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Review

Genus *Etlingera* - A review on chemical composition and antimicrobial activity of essential oils

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Essential oil from plants belonging to several species has been extensively used as natural resources in the prevention and treatment of a large number of diseases. The medicinal and aromatic plants of genus *Etlingera* produces high percentages of essential oil from its every organ and is recommended for a variety of health problems by traditional systems of medicine in south-east Asia. Essential oils from this genus demonstrated promising antimicrobial properties. As antibiotic resistance is emerging at an alarming rate, many infectious diseases have become difficult to treat. Thus, the genus *Etlingera* can be considered as a good source of essential oils, and extensive studies of biological activities of these oils may lead to the identification of new compounds which can be used in modern medicine, cosmetics and pharmaceutical industry, primarily as antimicrobial agents. This review summarizes the characteristics of essential oil of *Etlingera* species with particular attention to the chemical composition and antimicrobial activities from the data in the recent literature.

Key words: *Etlingera*, essential oil, chemical composition, antimicrobial activity.

INTRODUCTION

Etlingera is a genus of medicinal plants native to the Indo-Pacific region. It has been used by indigenous communities for its flavor, culinary and medicinal properties since antiquity. The well-established traditional uses may be explained by the presence of biologically active volatile components in this genus. However, despite the increasing scientific interest in this field, there is a lack of summarized data on herbal medicine composition, therapeutic applications and risks associated to their consumption. Therefore, the purpose of this article is to provide an overview of the published data results regarding chemical composition and antimicrobial activities of essential oil of *Etlingera* species.

Essential oils (EOs), also known as volatile or ethereal oil or aetherolea, are concentrated hydrophobic liquid containing volatile aroma compounds obtained from aromatic plants. An oil is “essential” in the sense that each plant oils contains characteristic “essence” of the plant's fragrance from which it is derived. Essential oils are generally highly odorous, volatile with penetrating taste. Although, their consistency is more like water than oil, it is lighter than water and give transparent to pale yellow color. It is a complex mixtures of various compounds containing about 20-100 components at quite

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different concentrations. Chemically, they are derived from terpenes and their oxygenated compounds and also include non-terpenic compounds such as alcohols, aldehydes, ethers, ketones, phenols, oxides and phenylpropanoids. Many essential oils have limited solubility in aqueous solutions but are soluble in alcohol, non-polar or weakly polar solvents, waxes and oils.

The term 'essential oil' was used for the first time in the sixteenth century by the Swiss reformer of medicine, Paracelsus von Hohenheim, who named the effective component of a drug as Quinta essential (Guenther, 1948). They are not essential for health but have been used to relieve a wide variety of human maladies including bronchitis, pneumonia, pharyngitis, diarrhea, periodontal disease, wounds and numerous other illnesses for thousands of years (Boire et al., 2013). Today, the term 'essential oils' is used to describe complex mixtures of low molecular weight (usually less than 500 daltons) compounds extracted from aromatic plants using conventional techniques. At present, around 3000 essential oils are known, of which 300 are commercially important especially used as flavoring agents in food products, drinks, perfumeries, pharmaceuticals and cosmetics.

Morphology of *Etlingera* species

The genus *Etlingera* belonging to Zingiberaceae family consists of more than 100 species. They are perennial herbs mainly grown in perhumid forest (Poulsen, 2007). The leafy shoots of some of the *Etlingera* species can be up to eight m tall and the bases of these shoots are so stout as to seem almost woody. Many of the *Etlingera* species grow as clumps of leafy shoots, while others have such long creeping rhizomes that each of their leafy shoots can be more than a meter apart. True, *Etlingera* is characterized by its unique and distinctive flowers which have exceptional ornamental value. The inflorescence shoots are found short and do not emerge from the ground. The flowers are characterized with prominent bright red petal-like structures (labella) radiating outward, with the flower tubes and ovaries being below ground level. The brightly colored flowers are thought to be pollinated by insects. Fruits ripen below ground, and the seeds are thought to be dispersed by wild pigs. These plants are very aromatic because of the high content of essential oil in its leaves, stems, flower, fruit and rhizomes.

Distribution

Etlingera is dominant in Indo-pacific terrestrial, native to India, Bangladesh, Burma, China, Laos, Cambodia, Vietnam, Thailand, Malaysia, Singapore, Indonesia, Philippines, Brunei, Papua New Guinea, Queensland and several Pacific Islands, predominantly close to the equator between sea level and 2500 m (Poulsen, 2006; Wojdyło et al., 2007). *Etlingera* are also naturalized in other warm places such as Hawaii, Puerto Rico, Trinidad, Central America, Mauritius and the islands of the Gulf of Guinea (Govaerts et al., 2011). These species are also cultivated in gardens, especially in Mexico and western parts of Indonesia (Java) as an ornamental plant and a source of condiment and spice. A total of 155 names of *Etlingera* species have been accepted in the World Checklist of Selected Plant Families facilitated by the Royal Botanic Gardens at Kew, England (Govaerts et al., 2011). Borneo rainforest is exceptionally rich and presently, at least forty species are known in this forest (Poulsen, 2006). Of these species, eighty-five percent of the *Etlingera* species in Borneo are endemics and thirty-three percent are found in Brunei Darussalam. Three new species of *Etlingera: Etlingera rubromarginata* (from Sabah, Sarawak and Brunei), *Etlingera belalongensis* (from the Temburong District of Brunei), and *Etlingera corrugata* (presently only known from Danum Valley, Sabah) from northern Borneo have recently been described (Poulsen et al., 1999). *Etlingera kenyalang* from Sarawak and *Etlingera palangkensis* from Central Kalimantan have also been reported (Poulsen and Christensen, 2003; Takano and Nagamasu, 2006). Fifteen *Etlingera* species in Malaysia Peninsular, nine species in Java, Indonesia and only three species (one endemic and one introduced) have been recorded in China (Chen and Boufford, 2000).

Traditional application

Plants of *Etlingera* have been used since ancient times as spice and vegetable as well as for medicinal purposes. The common traditional uses of different *Etlingera* species are shown in Table 1.

More specialized uses of few species include perfume (rhizome of *Etlingera baramensis*), shampoo (fruit of *Etlingera elatior* and *Etlingera pyramidosphaera*) and spice (rhizome of *Etlingera punica)* (Chan et al., 2013). Several other species of *Etlingera* also have been used as medicine in the prevention and therapy of diseases (e.g. rheumatism: *Etlingera foetens*; jaundice, fever, urinary ailments: *E. belalongensis;* stomach-ache*: E. pyramidosphaera;* snake bite: *Etlingera sessilanthera,* diarrhea: *E. pyramidosphaera*) (Poulsen, 2006; Sabli et al., 2012; Sirirugsa, 1997).

ESSENTIAL OILS IN COMMERCIAL PREPARATIONS

Several European countries have developed some essential oil-based industry in last few decades. Carvon, the principal constituent of the dill and caraway seeds EO, is currently marketed as Talent® in the Netherlands.

Table 1, Traditional uses of different plant parts of *Etlingera* species.

***Etlingera* species**	**Plant parts and preparation**	**Traditional use**	**Reference**
E. elatior	Young shoot, flower bud	As condiment	Abdelmageed et al. (2011b) and Noweg et al. (2003)
	Fruit decoction	In treating earache	
	Leaf	As wound cleaner, and body odor remover	
	Inflorescence	Food	Chan et al. (2007)
E. brevilabrum	Leaf (green)	To treat dry skin of the legs	Mahdavi (2014)
	Leaf (roasted)	To treat fever by rubbing on the bodies of children	
	Sap of heated young stem	To treat sore eyes	Poulsen (2006)
	Stolon	To cure stomachache	
E. linguiformis	Leaf and shoot	As vegetable	Kithan and Daiho (2014) and Ramana et al. (2012)
	Rhizome	To treat jaundice, sore throat, stomachache, rheumatism and respiratory complications	
	Sliced rhizome (with betel leaf)	To cure sore throat	Hossan et al. (2013)
E. labellosa	Juice of pseudostem	To cure body ache	Poulsen (2006)
E. littoralis	Young shoot, Flower bud, Fruit (raw or cooked)	As condiment	Noweg et al. (2003)
E. rubrolutea	Decoction of rhizome	To treat stomach ache, and as carminative and heart tonic	Chuakul and Boonpleng (2003)
E. coccinea, E. sessilanthera, E. volutina, E. rubromarginata	Inner sheath of leafy shoot	As condiment	Chan et al. (2007) and Poulsen (2006)
E. fimbriobracteata	Fruit	As condiment	
E. maingayi	Flower	As vegetable	

The preparation is used to inhibit the growth of storage pathogens and to suppress sprouting of potatoes in the warehouse (Hartmans et al., 1995). Soil Technologies Corporation (USA) has developed two natural products named Fungastop™ and Armorex™ which are commercially available for the control of various plant diseases in agriculture (Dubey et al., 2012). Eugenol based formulations (eugenol-Tween®;

eugenolethoxylate) showed potent inhibitory effect against four apple pathogens (*Phlyctema vagabunda, Penicillium expansum, Botrytis cinerea* and *Monilinia fructigena)* and thus used in post-harvest disease management of apple fruit (Amiri et al., 2008). Cinnamite™ (cinnamon), Valero™ (rosemary), Promax™ (thymus) and several other essential oil based pesticides are already commercially available (Prakash et al., 2015). EOs or their components (α-bisabolol, geraniol, elemene, *d*-limonene, diallyl trisulfide (DATS) and Eucalyptol) have been shown to exhibit cancer suppressive activity against glioma, colon cancer, gastric cancer, human liver tumor, pulmonary tumors, breast cancer, leukemia and others (De Angelis, 2001). Essential oils rich in terpinolene and/or eugenol have shown antioxidative activity against low density lipoprotein (LDL) oxidation thereby reducing the chance of atherosclerosis (Edris, 2007). Essential oils and their components are exploited for antibacterial properties in diverse commercial products as dental root canal sealers, antiseptics and feed supplements for lactating sows and weaned piglets (Burt, 2004). A few preservatives containing EOs are already available in the market, such as DMC Natural base, which comprises 50% essential oils (Speranza and Corbo, 2010). Beside these, essential oil and their individual constituents exhibited antiviral, antimycotic, antiparasitic and insecticidal properties (Bakkali et al., 2008; Dubey et al., 2010).

EXTRACTION PROCEDURES OF ESSENTIAL OIL

Extraction of essential oils can be achieved by various methods such as distillation, solvent extraction, effleurage, aqueous infusion, cold or hot pressing, supercritical fluid extraction, solvent free microwave extraction (SFME) and phytonic process. The method of extraction is normally dependent on what type of botanical material is being used. It is the key step that determines the quality of the oil as wrongly executed extraction method can damage the oil and alter the chemical signature of the essential oil. Today, hydro-distillation (with a collecting solvent that is then removed under vacuum) and steam distillation are widely used for extracting essential oils from plants. Volatile components in these methods can be distilled at temperatures lower than their individual boiling points and are easily separated from condensed water. Losses of some volatile compounds, low extraction efficiency, degradation of unsaturated or ester compounds of these widely used conventional methods have led to the consideration of the use of new “green” technique in essential oil extraction, which typically use less solvent and energy, such as supercritical fluids, ultrasound and microwave extraction. Berka-Zougali et al. (2012) describes a new innovative method, solvent free microwave extraction (SFME), which yields an essential oil with higher amounts of more valuable oxygenated compounds and allows substantial savings of costs, in terms of time, energy and plant material. However, essential oils obtained by SFME were quantitatively (yield) and qualitatively (aromatic profile) similar to those obtained by conventional method (hydro-distillation), while SFME is highly effective for reducing extraction time (30 min for SFME against 180 min for hydro-distillation) (Périno-Issartier et al., 2013). In *Etlingera* species, hydro-distillation technique is extensively used for extracting essential oils except from the flower of *E. elatior,* where steam distillation is used.

Yield and chemical composition of essential oil of *Etlingera*

Essential oils are derived from almost all parts of plant (leaf, stem, flower, peduncle, bark, rhizome, seed and fruit) in *Etlingera* species. Total oil content in this genus was found very low and rarely exceeds 1% by mass. For example, the essential oil yields in leaf, stem, flower, peduncle, rhizome and whole plant of *Etlingera* species were found in the range 0.031 - 1.94%, 0.001 - 0.02%, 0.014 - 0.9%, 0.005 - 0.1% 0.006 - 1.4% and 0.004 - 0.07% (w/w) respectively as shown in Table 1. Leaves showed the highest yield, while least percentage was obtained from stems. The ranking was in the order: leaf>rhizome>flower>peduncle>whole plant>stem. Essential oil yields of the same plant parts can also vary on the time of collection (Vahirua-Lechat et al., 2010). Leaves of *Etlingera cevuga* showed the highest percentage of yield than other *Etlingera* species. The most extensively studied plant in *Etlingera* species is *E. elatior* and the plant part is rhizome.

Essential oils and their components have gained wide acceptance by consumers because of promising biological activities, safety, and exploitation for potential multi-purpose uses. The chemical composition of the *Etlingera* (around 21 different species including 2 varieties) essential oil has been described by many authors that has been summarized in Table 2.

Table 2 shows that different species *Etlingera* are dominated by different chemical components. Vahirua-Lechat et al. (2010) analyzed the essential oil of *E. cevuga* by capillary gas chromatography and combined GC/MS. Thirty-one components were identified where methyl eugenol (40.9-45.7%) and (E)-methyl isoeugenol were the major constituents. The author also reported that the percentage of major chemical constituents of *E. cevuga* varied with the location of plant. Chemical compositions of essential oil for the *Etlingera* species were found different in different parts of the same plant (Jaafar et al., 2007; Khaleghi et al., 2012a, b). Many reports demonstrated that the fragrance and chemical composition of essential oils can vary based on geography (soil type, climate, altitude, amount of water available, harvesting season) and method of preparation

Table 2. Yield and percentage of major chemical component(s) identified in *Etlingera* species.

Etlingera Species	Plant Source	Plant Part[a]	Yield (%)	Major Constituent	% Composition	Reference
E. brevilabrum	Borneo	R	0.28	Elemicin -	35.6	Vairappan et al. (2012)
				Methyl isoeugenol	19.2	
				β-Farnesene	10.7	
	Malaysia	L	0.24	β-pinene	52.6	Mahdavi et al. (2012)
				α-thujene	28.6	
				o-cymene	7.8	
		S	0.07	limonene	28.6	
				β-pinene	21.6	
				α-thujene	13.9	
		R	0.03	Eucalypto	27.6	
				β-pinene	13.4	
				Caryophyllene oxide	10.5	
				α-thujene	10.1	
E. cevuga	Malaysia	L	0.03	α-thujene	38.1	Mahdavi et al. (2016)
				p-cymen-7-ol	8.0	
		Stolon	0.016	β-pinene	30.6	
				p-cymen-7-ol	25.0	
		S	0.011	δ-3-carene		
				α-thujene	17.7	
		R	0.018	Perilla aldehyde	19.6	
				Bornyl acetate	17.6	
	Tahiti Island	R	NM	Methyl eugenol	47.4	Vahirua-Lechat et al. (2010)
				(Z)- and (E)-methyl isoeugenol	18.8	
		L	0.48-1.94	Methyl eugenol	40.9–45.7	
				(E)-methyl isoeugenol	8.6–16.5	
				α-pinene	6.9-11.6	
				β-pinene	5.6-10.3	
E. coccinea	Borneo	R	0.3	Borneol	28.3	Vairappan et al. (2012)
				L-calamenene	18	
E. elatior	Borneo	R	0.38	1-Dodecanol	46.2	Bhuiyan et al. (2010)
				Cyclodecanol	34.1	

Table 2. Contd.

Malaysia	L	86 mg/100 g	(E)-farnesene	13.6	Chiang et al. (2010)
			(E)-caryophyllene	8.56	
Malaysia	L	0.0735	(E)-ß-farnesene	27.9	Jaafar et al. (2007)
			ß–pinene	19.7	
			caryophyllene	15.36	
	S	0.0029	1,1-dodecanediol diacetate	34.26	
			(E)-5-dodecene	26.99	
			1,1- dodecanediol diacetate	24.38	
	F	0.0334	cyclododecane	40.32	
			α-pinene	6.3	
	R	0.0021	1,1- dodecanediol diacetate	47.28	
			cyclododecane	34.45	
Malaysia	WP	NM	β-pinene	24.92	Abdelwahab et al. (2010)
			1-dodecene	24.31	
			Bicyclo[3.1.1]hept-2-ene, 2,6,6-trimethyl	11.59	
Malaysia	L	0.7	myrcene	13.5	Wong et al. (2010)
			a-humulene	11.8	
			b-caryophyllene	10.7	
	R	0.4	Camphene	18	Wong et al. (2010)
			b-pinene	16.9	
			bornyl acetate	9.2	
Malaysia	F	NM	dodecanal	16.87	Susanti et al. (2013)
			n-dodecyl acetate	16.4	
			1-hexadecanol	16.34	
			cis-9-tetradecen-1-ol	16.29	
			1-decanol	16.27	
Malaysia	L-6[b]	0.1	2-cyclohexen-1-one	93.42	Abdelmageed et al. (2011a)
	L-24	0.1	β-pinene	39.1	
			1-Dodecanol	16.03	
	L-48	0.16	β-pinene	37.3	
			1,6,10-Dodecatriene	-	
			1-Dodecanol	10.9	
	L-72	0.14	1-Dodecanol	42.3	
			Dodecanal	19.21	

Table 2. Contd.

				β-pinene	8.16
		PS-6	0	-	-
		PS-24	0.013	2-Tridecanone	51.5
				1-Tridecyne	12.18
		PS-48	0	-	-
		PS-72	0.007	Dodecanal	22.43
		F-6	0	-	-
		F-24	0.1	-	-
		F-48	0.08	1-Dodecanol	54.48
				Dodecanal	16.53
				Undecanoic acid	10.1
		F-72	0.1	1-Dodecanol	53.12
				Dodecanal	17.27
				n-Decanoic acid	12.33
		R-24	0.047	1-Dodecene	41.6
				Dodecanal	32.9
				1-Dodecanol	48.15
				Dodecanal	25.99
		R-48	0.013	1-Dodecanol	63.64
				Dodecanal	17.01
		R-72	0.02	1-Dodecanol	54.3
				Dodecanal	20.17
				1-Tetradecanol	7.81
	Brazil	F	0.9	Dodecanol	42.5
				α-pinene	22.2
				Dodecanal	14.5
		P	0.1	Dodecanol	34.6
				caryophyllene oxide	22.5
				Dodecanal	21.5
				α-pinene	6.3
	Indonesia	F		1-dodecanol	13.82
				dodecanal	12.10
				17-pentatriacontene	10.52
E. elatior vai Thai queen	Malaysia	L	0.9	a-pinene	24.4
				dodecanol	18.9

Zoghbi and Andrade (2005)

Maimulyanti and Prihadi (2015)

Wong et al. (2010)

Table 2. Contd.

				dodecanal	15.9	
		R	0.6	camphene	15.1	
				dodecanol	12.9	
				dodecanal	10.6	
E. fimbriobractata	Drunei Darussalam	L	0.126	β-pinene	67.8	Ud-Daula et al. (2016)
				α-pinene	17.1	
		AS	0.031	1,8-cineole	37.2	
				β-pinene	18.2	
		BS	0.031	decanal	27.5	
				1,8-cineole	18.7	
				β-pinene	10.6	
		R	0.018	decanal	34.4	
				β-pinene	10	
E. fulgens	Malaysia	L	133 mg/100 g	dodecyl acetate	21.6	Chiang et al. (2010)
				pentadecanol	14.1	
	Malaysia	L	Very Little Amount	(Z)-9-Hexadecen-1-ol	15.04	Khaleghi et al. (2012b)
				cyclotetradecane	8.93	
				oleyl alcohol	8.72	
				(Z)-11-hexadecen-1-ol acetate	7.74	
		S	0.0173	n-dodecyl acetate	16.62	
				cyclodecane	12.22	
				(Z)-9-hexadecen-1-ol	7.21%	
		R	0.0069	n-dodecyl acetate	16.28	
				cyclotetradecane	13.48	
				cyclododecane	11.3	
				cadinene	9.71	
		F	0.0493	n-dodecyl acetate	25.71	
				cyclododecane	22.91	
		F		cyclodecane	9.43	
		P	0.0213	(E)-2-tetradecene	26.24	
				cyclododecane	16.21	
				n-dodecyl acetate	15.18%	
		WP	0.0143	n-dodecyl acetate	18.59	

Table 2. Contd.

				cyclododecane	8.61	
				cyclotetradecane	6.16	
		L	0.2	α-phellandrene	12.1	
				diprene	11	
		S	0.03	1,8-cineole	16.9	
				α-phellandrene	11.5	
				β-trans-ocimene	7.9	
E. hornstedia	Malaysia	R	0.04	1,8-cineole	17.4	Yahya et al. (2010)
				α-phellandrene	9.5	
				1S-α-pinene	9.4	
		WP	0.07	β-pinene	13	
				p-menth-1-en-8-ol	8.5	
				α-pinene	8.1	
				α-phellandrene	8.1	
		L	1.15	eucalyptol	20.02	
				β-pinene	15.7	
				α-pinene	11.76	
	India			linalool	9.48	Kithan and Daiho (2014)
				methyl chevicol	49.93	
E. linguiformis		R	1.4	methyl eugenol	32.3	
				ß-pinene	13.46	
				Asarone	10.5	
		L	1.15	eucalyptol	39.7	
	Bangladesh			β-pinene	13.3	Bhuiyan et al. (2010)
		R	2.4	methyl chevicol	49.9	
				methyl eugenol	32.3	
				(E)-methyl isoeugenol	37.7	
E. littoralis	Malaysia	L	0.2	b-pinene	30.4	Wong et al. (2010)
				b-phellandrene	8.6	
		R	0.2	(E)-methyl isoeugenol	58.1	
E. maingayi	Malaysia	L	1317 mg/100 g	dodecanoic acid	44.6	Chiang et al. (2010)
				decanoic acid	42.6	

Table 2. Contd.

E. megalocheilos	Borneo	R	0.25	Aromadendrene oxide Terpineol oxide Aromadendrene	24.8 13 8.9	Edris (2007) and Vairappan et al. (2012)
E. pavieana	Thailand	R	0.07	trans-anethole p-anisaldehyde d-cadinene (7.2%).	48.6 13.8 7.2	Tachai et al. (2014)
E. punicea	Thailand	R	NM	Methyl chavicol	95.73	Tadtong et al. (2009)
E. pyramidosphaera	Borneo	R	0.45	Lauryl acetate 1-Tetradecano 1-Dodecanol Lauricaldehyde	29.6 20.2 28 11.2	Vairappan et al. (2012)
E. rubrostriata	Malaysia	L	39 mg/100 g	dodecyl acetate dodecanoic acid	6.01 4.41	Chiang et al. (2010)
E. sphaerocephala	Malaysia	L	0.17	δ-cadinene nerolidol	12.49 9.21	Jani et al. (2011)
		S	0.02	nerolidol limonene α-phellandrene	7.64 7.14 6.2	
		R	0.03	isoeugenol α-pinene β-pinene	9.9 9.23 8.4	
E. Sphaerocephala var. grandiflora	Malaysia	L	0.17	α-phellandrene diprene	12.3 10.3	Yahya et al. (2010)
		S	0.02	1,8-cineole α-phellandrene 1S-α-pinene	17.4 9.7 9.5	
		R	0.3	1,8-cineole α-phellandrene β-trans-ocimene	16.8 12.7 8.9	

Table 2. Contd.

		WP	0.05	β-pinene α-pinene p-menth-1-en-8-ol α-phellandrene	12.2 8.6 8.5 8.5	
E. sayapensis	Malaysia	L S R		Carvone cis-carveol a-terpineol linalool formate Linalool formate Eugenol	21.35 13.49 39.86 30.55 25.47 11.84	Mahdavi et al. (2017)
E. venusta	Malaysia	L	0.031	α-Terpineol linalool 1-methyl-3-(1-methylethenyl) cyclohexene (9.7 %),	13.4 10.9 9.7	Khaleghi et al. (2012a)
		AS	0.001	1-methyl-3-(1-methylethenyl) cyclohexene α-terpineol α-pinene	8.3 8.1 7.9	
		R	0.006	dodecanoic acid (37.8 %), cyclododecane (Z)-13-octadecen-1-ol acetate	37.8 15.8 10.9	
		F	0.014	Cyclododecane dodecanoic acid cyclododecanol	17.1 15.6 11.5	
		P	0.005	(E)-2-tetradecene cyclododecane cyclotetradecane	28.5 17.6 15.7	
		WP	0.004	Carene dodecanoic acid n-hexadecanoic acid	8 6.6 6.1	
E. yunnanensis	Vietnam	L	0.25	germacrene D β-pinene α-amorphene	19.2 11.6 11.2	Guo et al. (2015)

Table 2. Contd.

S	0.20	β-pinene	23.7
		1,8-cineole	11.0
		α-pinene	9.6
Ro	0.31	β-pinene	31.9
		α-pinene	13.7
		1,8-cineole	9.4
R	0.14	estragole	65.20
		β-caryophyllene	6.4
		1,8-cineole	6.5

[a] L - Leaf, S - stem, PS - pseudostem, R - rhizome, Ro-root, F - flower, P - peduncle, WP - whole plant. [b] drying time in hours, NM: not mentioned.

(Luna, 2002). Essential oil concentration is also fluctuated during its different stages of ripening (Wannes et al., 2009). These variations in chemical composition lead to the notion of chemotypes, which are generally defined as a chemically distinct entity in a plant with different compositions of the secondary metabolites(Djilani and Dicko, 2012). An essential oil chemotype can distinguish *Etlingera* of different origins, as well as seasonal variations throughout the vegetative cycle of plants.

Because chemotypes are defined only by the most abundant secondary metabolite, *Etlingera brevilabrum* may be assigned to two different chemotypes, depending on dominant component of the essential oil; elemicin and eucalyptol. Such chemotypes may be indicated as *E. brevilabrum* ct. elemicin, or *E. brevilabrum* ct. eucalyptol, although such indication has no taxonomic standing. Individuals of these chemotypes have vastly different chemical profiles, varying in the abundance of the kind of the next most abundant chemical. This can be a very qualitative assessment of an individual's chemical profile, under which may be hiding significant chemical diversity. Plant genotype is another important factor that can also affect the changes of the chemical composition of essential oils (Djilani and Dicko, 2012). Therefore, genetic and epigenetic factors influence the biochemical synthesis of essential oils in a given plant resulting in different chemical compositions and therapeutic activities.

Essential oil compositions of *Etlingera* were also varied on drying condition of the plants (Abdelmageed et al., 2011a; Mahdavi et al., 2012). The major constituents found in fresh rhizomes of *E. brevilabrum* were elemicin (35.6%) and methyl isoeugenol (19.2%) whereas eucalyptol (27.6%) and β-pinene (13.4%) were abundant in air-dried rhizomes of same species collected from Sabah, Malaysia (Mahdavi et al., 2012). Abdelmageed et al. (2011a) reported that fresh and dried of same plants parts (leaves, pseudostems, flower and rhizomes) of *E. elatior* produced different oil composition with increasing drying time. The author also found that different drying periods had an effect on the percentage of the main compounds and resulted in slight losses in volatile compounds compared with the fresh herb. Some authors also reported that the concentrations of various volatile substances increased after air drying (Díaz-Maroto et al., 2002; Faridah et al., 2010). This might be due to chemical transformation (breakdown of glycosylated forms, dehydration reactions, or oxidation reactions) or loss of compounds during drying process or due to the rupture of the plant cells in which the volatiles are stored.

The major constituents of *Etlingera* leaf essential oil are 1,8-cineole, 1-dodecanol, dodecyl acetate, elemicin, eucalyptol, methyl eugenol, (E)-methyl isoeugenol, α-pinene, β-pinene, and thujene. Stem of the plants contains mainly (E)-5-dodecene, 1,1-dodecanediol diacetate, 1,8-cineole, limonene, β-pinene and α-phellandrene, whereas cyclododecane, 1,1- dodecanediol diacetate, dodecanal, n-dodecyl acetate, 1-decanol and 1-hexadecanol are dominant in flowers. The main essential oil components in the rhizome are aromadendrene oxide, trans-anethole, 1,8-cineole, 1-dodecanol, n-dodecyl acetate, decanoic acid methyl chevicol, lauryl acetate, (E)-methyl isoeugenol as well as other compounds including cyclododecane, dodecanoic acid, linalool, α-pinene, β-pinene and α-

phellandrene. Cyclododecane, dodecanol, dodecanal, (E)-2-tetradecene and caryophyllene oxide are the dominant component in peduncle essential oil and it is followed by α-pinene n-dodecyl acetate and cyclotetradecane. Analysis of whole plant essential oil composition showed that caryophyllene oxide n-dodecyl acetate, α-pinene, β-pinene, nerolidol, phellandrene, cyclododecane and 1-dodecene are abundant in *Etlingera* species.

Classification of *Etlingera* essential oil

Essential oil compounds can be classified into three main categories: terpenes (monoterpene hydrocarbons and sesquiterpene hydrocarbons), terpenoids (oxygenated monoterpenes and oxygenated sesquiterpenes) and non-terpenic compounds. Terpenes are hydrocarbons derived from five carbon atoms attached to eight hydrogen atoms regarded as isoprene units (C_5H_8). They form structurally and functionally diverse classes of organic compounds. They are synthesized in the cytoplasm of plant cells, where two molecules of acetic acid are combined to form mevalonic acid ($C_6H_{12}O_4$) (Dhifi et al., 2016). Terpenes are usually grouped according to the number of isoprene units in the molecule, which can be rearranged into cyclic structures by cyclases, thus forming monocyclic or bicyclic structures. Monoterpenes ($C_{10}H_{16}$) contain two isoprene units; three sesquiterpenes ($C_{15}H_{24}$); four diterpenes ($C_{20}H_{32}$); six triterpenes ($C_{30}H_{48}$); and eight tetraterpenes ($C_{40}H_{64}$). *Etlingera* essential oils are abundant with monoterpenes and sesquiterpenes but longer chains also exist. Themonoterpene hydrocarbons found in *Etlingera* essential oils include α-pinene, β-pinene, α-phellandrene and limonene as seen in Table 3. On the other hand, cadinene, caryophyllene, (E)-farnesene and tetradecadiene are the major sesquiterpene hydrocarbons observed in *Etlingera* oil.

Terpenoids can be thought of as modified terpenes which undergo biochemical modifications *via* enzymes that add oxygen molecules and move or remove methyl groups (Williams et al., 1989). It can be sub-divided according to the number of isoprene units: monoterpenoids (2 isoprene units), and sesquiterpenoids (3 isoprene units) etc. The principle monoterpenoids found in *Etlingera* essential oil are 1,8-Cineole, 1,1-dodecanediol diacetate, eucalyptol, linalool and α-terpineol while caryophyllene oxide and nerolidol are major sesquiterpenoids.

Terpenic compounds are dominant in all parts of *Etlingera* species except *E. fulgens* and *Etlingera venusta.* Monoterpene hydrocarbon was found in greater percentage in essential oil of *Etlingera* species followed by oxygenated monoterpene, sesquiterpene and oxygenated sesquiterpene. Among different plant parts (leaves, stems, flowers, peduncle and rhizomes), leaves are found to contain highest proportion of monoterpene hydrocarbon than any other parts.

Phenylpropanoids (non-terpenic compounds) are a wide-spread class of plant-derived natural products synthesized from the amino acid precursor, phenylalanine. They serve as essential components of a number of structural polymers, provide protection from ultraviolet light, defend against herbivores and microbial attack and acts as signaling molecules (Korkina, 2007).

In *Etlingera* essential oils, most thoroughly studied phenylpropanoids are elemicin, eugenol, methyl eugenol, (*E*)-methyl isoeugenol and methyl chavicol. Out of 16 different *Etlingera* species, they were identified only in 8 species such as *Etlingera bravlabrum* (R), *E. cevuga* (L,R*), Etlingera linguiformis* (R), *E. linguiformis* (R) *Etlingera littoralis* (L,R), *E. pavieana* (R*), E. punica* (R), *Etlingera sphaerocephala* (R)*, E. venusta* (L) (Bhuiyan et al., 2010; Khaleghi et al., 2012a; Tachai et al., 2014; Vahirua-Lechat et al., 2010; Vairappan et al., 2012; Wong et al., 2010; Yahya et al., 2010).

Various other groups of non-terpenic compounds such as alcohol, aldehyde, ketone, esters, carboxylic acid and hydrocarbon were also observed in *Etlingera* species. Examples of this group of compounds are dodecanoic acid, decanoic acid, dodecyl acetate, dodecanol, cyclododecane and (E)-2-tetradecene. *E. fulgen, Etlingera fimbriobracteata* and *E.venusta* are characterized by high proportion of non-terpene hydrocarbon, alcohol and esters (Chiang et al., 2010; Khaleghi et al., 2012a; Ud-Daula et al., 2016).

However, *E. bravilabrum, E. cevuga, E. coccinea, E. elatior* (S, R)*, E. linguiformis, E. littoralis, E. pyramidosphaera* and *E. Sphaerocephala var. grandiflora* are devoid of these compounds (Jaafar et al., 2007; Kithan and Daiho, 2014; Mahdavi et al., 2016; Vahirua-Lechat et al., 2010; Vairappan et al., 2012).

ANTIMICROBIAL ACTIVITY

Test of antimicrobial assays of essential oils in *Etlingera*

An overview of the literature reporting antimicrobial assays of *Etlingera* EOs is presented in Table 4.

Determination of antimicrobial activity of *Etlingera* EOs was generally done by agar disk diffusion, agar dilution and broth micro dilution method. Screening of antimicrobial properties of EOs is generally by the agar disk diffusion method, where a sterile paper disk impregnated with EO is laid on top of an inoculated agar plate.

This method is not considered an ideal method for essential oils, normally used as a preliminary check for antimicrobial activity prior to more detailed studies. Agar dilution or broth microdilution methods are most widely used methods to determine minimum inhibitory

Table 3. Classes of major *Etlingera* essential oils compounds and their bioactivities in different plant parts.

Chemical class	Chemical subclass	Plant parts	Subclass content			Major EO compound	Bioactivity	References for bioactivity
			Highest (%)	Lowest (%)	Absent			
Terpene	Monoterpene hydrocarbons[a] $C_{10}H_{16}$	L	[a]e.b(89.8)	es(7.1)	ef, em, er	β-pinene, α-thujene, myrcene	Stimulant, antiviral, antitumor, decongestant, antibacterial, hepatoprotective	Bakkali et al. (2008) Kalemba and Kunicka (2003)
		S	eb(66.8)	e sp(26.5)	ef	α-pinene, β-pinene, α-phellandrene, limonene,		
		F	ee(47.5)	ev(8.3)	ef	β-pinene,		
		P	ee(6.3)	-	ef,ev	α-pinene, β-pinene		
		R	esv(56)	epv(2.1)	eb(1A), ec, eco, elt, ef, eme, epy, ev	α-pinene, β-pinene, α-phellandrene		
		WP	esv(53.3)	ev(21.9)	ef	α-pinene, β-pinene, phellandrene,		
	Sesquiterpene hydrocarbons[b] $C_{15}H_{24}$	L	ee(45)	eh(0.94)	eb(1B), ec, ef, elt, em, er, egr, ev	(E)-farnesene, dodecyl acetate	Aphicide, Antiviral, Local anaesthetic	Edris (2007) and Nerio et al. (2010)
		S	e sp(13.7)	egr(3.5)	ef	caryophyllene		
		F	ee(9.9)	-	ef,ev	Tetradecadiene		
		P	-	-	ee, ef, ev	-		
		R	eb-1B(47)	eh(3.45)	ec,el, elt,epu, epy, ev,	cadinene, caryophyllene		
		WP	ee(4.5)	egr(3.3)	ev	n-dodecyl acetate		
Terpenoid	Oxygenated monoterpenes	L	ev(58)	esp(6.5)	ec,ef,elt,em,	1,8-Cineole, α-Terpineol, linalool, eucalyptol	Antimicrobial, Antioxidant, insecticidal	Edris (2007) Kalemba and Kunicka (2003) and Nerio et al. (2010)
		S	ee(54.3)	esp(11.5)	-	1,8-cineole, 1,1-dodecanediol diacetate		
		F	ee(30.8)	ev(22.8)	ef	α-Terpineol		
		P	-	-	ee,ef,ev	-		
		R	epy(80.1)	epv(0.6)	ec,ef,elt,epu	1,8-cineole, Borneol		
		WP	ev(31.2)	egr(22.6)	ee,ef	α-Terpineol		
	Oxygenated sesquiterpene[c]	L	esp(17.7)	eh(0.13)	eb(1B),ec,ef,el,elt,em, esv	caryophyllene oxide	Antimicrobial, analgesic, anti-inflammatory	Kalemba and Kunicka (2003)
		S	esp(10.1)	eh(0.32)	ef, egr, ev	nerolidol		
		F	-	-	ee, ef, ev	caryophyllene oxide		
		P	-	-	ee, ef, ev	caryophyllene oxide		
		R	eme(28.6)	eh(0.7)	eb(1A), ec, ee, ef, el, elt, epu, esv, ev	nerolidol		
		WP	ev(7.7)	-	ee, ef, esv	nerolidol, caryophyllene oxide		
Non-terpene	alcohol, aldehyde, ketone, esters, carboxylic acid, hydrocarbon	L	eh(0.29)	ef(85.78)	eb(1B), el, egr	dodecanoic acid ,decanoic acid, dodecyl acetate, (E)-methyl isoeugenol, methyl eugenol	Antimicrobial, Antioxidant, chelating agent	Edris (2007) Kalemba and Kunicka (2003)
		S	ef(65.84)	eh(0.13)	eb(1B), ee, esv	(E)-5-dodecene, n-dodecyl acetate		
		F	ef(84.06)	ev(58.5)	-	Cyclododecane, dodecanol		

Table 3. Contd.

and phenylpropanoids					
	P	ev(96.4)	ee(59.3)	-	(E)-2-tetradecene, dodecanol, dodecanal, cyclododecane
	R	epu(95.8)	eh(0.34)	eb(1B), eco, ee, eme, epy, esv	methyl chavicol ,1-Dodecanol, methyl isoeugenol, eugenol, Dodecanal, trans-anethole
	WP		eh(0.09)	esv	cyclododecane, 1-dodecene

'-' indicates not mentioned. [a]eb: *E. brevilabrum*, ec: *E. cevuga*, eco: *E. coccinea*, ee: *E. elatior*, eet: *E. elatior* vai Thai queen, ef: *E. fulgens*, eh: *E. hornstedia*, elt: *E. littoralis*, el: *E. linguiformis*, eme: *E. megalocheilos*, em: *E. maingayi*, ep: *E. pavieana*, epu: *E. punicea*, epy: *E. pyramidosphaera*, er: *E. rubrostriat*, es: *E. Sphaerocephala*, esv: *E. Sphaerocephala* var. grandiflora, ev: *E. venusta*

concentration (MIC) (Rios et al., 1988).

MIC is generally regarded as the most important parameter to confirm resistance of micro-organisms to an antimicrobial agent and also to monitor the activity of new antimicrobial agents (Andrews, 2001). In addition to MIC, some author also stated minimum bactericidal concentrations (MBCs) and bacteriostatic concentration (Susanti et al., 2013).

ANTIMICROBIAL EFFECTS OF *ETLINGERA* EOS

Researchers expedite their relentless venture to discover and synthesize new antimicrobial agents due to emergence of antibiotic resistance, increased population with lower immunity, undesirable effects of current drugs, etc. Plant molecules are well known for their antimicrobial properties; especially many essential oils and their components are known to possess significant activity against wide range of microorganisms (Bakkali et al., 2008; Bassolé and Juliani, 2012). Therefore, detail study of the plant EOs could be helpful to identify novel drugs and targets for controlling the infectious diseases worldwide. The antibacterial activity of *Etlingera* essential oils against gram-positive and gram-negative bacteria were reported in few studies and obtained results is presented in Table 4.

Tadtong et al. (2009) indicated antimicrobial effect of *E. punicea* rhizome essential oil on some pathogenic bacteria, particularly *Staphylococcus aureus*, *Escherichia coli* and *Salmonella albany,* and also possesses fungicidal activity against *Candida albicans*. Essential oils from rhizomes of *E. pyramidosphaera, E. megalocheilos, E. coccinea*, *E. elatior* and *E. brevilabrum* were tested against four strains of clinical bacteria (*S. aureus*, *Staphylococcus* sp., *Streptococcus pyrogenes* and *Salmonella enteritidis*), where *E. megalocheilos, E. coccinea* and *E. elatior* inhibited all the four tested bacteria with MIC values of less than 10 µg/mL, and the other two *Etlingera* species, *E. pyramidosphaera* and *E. brevilabrum* also showed interesting selective inhibition against *S. aureus* and *S. pyrogenes*, with MIC values ranging from 40.0 to 120.0 µg/mL (Vairappan et al., 2012). The author also claimed that the dominant presence of borneol (28.3%) and the availability of camphor (3.8%) could possibly be responsible for potent antibacterial activity.

In the study of Chiang et al. (2010), the oils from leaves of *E. elatior*, *E. fulgens* and *E. maingayi*, and *E. rubrostriata* showed inhibition against gram-positive bacteria of *B. cereus*, *M. luteus* and *S. aureus* with MIC values ranging from 6.3-100 mg/ml and ranking was in the order: *E. maingayi* > *E. rubrostriata* > *E. elatior* > *E. fulgens*. Of the gram-positive bacteria, *M. luteus* was the most susceptible with all four *Etlingera* species having MIC of 6.3 mg/ml, whereas no activity was observed on Gram-negative bacteria of *E. coli*, *P. aeruginosa* and *S. choleraesuis* (Chiang et al., 2010).

Dodecanoic (lauric) acid and decanoic (capric) acid which constitute more than 87% of *E. maingayi* oil content might be responsible for strong antibacterial activity. The oils (leaves, aerial stems, basal and rhizomes) from *E. fimbriobracteata* exhibited moderate to potent broad-spectrum antimicrobial activity against gram positive (*Bacillus subtilis, Bacillus spizizenii*. and *S. aureus*), gram negative (*E. coli*) and fungi (*C. albicans* and *Saccharomyces cerevisiae*) (Ud-Daula et al., 2016). (Reference) *E. sayapensis* essential oils also displayed a broad spectrum of antimicrobial activity against gram positive (*B. subtilis, B. thuringiensis, MRSA, S. aureus*), gram-

Table 4. Antimicrobial activity of essential oils of *Etlingera* species.

Etlingera Species	Plant Part	Microorganism	Test method	Activity	Reference
E. punicea (20 µl/6 mm disk)	R	*Staphylococcus aureus*	Disk Diffusion	0.87 mm	Tadtong et al. (2009)
		Escherichia coli		0.78 mm	
		Pseudomonas aeruginosa		-	
		Salmonella albany		0.82 mm	
		Candida albicans		1.18 mm	
E. brevilabrum	L	*MRSA*	Disk Diffusion	8.1 mm	Mahdavi et al. (2016, 2018)
		Staphylococcus aureus		12.6 mm	
		Bacillus subtilis		-	
		Bacillus thuringiensis		-	
		Escherichia coli		-	
		Proteus vulgaris		10.3 mm	
		Salmonella typhimurium		-	
		Proteus mirabilis		16.4 mm	
		Pseudomonas aeruginosa		-	
	S	*MRSA*		10 mm	
		Staphylococcus aureus		11.2 mm	
		Bacillus subtilis		-	
		Bacillus thuringiensis		-	
		Escherichia coli		13.5 mm	
		Proteus vulgaris		9.5 mm	
		Salmonella typhimurium		-	
		Proteus mirabilis		-	
		Pseudomonas aeruginosa		-	
	R	*MRSA*		12.6 mm	
		Staphylococcus aureus		17.7 mm	
		Bacillus subtilis		-	
		Bacillus thuringiensis		-	
		Escherichia coli		-	
		Proteus vulgaris		-	
		Salmonella typhimurium		-	
		Proteus mirabilis		13.2 mm	
		Pseudomonas aeruginosa		-	

Table 4. Contd.

E. coccinea	R	*Staphylococcus aureus* *Staphylococcus sp.,* *Streptococcus pyrogenes,* *Salmonella enteritidis*		5 µg/ml 10µg/ml 8 µg/ml 8 µg/ml	Vairappan et al. (2012)
E. pyramidosphaera	R	*Staphylococcus aureus* *Staphylococcus sp.,* *Streptococcus pyrogenes,* *Salmonella enteritidis*		90 µg/ml - 120 µg/ml -	
E. megalocheilos	R	*Staphylococcus aureus* *Staphylococcus sp.,* *Streptococcus pyrogenes,* *Salmonella enteritidis*	Agar dilution method	8 µg/ml 8 µg/ml 6 µg/ml 9 µg/ml	
E. brevilabrum	R	*Staphylococcus aureus* *Staphylococcus sp.,* *Streptococcus pyrogenes,* *Salmonella enteritidis*		40 µg/ml - 40 µg/ml -	
E. elatior	R	*Staphylococcus aureus* *Staphylococcus sp.,* *Streptococcus pyrogenes,* *Salmonella enteritidis*		80 µg/ml 90 µg/ml 75 µg/ml 60 µg/ml	
E. elatior	L	*Bacillus cereus* *Micrococcus luteus* *Staphylococcus aureus*	Disk Diffusion	25 mg/ml 6.3 mg/ml 50 mg/ml	
E. fulgens	L	*Bacillus cereus* *Micrococcus luteus* *Staphylococcus aureus*		25 mg/ml 6.3 mg/ml 100 mg/ml	Chiang et al. (2010)
E. maingayi	L	*Bacillus cereus* *Micrococcus luteus* *Staphylococcus aureus*	Disk Diffusion	6.3 mg/ml 6.3 mg/ml 12.5 mg/ml	
E. rubrostriata	L	*Bacillus cereus* *Micrococcus luteus* *Staphylococcus aureus*	Disk Diffusion	12.5 mg/ml 6.3 mg/ml 50 mg/ml	
Etlingera elatior (5 µl/6 mm disk)	WP	*Staphylococcus aureus* *Pseudomonas aeruginosa*	Disk Diffusion	- -	Abdelwahab et al. (2010)

Table 4. Contd.

		Salmonella choleraesuis		-	
		Bacillus subtilis		-	
E. elatior (10 µl/6 mm disk)	F	*Staphylococcus aureus*	Disk Diffusion / Microdilution	12 mm, MIC 33.3 µl/ml, MBC 33.3 µl/ml,	Susanti et al. (2013)
		Bacillus cereus		11.5 mm, MIC 33.3 µl/ml, MBC 33.3 µl/ml,	
		Pseudomonas aeruginosa		-	
		Escherichia coli		-	
		Candida albicans		10 mm, MIC 0.4 µl/ml, MBC 11.1 µl/ml	
		Candida neoformans		20 mm, MIC 0.05 µl/ml, MBC 12 µl/ml	
E. fimbrobractata	L	*Pseudomonas aeruginosa*	Disk Diffusion / Microdilution	-	Ud-Daula et al. (2016)
		Escherichia coli		8 mm, MIC 625 µg/ml	
		Bacillus subtilis		12.33 mm, MIC 78 µg/ml	
		Bacillus spizizenii		9.5 mm, MIC 156 µg/ml	
		Staphylococcus aureus		9.17 mm, MIC 156 µg/ml	
		Candida albicans		17 mm, MIC 78µg/ml	
		Saccharomyces cerevisiae		34.33 mm, MIC 78 µg/ml	
	AS	*Pseudomonas aeruginosa*	Disk Diffusion /Microdilution	-	
		Escherichia coli		10.28 mm, MIC 312.5 µg/ml	
		Bacillus subtilis		15.0 mm, MIC 39 µg/ml	
		Bacillus spizizenii		13.33 mm, MIC 39 µg/ml	
		Staphylococcus aureus		12.0 mm, MIC 156 µg/ml	
		Candida albicans		11.33 mm, MIC 156 µg/ml	
		Saccharomyces cerevisiae		29.83 mm, MIC 39 µg/ml	
	BS	*Pseudomonas aeruginosa*	Disk Diffusion /Microdilution	-	
		Escherichia coli		8mm, MIC 156 µg/ml	
		Bacillus subtilis		12.33 mm, MIC 19.5µg/ml	
		Bacillus spizizenii		9.5 mm, MIC 19.5µg/ml	
		Staphylococcus aureus		9.17 mm, MIC 78µg/ml	
		Candida albicans		17 mm, MIC 78µg/ml	
		Saccharomyces cerevisiae		34.33 mm, MIC 9.7µg/ml	
	R	*Pseudomonas aeruginosa*	Disk Diffusion /Microdilution	-	
		Escherichia coli		12 mm, MIC 156 µg/ml	
		Bacillus subtilis		20.2 mm, MIC 19.5 µg/ml	
		Bacillus spizizenii		16.67mm, MIC 19.5µg/ml	

Table 4. Contd.

		Staphylococcus aureus		13.67 mm, MIC 39 µg/ml	
		Candida albicans		26 mm, MIC 39 µg/ml	
		Saccharomyces cerevisiae		58.67 mm, MIC 2.4 µg/ml	
E. sayapensis	L	*Bacillus subtilis*	Microdilution	0.78 MIC mg/ml	Mahdavi et al. (2017)
		Bacillus thuringiensis		5.29 MIC mg/m	
		Enterococcus faecalis		-	
		MRSA		8.33 MIC mg/m	
		Staphylococcus aureus		0.78 MIC mg/m	
		Staphylococcus epidermidis		4.16 MIC mg/m	
		Aeromonas hydrophila		5.21 MIC mg/m	
		Enterobacter aerogenes		2.6 MIC mg/m	
		Escherichia coli		1.56 MIC mg/m	
		Proteus mirabilis		20.83 MIC mg/m	
		Proteus vulgaris		-	
		Pseudomonas aeruginosa		-	
		Salmonella typhimurium		-	
		Serratia marcescens		5.21 MIC mg/m	
		Shigella sonnei		1.56 MIC mg/m	
		Vibrio parahaemolyticus		6.25 MIC mg/m	
		Candida albicans		4.16 MIC mg/m	
		Candida parapsilosis		3.64 MIC mg/m	
	R	*Bacillus subtilis*		0.78 MIC mg/m	
		Bacillus thuringiensis		4.16 MIC mg/m	
		Enterococcus faecalis		-	
		MRSA		0.52 MIC mg/m	
		Staphylococcus aureus		1.56 MIC mg/m	
		Staphylococcus epidermidis		-	
		Aeromonas hydrophila		3.12 MIC mg/m	
		Enterobacter aerogenes		2.6 MIC mg/m	
		Escherichia coli		3.12 MIC mg/m	
		Proteus mirabilis		1.56 MIC mg/m	
		Proteus vulgaris		-	
		Pseudomonas aeruginosa		-	
		Salmonella typhimurium		-	

Table 4. Contd.

	Serratia marcescens	0.52 MIC mg/m
	Shigella sonnei	6.25 MIC mg/m
	Candida albicans	2.6 MIC mg/m
	Candida parapsilosis	0.52 MIC mg/m

'-' indicates no inhibition.

negative bacteria (*A. hydrophila, E. aerogenes, E. coli, P. mirabilis, S. marcescens, S. sonnei, V. parahaemolyticus*) and fungi (*C. albicans* and *C. parapsilosis*) (Mahdavi et al., 2017).

Abdulwahab et al. (2010) reported that essential oils from whole plant of *E. elatior* failed to inhibit gram positive (*B. subtilis)* and gram-negative (*S. choleraesuis* and *P. aeruginosa)* bacteria. However, Susanti et al. (2013) demonstrated the activity of *E. elatior* flowers essential oil against Gram positive bacteria of *S. aureus*, *Bacillus cereus* with zone of inhibition of 13 and 12.3 mm, respectively but not sensitive against *P. aeruginosa* and *E. coli*. The oil also showed potent inhibitory effects against fungi with mean inhibition zones 10 mm for *C. albicans* and 20 mm for *C. neoformans. C. neoformans* was found as the most susceptible among all four microorganisms with MIC value of 0.05 µL/mL. antifungal property of *Etlingera* is suspected to be associated with their high contents of monoterpene hydrocarbon and phenylpropanoids.

It has been generally reported that gram-negative bacteria are more resistant than gram-positive bacteria. Similar observations were also found in *Etlingera* species where Gram-negative bacteria (*E. coli* and *P. aeruginosa*) showed less susceptibility to the *Etlingera* essential oil than Gram positive bacteria (Abdelwahab et al., 2010; Susanti et al., 2013). Of the gram-negative bacteria, *P. aeruginosa* was found resistant to the action of essential oil. This could be due to highly restricted outer membrane of these bacteria that slows down the passage of essential oils, whereas lacking of outer membrane enables Gram positive bacteria to be more susceptible to *Etlingera* essential oils.

Mode of antimicrobial action

Different modes of action are involved in the antimicrobial activity of essential oils, because of the variability of quantity and chemical profiles of essential oil. Antimicrobial action of essential oils and their components may be attributed by any or a combination of six possible mechanisms which include: (1) disintegration of cytoplasmic membrane, (2) damage of membrane proteins (ATPases and others), (3) degradation of cell wall with the release of lipopolysaccharides, (4) leakage of ions and other cell content, (5) coagulation of cytoplasm and (6) inhibition of enzyme synthesis (Bakkali et al., 2008; Bouhdid et al., 2010; Burt, 2004). Until now, the mechanisms of antimicrobial action of *Etlingera* essential oil and their components are not demonstrated in any published paper. Precise mechanisms of antibacterial activity of few individual pure essential oil components from different genera were reported. Carvacrol and thymol are able to disintegrate the outer membrane, increasing membrane fluidity, which in turn increases the permeability of the cytoplasmic membrane to ATP (Ultee et al., 1999). Eugenol - a major component (approximately 85%) of clove oil binds with membrane protein of both gram positive and negative bacteria altering their structure and increases permeability. Farnesol, nerolidol and plaunotol has been regarded as a cause for the loss of potassium and sodium ions in *S. aureus* (Carson and Hammer, 2011). *p*-cymene, terpinen-4-ol, 1,8-cineole, terpinolene and γ-terpinene increase membrane fluidity that breaches membrane integrity and allows small intracellular components such as hydrogen, potassium and sodium to pass through the cell membrane and ultimately causes cell death.

AREAS OF FUTURE RESEARCH

All the literature in the last 15 years mainly focuses on the chemical composition of essential oil of *Etlingera* species. Only three literatures demonstrated the antimicrobial properties of essential oil in which two papers reported antifungal properties. Also, the oil from *E. punicea* and *E. elatior* showed potent antifungal properties than bacteria. However, some limitations have also been identified in the investigation of antimicrobial activity (only 2 species of fungi were used). Fungi are eukaryotic cells and

consequently most agents that are toxic to fungi are also toxic to the host, hence development of antifungal agents has lagged behind that of antibacterial agents. Fungi grow slowly and many of them have multicellular forms which complicates in developing new antifungal agents and in understanding the existing ones. The action of essential oil and their components against bacterial and fungal cell is not fully identified and is a focal area for future research. Thus, elucidation of mechanism of action of essential oil and their components would provide insights that may lead to identification of new antibiotic target and exploitation of novel biochemical pathways.

CONCLUSION

This review summarizes and characterizes the importance of essential oils obtained from different parts of *Etlingera* species which comprise diverse chemical constituents. The essential oil of *Etlingera* showed promising antimicrobial properties that could be an alternative source of synthetic antibiotics in order to combat emerging drug resistance. Since, essential oil and their components possess many important medicinal activities, seeking new drugs from aromatic and medicinal plant like *Etlingera* is crucial. Many studies have demonstrated the chemical composition of this plant but only few researchers have investigated the antimicrobial properties of essential oil. Thus, more studies are needed to understand and elucidate the mechanism of action of essential oil and their constituents.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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