Larvicidal Activity

To minimize and eradicate the occurrence of mosquito-borne diseases, many steps have been taken to prevent their spread to different extents, for example, mosquito eradication at an early stage, disease prevention via prophylactic drugs and vaccines, and the prevention of mosquito bites using repellants. Out of these, larviciding has the greatest control impact on the mosquito population because the larvae are concentrated, immobile, and accessible. Nour et al. (2012) studied the essential oils from two basil accessions for larvicidal activity against instar *Aedes aegypti*, the major vector for dengue and yellow fever disease larvae. The steam-distilled essential oil showed larvicidal activity against *A. aegypti*. The larval mortality increased as the essential oil concentration increased, and the essential oil from the different accessions had different activities. The highest dose (500 μg/ml) caused 100% mortality after 3 hours' incubation with oil of the methylchavicol accession (MCV), whereas the same dose caused 100% mortality after 6 hours with geranial–geraniol accession (GGV).

5.8.3 Basil Oil as a Repellant

How repellents work in different arthropods is unclear, since conflicting evidence exists, such as different insects detecting the same substance by different organs, but there is a difference in sensitivity to the same repellent. Hairs on the mosquito antennae are temperature and moisture sensitive. The repellant molecules interact with the female mosquito olfactory receptors, thereby blocking the sense of smell. Very little is known about the receptors responsible for the repellent responses in cockroaches. Oleic acid and linoleic acid have been indicated in death recognition and death aversion in cockroaches, and the

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term *necromone* has been proposed to describe the compound responsible for this type of behavior (Rollo et al. 1995).

The hexane extract of *O. basilicum* in melted petroleum jelly has been studied for repellent activities against mosquito *A. aegypti*. At a concentration of 2%, repellency was observed to be 91.45%. Complete protection was obtained at 3% of the oil, but beyond this concentration, there was no significant change in activity observed. Furthermore, mosquito paralysis occurred at this concentration, which is similar to the reported effect of DEET (Kiplang'at and Mwangi 2013). The essential oil from *O. basilicum* at a 2% level showed a significant mortality, repellency, and an antireproductive effect against the rice weevil *Sithophilus oryzae* L. (Popovic et al. 2006).

5.9 Advantages of Basil Oil as a Pesticide

The constituents of basil oil have little or no harmful effect on the environment and the nontarget organism. Due to the multiple sites of action through which the constituents can act, the probability of developing a resistant population is very low. These botanical insecticides degrade rapidly in air and moisture, and detoxification enzymes break them readily. Due to rapid breakdown, they have less persistence in the environmental, larvicidal, and adulticidal activities to sublethal effects, including oviposition, deterrence, and repellent actions (Koul et al. 2008).

5.10 Constraints of Basil Oil as a Pesticide

Although essential oils as pesticides have a number of advantages over synthetic pesticides, there are some specific constraints. The efficacy of these materials falls short when compared with synthetic pesticides. The commercial application of plant essential oil–based pesticides includes the availability of sufficient quantities of plant material, standardization and refinement of pesticide products, protection of technology (patents), and regulatory approval. In addition, as the chemical profile of plant species can vary naturally depending on geographic, genetic, climatic, or seasonal factors, pesticide manufacturers have to take additional steps to ensure that their product will perform consistently. All this requires substantial costs, and smaller companies are not willing to invest the required funds unless there is high probability of recovering the costs through some form of market exclusivity. Finally, once all these issues are addressed, regulatory approval is required (Mohan et al. 2011).

5.11 Basil Oil–Based Insecticides

In spite of considerable research efforts in many laboratories throughout the world and an ever-increasing volume of scientific literature on the pesticidal properties of essential oils and their constituents, surprisingly few pest control products based on basil essential oils

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have appeared in the marketplace. This may be a consequence of regulatory barriers to commercialization, or the fact that the efficacy of essential oils toward pests and diseases is not apparent or as obvious as that seen with currently available products.

Krishnamurthy et al. (2012) studied a 25% emulsifiable concentrate (EC) formulation of basil and geranium, each at a 1 ml/L dose, that is, 0.0125% active ingredient of essential oil. This formulation at this dose was again found to be effective against chilli thrips. This EC formulation of essential oils evaluated under field conditions and consistently gave good results against thrips in *Capsicum* under polyhouse conditions by basil formulation. Hence, there is great potential in using essential oils and their formulations in the integrated pest management (IPM) of thrips in these crops. The main constraint in using essential oils as such is their cost, as well as the quality. The cost can be reduced by using commercial ready-to-use essential oil formulations; a few are available in the United States.

Environmentally and user-friendly emulsifiable concentrates of basil oil using biodegradable vegetable oil and a suitable emulsifier such as Tween 80 and Tween 60 have been developed. The EC with 5% (w/w) using methyl oleate as a solvent has been found to have antifungal activity, which may be due to the presence of linalool, beta linalool, and methyl chavicol in the basil oil (Thakur et al. 2014).

5.12 Conclusion

The constituents of basil oil exhibit various activities, such as antimicrobial, antioxidative, anticarcinogenic, antiviral, and insecticidal activities. Therefore, this oil has great potential not only in food, pharmaceuticals, and cosmetics, but also as insect repellant and insecticide, and in a variety of ways to control a large number of pests. Although the efficacy of these basil oil constituents is comparatively less than that of synthetic pesticides, it is gaining momentum as far as environment pollution and human health are concerned. It is expected that the innovative formulations of pesticides based on this essential oil will find their greatest commercial applications in urban pest control, vector control vis-à-vis human health, and pest control in agriculture, and will help in organic food production systems, where few alternative pesticides are available.

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