# Chemical Composition, Antibacterial and Antioxidant Activities of Leaves Essential Oils from *Syzygium cumini* L., *Cupressus sempervirens* L. and *Lantana camara* L. from Egypt

Hosam O. Elansary<sup>1</sup>, Mohamed Z.M. Salem<sup>2</sup>, Nader A. Ashmawy<sup>3</sup> & Mohamed M. Yacout<sup>4</sup>

<sup>1</sup> Floriculture, Ornamental Horticulture and Garden Design Department, Faculty of Agriculture (El-Shatby), Alexandria University, Alexandria, Egypt

<sup>2</sup> Forestry and Wood Technology Department, Faculty of Agriculture (EL-Shatby), Alexandria University, Alexandria, Egypt

<sup>3</sup> Plant Pathology Department, Faculty of Agriculture (El-Shatby), Alexandria University, Alexandria, Egypt

<sup>4</sup> Genetics Department, Faculty of Agriculture (El-Shatby), Alexandria University, Alexandria, Egypt

Correspondences: Hosam O. Elansary, Floriculture, Ornamental Horticulture and Garden Design Department, Faculty of Agriculture (El-Shatby), Alexandria University, Alexandria, Egypt. Tel: 20-109-951-4043. E-mail: hosammail2003@yahoo.com / Mohamed Z. M. Salem, Forestry and Wood Technology Department, Faculty of Agriculture (EL-Shatby), Alexandria University, Alexandria, Egypt. Tel: 20-1045-6137. E-mail: zidan forest@yahoo.com

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# Abstract

Leaves essential oils of *Syzygium cumini* L. *Cupressus sempervirens* L. and *Lantana camara* L., from Alexandria, Egypt were analysed by Gas chromatography-mass spectrometry (GC-MS). The antibacterial and antioxidant activities were assessed for each oil using minimum inhibitory concentrations (MICs) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) methods; respectively. The main oil constitutes were  $\alpha$ -pinene (17.53%),  $\alpha$ -terpineol (16.67%) and alloocimene (13.55%) in *S. cumini*, trans-caryophyllene (15.57%) and  $\alpha$ -humulene (9.16%) in *L. camara* and cedrol (21.29%),  $\Delta^3$ -carene (17.85%) and  $\alpha$ -pinene (6.9%) in *C. Sempervirens*. The antibacterial activity was studied against some bacterial strains. The oil of *S. cumini* demonstrated strong inhibition activity against the tested bacterial strains. The total antioxidant activity (TAA %) was 11.13, 9.13, 3.13% for *S. cumini*, *L. camara* and *C. sempervirens*, respectively. Our work revealed that the studied leaf oils are new promising potential sources of antioxidants and antibacterial compounds and good future practical applications for human and plant health.

Keywords: Syzygium cumini, Cupressus sempervirens, Lantana camara, essential oil, antibacterial activity, antioxidant activity

# 1. Introduction

The essential oils from the leaves of plants and their therapeutic properties were known by Arabs in the middle ages who developed hydro-distillation (Bakkali et al., 2008). Nowadays, essential oils are well known for their cosmetic, pharmaceutical, agricultural and industrial applications. The main focus of pharmaceutical and agricultural companies in the Mediterranean countries now is the commercial production of known aromatic herbs like mint and basil (Edris et al., 2003), and neglecting the utilization of trees like *Syzygium cumini* (L.) Skeels. (*S. cumini*), *Cupressus sempervirens* L. (*C. sempervirens*) and shrubs like *Lantana camara* L. (*L. camara*). Additionally, these trees and shrubs are globally distributed and have massive diversity which may provide new sources of flavors, perfumes and remedies for aging as well as other medical and agricultural applications.

*S. cumini* or *Eugenia jambolana* Lam. is an evergreen tropical tree in the flowering plant belongs to the family Myrtaceae and a genus of 1000 species (Ayyanar & Subash-Babu, 2012). The tree has long been considered to have medicinal properties such as being used against dysentery and to treat inflammation, diabetes mellitus, constipation, leucorrhoea, stomachalgia, fever, gastropathy, trangury and dermopathy and to inhibit blood discharges in the faces (Bhandary et al., 1995; Shafi et al., 2002). The extracts from leaves, fruit, root-bark and stem-bark showed antifungal activity (Jabeen & Javaid, 2010). The seeds of *S. cumini* have anti-diabetic properties (Modi et al., 2010). The fruits and leaves extract (Ruan et al., 2008) have been reported to have antioxidant activity.

*C. sempervirens*, a medium-sized evergreen tree to 35 m belongs to the Family Cupressaceae, is a species of cypress native to the eastern Mediterranean region. The plant is known for its leaf oil "commercially known as Cypress oil" which is used to protect stored grains from insect infestation (Tapondjou et al., 2005). The cone oil of an Egyptian local cultivar showed antibacterial activity (Kassem et al., 1991) and antiviral activity (Amouroux et al., 1998). *C. sempervirens* leaf extracts showed a remarkable effect in enhancing liver and kidney functions (Ali et al., 2010). The antioxidant activity of leaves oil is not well studied and there is only one study concerned one commercial cultivar in Italy (Sacchetti et al., 2005). Additionally, some therapeutic properties of cypress oil are claimed by commercial companies and dozens of websites like astringent, antiseptic, antispasmodic, deodorant, diuretic, haemostatic, hepatic, styptic, sudorific, vasoconstrictor, respiratory tonic and sedative.

*L. camara*, wild or red sage, is the most widespread species of this genus, originated in subtropical America and the genus include 150 cultivar used as ornamental plants (Ghisalberti, 2000; Sousa et al., 2012). The essential oil of *L. camara* from different regions of the world has been studied (Sefidkon, 2002; Kasali et al., 2004) and the oils differ in their chemical composition according to geographic origin of the plants. Previously, the leaf oil showed strong antibacterial activity (Saikia & Sahoo, 2011). The plant had antimycobacterial (Beguma et al., 2008), antifungal (Sonibare & Effiong, 2008) and nematicidal effect (Qamar et al., 2005). Furthermore, the leaves and flowers essential oil of *L. camara* grown in Egypt showed insecticidal activity (Abdel-Hady et al., 2005). The antioxidant activities of the plant were not well documented, where one Peruvian leaf oil was studied (Benites et al., 2009).

Although a large variety of plants had been studied for their antioxidant or antibacterial activities in the world, either there are no or few reports concerning the leaves oil of these species in Egypt or the world, the search for natural antioxidants which is available in the plant natural products like phenolic compounds and others is gaining more momentum now days. Sequentially, we aimed to analyse the essential oils composition using GC-MS of three different local species belonging to different families and to elucidate their antibacterial activities and antioxidant activities as guide step for pharmaceutical, bactericidal industries and health improvement.

# 2. Materials and Methods

#### 2.1 Plant Material

Leaf samples from grown trees of *S. cumini* (GPS: Latitude 31.20451: Longitude 29.94859) and *C. sempervirens* (GPS: Latitude 31.20440: Longitude 29.94928) and one shrub which is *L. camara* (GPS: Latitude 31.20394: Longitude 29.94968) located in Antoniadis Garden, Horticultural Research Institute, Alexandria, Egypt. *S. cumini, C. sempervirens* and *L. camara* samples identified by Dr. Nader Elshanhory and Dr. Hesham Ali, Antoniadis Research Centre and obtained voucher numbers at Egypt barcode of life project (www.egyptbol.org), Faculty of Agriculture, Alexandria University and their voucher numbers were Hosam00014, Hosam00020, Hosam00090, respectively.

# 2.2 Extraction of Essential Oils

The collected freshly-green leaves (200 g) of *S. cumini* and *L. camara* and the twigs and needles of *C. sempervirens* were cut into small pieces and hydro-distilled for 3 h using a Clevenger-type apparatus (Clevenger, 1928). The essential oil was dried over anhydrous sodium sulphate, filtered, kept dry in sealed brown bottles and stored at 4°C to wait chemical and bioassay analyses. The amount of oil obtained was measured and the oil percentage was calculated based on the fresh weight (v/w %).

# 2.3 GC-MS Analysis Conditions

The analysis of the essential oils was performed using an Agilent 6890 gas chromatography equipped with an Agilent mass spectrometric detector; with a direct capillary interface and fused silica capillary column HP-5MS (30 m  $\times$  0.32 mm  $\times$  0.25 um film thickness). Samples were injected under the following conditions: Helium was used as carrier gas at approximately 1.0 mL/min., pulsed splitless mode. The solvent delay was 3 min. and the injection size was 1.0 µL. The MS detector was operated in electron impact ionization mode with ioning energy of 70 eV scanning from *m*/*z* 50-500. The ion source temperature was 230°C and the quadruple temperature was 150°C. The electron multiplier voltage (EM voltage) was maintained 1050 V above auto tune. The instrument was manually tuned using perfluorotributyl amine (PFTBA). The GC temperature program was started at 60°C (3min) then elevated to 280°C at rate of 8°C/min. The detector and injector temperature were set at 280 and 250°C, respectively.

Identification of the constituents was performed on the basis of MS library search (NIST and Wiley), and by comparing with the MS literature data (Adams, 1995; Davies, 1990). The relative amounts of individual components were calculated based on the GC peak area (FID response) without using correction factors.

## 2.4 Antibacterial Susceptibility Test

The biological assays were carried out on the oils with concentration of 1000  $\mu$ g/mL against the gram positive bacteria; *Bacillus subtilis* ATCC 6633 (*B. subtilis*), *Staphylococcus aureus* ATCC 6538 (*S. aureus*) and *Sarcina* 

*lutea* ATCC 9341 (*S. lutea*) and the gram negative bacteria; *Escherichia coli* ATCC 8739 (*E. coli*) and *Pseudomonas aeruginosa* ATCC 9027 (*P. aeruginosa*). Furthermore, a Gram-negative bacteria; *Agrobacterium tumefaciens* ATCC 1593-2 (*A. tumefaciens*) and *Pectobacterium carotovorum subsp. carotovorum* ATCC 39048 (*P. carotovorum*) were used as phytopathogenic bacteria. Bacterial strains were provided by the Laboratory of Bacteriology (Faculty of Agriculture, Alexandria University, Egypt). Nutrient agar (NA) medium was used for maintenance of the tested bacterial organisms. Mueller Hinton agar (MHA) was used in all bioassays applying the disc diffusion method.

# 2.4.1 Disc Diffusion Method

The agar disc diffusion method was employed for the determination of antimicrobial activities of the essential oils (NCCLS, 1997). A suspension of the tested bacteria (0.1 mL of  $10^8$  CFU/mL) was spread on the solid media plates. Filter paper discs (5 mm in diameter) were loaded with 20 µL of the oil and placed on the inoculated plates and, after staying at 4°C for 2 h, the plates were incubated at 37°C for 24 h. The diameters of the inhibition zones (IZs) were measured in millimetres. Negative control was prepared using respective solvent. Tetracycline (20µg/disc) was used as a positive control with the tested bacteria. All tests were performed in duplicate.

#### 2.4.2 Determination of Minimum Inhibitory Concentration of Oils

Minimum inhibitory concentrations (MICs) were determined by serial dilution of oils (250, 500, 1000, 2000, 4000 and 5000  $\mu$ g/mL). This was performed in 96-well micro-plates (Eloff, 1998) by filling all wells, with 50  $\mu$ L sterile Mueller Hinton Broth (MHB) with minor modification. Two wells were used as a sterility and growth control respectively with the sterility control containing only Oxoid<sup>®</sup> MHB, whilst the growth control containing both MHB as well as test organism. After adding 50  $\mu$ L of the bacterial suspension (10<sup>5</sup> CFU/mL) to each row (except for the sterility control), the micro-plate was covered and incubated at 37°C at 100% relative humidity overnight. The following morning 50  $\mu$ L of a 0.2 mg/mL solution of *p*-iodonitrotetrazolium violet (INT, Sigma-Aldrich) was added to each well. Inhibition of the growth was indicated by a clear solution or a definite decrease in colour reaction. Oils used for the determination of MICs were dissolved in 10% dimethyl sulfoxide (DMSO, Sigma-Aldrich) with Tween 80 (0.5 v/v) and was made up as a stock solution (5000  $\mu$ g/mL) with distilled water.

#### 2.5 Antioxidant Capacity / DPPH Radical-scavenging Assay

Free radical scavenging activity of the samples was determined using the 1,1,-diphenyl-2-picryl-hydrazyl (DPPH) method (Shirwaikar et al., 2006) with some modifications. An aliquot of 2 mL of stock solution of 0.1 mM DPPH (Sigma-Aldrich) reagent dissolved in pure methanol was added to a test tube with 2 mL of the sample solution in methanol ( $200 \mu g/L$ ). The reaction mixture was mixed for 10 s and left to stand in fibber box at room temperature in the dark for 30 min. The absorbance was measured at 517 nm, using a UV scanning spectrophotometer (Unico<sup>®</sup> 1200). Pure methanol (Sigma-Aldrich) was used to calibrate the spectrophotometer. The decrease of the absorbance of the DPPH solution indicates an increase in DPPH radical scavenging activity. Total antioxidant activity (TAA %) was expressed as the percentage inhibition of the DPPH radical and was determined by the following equation:

$$\Gamma AA (\%) = (A_{\text{control}} - A_{\text{sample}} / A_{\text{control}}) \times 100$$
(1)

where TAA is the total antioxidant activity,  $A_{\text{control}}$  is the absorbance of the control reaction (containing all reagents except the test compound) and  $A_{\text{sample}}$  is the absorbance of the test compound. The control contained 2 mL of DPPH solution and 2 mL of methanol. The measurements of DPPH radical scavenging activity were carried out for two replicates, and values are an average of two replicates. The results are expressed as mean values  $\pm$  standard deviation (SD).

#### 3. Results and Discussion

#### 3.1 Chemical Composition of the Essential Oils

Essential oils yields (v/w %) were 0.125, 0.35 and 0.125% for *S. cumini*, *C. sempervirens* and *L. camara*, respectively. Oil yields were similar to studies on *C. sempervirens* (da Silva et al., 1999), and higher than Indian *S. cumini* (Shafi et al., 2002).

The main constituents of *S. cumini* oil (Table 1) were  $\alpha$ -pinene (17.53%),  $\alpha$ -terpineol (16.67 %), alloocimene (13.55%),  $\alpha$ -bornyl acetate (6.37%), 2- $\beta$ -pinene (5.34%), caryophyllene (5.41%), caryophyllene oxide (4.81%), L-limonene (4.5%),  $\alpha$ -humulene (3.08%) and  $\alpha$ -terpineolene (2.71%). In Brazil, it was found that 85% of leaf oil was composed of  $\alpha$ -pinene (30%), cis-ocimin (9%), trans-ocimine (9.5%), 2- $\beta$ -pinene (20%) and  $\alpha$ -humulene with 2.8% (Craveiro et al., 1983). The most components found in the Egyptian *S. cumini* were found before (Machado et al., 2011) in buds oil of *S. aromaticum* but the percentages were much smaller because of the abundance of the

eugenol (85.3%). The major difference found here between both of the species is the availability of the alloocimene in high ratio within *S. cumini*.

S. cumini			C. sempervirens			L. camara		
Constituent	RT (min)*	Oil% <sup>†</sup>	Constituent	RT (min)*	Oil% <sup>†</sup>	Constituent	RT (min)*	Oil% <sup>†</sup>
α-Pinene	4.3	17.53	α-Pinene	4.48	6.9	α-Pinene	4.24	2.11
2-β-Pinene	5.27	5.34	2-β-Pinene	5.5	2.18	Thujene	4.11	0.03
L-Limonene	6.33	3.33	Δ3-Carene	5.95	17.85	Sabinene	5.17	4.28
Cis-Ocimene	6.62	4.11	Limonene	6.34	5.82	γ-Terpinene	5.91	0.42
1,3,6-Octatriene	6.83	0.72	γ-Terpinene	7.05	0.45	∆3-Carene	6.09	1.22
α-Terpinolene	7.69	2.71	α-Terpinolene	7.7	9.17	1,8-Cineol	6.59	1.43
Alloocimene	8.59	13.55	α-Terpineol	9.92	3.13	7-Isoquinolinol	7.17	0.46
Campheniolanol	9.02	0.28	Carvacrol methyl ether	10.92	0.87	Trans-Sabinene	7.31	1.09
Boreneol L	9.41	0.89	$\alpha$ -Bornyl acetate	11.68	1.02	α-Terpinolene	7.7	0.42
α-Terpineol	10.05	16.67	γ-Terpinene	12.67	1.23	Linalool	8.01	1.61
Fenchyl acetate	10.43	2.53	Camphene	12.88	7.3	Alloocimene	8.6	0.41
α-Bornyl acetate	11.71	6.37	Cedrene	14.08	0.63	Camphor	8.93	3.57
1(7),5,8-0-Menthatriene	11.96	0.5	α-Caryophyllene	14.08	0.8	Borneol	9.43	1.15
Caryophyllene	14.09	5.41	α-Humulene	14.64	0.37	4-Terpineol	9.63	1.78
p-Cymol	14.18	0.23	Germacrene-D	15.1	2.75	α-Terpineol	9.93	1.77
p-Mentha-1	14.41	0.63	Phenol	15.61	0.48	Bicycloelemene	12.62	3.78
α-Humulene	14.66	3.08	Δ-Cadinene	15.78	0.84	α-Cubebene	13.29	1.15
Aromadendrene	14.91	0.2	β-cedrene	16.93	0.13	Germacrene-D	13.6	5.23
Phenol	15.61	0.37	Cedrol	17.22	21.29	Trans-Caryophyllene	14.26	15.57
α-Farnesene	15.83	1.17	Diepi-α- cedrene-1	17.52	1.03	α-Humulene	14.8	9.16
O-Menth-8-ene	16.29	0.42	α-Cadinol	17.89	1.18	Bicyclogermacrene	15.48	6.69
Caryophyllene alcohol	16.6	1.65	Cycloisolongifolene	18.38	1.03	Farnesol	16.54	6.38
Caryophyllene Oxide	16.81	4.81	Phenanthrene	22.5	7.81	Davanone	16.66	0.99
α-Caryophyllene	17.04	0.23	Phenanthrenol	23.26	1.81	Spathulenol	16.88	6.02
Epiglobulol	17.1	1.36	13(16), 14-Labdien-8-ol	23.88	2.36	Caryophylla-3,8	17.31	2.97
Decalina	17.22	1.65	Ferruginol	26.48	0.18	γ-Cadinene	17.49	0.08
Trans/trans-photocitral	17.58	0.77	Traces	-	1.39	α-Cyclocitral	17.66	5.08
Junipene	17.65	0.95				∆-cadinene	17.74	2.9
Globulol	17.85	0.44				Calamanene	17.94	3.22
Traces	-	2.10				Trans/Trans-Photocitral	18.32	4.22
						Quinoline	18.45	0.15
						Caryophyllene Oxide	19.1	0.21
						Isoaromadendrene epoxide	19.57	0.32
						Oxiranone	1637	2.65
						Traces	-	1.48

Table 1. Essential oil composition of S. cumini, C. sempervirens and L. camara leaves

\* RT: Retention time (min).

<sup>†</sup>Percentage of total FID area obtained on HP-5 capillary column.

The main constituents of *L. camara* (Table 1) were trans-caryophyllene (15.57%),  $\alpha$ -humulene (9.16%), bicyclogermacrene (6.69%), farnesol (6.38%), spathulenol (6.02%), germacrene-D (5.23%),  $\alpha$ -cyclocitral (5.08%), sabinene (4.28%), trans-photocitral (4.4%), bicycloelemene (3.78%), camphor (3.57%), calamanene

The main constituents of *C. sempervirens* oil (Table 1) where cedrol (21.29%),  $\Delta^3$ -carene (17.85%),  $\alpha$ -pinene (6.9%), 2- $\beta$ -pinene (2.18%), limonene (5.82),  $\alpha$ -terpineolene (9.17%), phenanthrene (7.81%), camphene (7.3%),  $\alpha$ -terpineol (3.13%) and germacrene-D (2.75%). Some of these components were found in the same species in the Cameron (Tapondjou et al., 2005) although the percentage of cedrol was lower (3.3%), both of sabinene (14.8%) and  $\alpha$ -pinene were higher (9.9%). The camphene percentage herein was 7.3% which may prove to have a chemotaxonomic value in the Turkish cultivars of *C. sempervirens* of Horizontal and Pyrimidalis (Uçar et al., 2007). High sesquiterpenes including cedrol content in the essential oil of *C. dupreziana* Dejant in Algeria was found (Ramadani & Lograda, 2009) which is similar to our tested oil from *C. sempervirens*. The percentage of limonene achieved here is quite low (16.5%) compared to that of the Brazilian cultivars (da Silva et al., 1999).

(3.22%), calarene (3.19%),  $\Delta$ -cadinene (2.9%),  $\alpha$ -pinene (2.11%) and trans-sabinene (1.09%). These results were in agreement with most published data like the South Chinese essential oil revealed by Sundufu and Shoushan (2004) where they found almost exact  $\alpha$ -humulene (9.3%), higher 1,8-cineol (4.6%), lower sabinene (1.1%), lower  $\alpha$ -pinene (0.8%), lower isomer  $\beta$ -caryophyllene (13.35%) and dominant higher germacrene D (15.9%) also higher germacrene D was found in other studies (da Silva et al., 1999; Möllenbeck et al., 1997). Recent study in Algeria reported that  $\beta$ -caryophyllene was dominant (35.7%) and lower  $\alpha$ -humulene (3.72%), in *L. camara* oil (Zoubiri & Baaliouamer, 2011). A previous study (Abdel-Hady et al., 2005) on *L. camara* located in Cairo, Egypt found that the major constituents of the leaves essential oil were caryophyllene (9.76%), 1,8-cineol (9.37%) and  $\beta$ -pinene (8.15%).

#### 3.2 In vitro Antibacterial Test

The antibacterial activates of the essential oils from the leaves of *S. cumini*, *C. sempervirens* and *L. camara* using disc diffusion and MIC methods are shown in Table 2. The results showed that the antibacterial activities were ranged between 7 and 18 mm at 1000  $\mu$ g/mL oil concentration.

Table 2. Antibacterial activity of the essential oils of *S. cumini*, *C. sempervirens* and *L. camara* using agar disc diffusion and minimum inhibitory concentration methods

Bacterial	Essen	tial oils	Negative	Positive				
strain	S. cumini		C. sempervirens		L. camara		control	control*
	IZ	MIC	IZ	MIC	IZ	MIC	(mm)	(mm)
B. subtilis	16	< 250	18	< 250	13	500	n.a.	17
S. aureus	14	500	13	500	14	500	n.a.	22
E. coli	12	< 250	17	< 250	12	500	n.a.	18
P. aeruginosa	14	500	n.a.	2000	n.a.	5000	n.a.	26
S. lutea	17	< 250	16	< 250	15	< 250	n.a.	30
A. tumefaciens	12	< 250	8	500	n.a.	2000	n.a.	n.t.
<i>P</i> .	10	< 250	7	500	n.a.	2000	n.a.	n.t.
carotovorum								

IZ: Diameter of inhibition zone (mm) including disc diameter of 5 mm at 1000 µg/mL.

MIC: Minimum inhibitory concentration; values given as µg/mL.

n.a. Not active.

n.t. Not tested.

\*. Tetracycline (20 µg/disc).

Extracts from *Syzygium* species are known antibacterial (Chattopadhyay et al., 1998). The essential oil from *S. cumini* leaves demonstrated strong inhibition activity against the tested bacterial strains. The results demonstrated that the IZs were 16, 14, 12, 14 and 17 mm for *B. subtilis, S. aureus, E. coli, P. aeruginosa* and *S. lutea*, respectively. Our results were in agreements with previous study (Shafi et al., 2002), where the IZs were 13, 14, 12 and 17 mm for *B. subtilis, S. aureus, E. coli* and *P. aeruginosa*. Moreover, the essential oil of plants belonging to the family Myrtaceae are known for their biological activities which is suggested to be due to the presence of 1,8-cineole (Lis-Balchin et al., 1998) even the tested oil in this work didn't contain 1,8-cineole.

The essential oil of *C. sempervirens* presented antibacterial activity against the tested strains (Gram-positive and Gram-negative bacteria), showing the biggest IZs with *B. subtilis* and *E. coli* with 18 and 17 mm, respectively and nearly equivalent to the positive control used (18 mm with tetracycline). Moreover, *S. aureus* and *P. aeruginosa* strains showed resistant to the oil concentration at 1000  $\mu$ g/mL compared to positive control used as can be seen from Table 2. Additionally, the oil showed moderate activity against most of the bacteria tested (*E. coli* and *S. aureus*). On the other hand, it was noted that *P. aeruginosa* was not very sensitive to all the tested oils (Chéraif et al., 2007). Additionally, the cypress essential oil was found to have moderate antimicrobial activity when compared to vancomycin (30 mcg) and erythromycin (15 mcg) as antibiotics (Toroglu, 2007). In addition, the highest MIC values of *C. sempervirens* essential oil (4.0 µg/mL) are determined against *E. coli* (Toroglu, 2007). Furthermore, the MIC of the essential oil (% v/v) was >2.0 against *E. coli* (Hammer et al., 1999), where it was <250 µg/mL in our study.

The essential oil from *L. camara* shows activity against *B. subtilis*, *S. aureus*, *E. coli* and *S. lutea* at MICs values of 500, 500 and  $< 250 \mu g/mL$ , respectively. It shows activity against *P. aeruginosa* but at high concentration with MIC of 5000  $\mu g/mL$ . This result is lower by half in comparison with the study of Sonibare and Effiong (2008) which found that oil of *L. camara* had an antibacterial against *B. subtilis* and *P. aeruginosa* at MIC values of 1000 and 10000 ppm, respectively. In addition, all the values of IZs were lower than the positive control used (tetracycline). Furthermore, Saikia and Sahoo (2011) found that the major essential oil compounds in fresh leaves of *L. camara* were  $\alpha$ -caryophyllene (27.0%),  $\alpha$ -humulene (11.8%), sabinene (9.7%), bicyclogermacrene (8.1%) and davanone (4.7%) and the essential oil exhibited significant antibacterial activity against *E. coli*, *B. subtilis* and *S. aureus*.

The tested oils of *L. camara* and *C. sempervirens* showed weak activities against *A. tumefaciens* and *P. carotovorum* at 1000 µg/mL which might be attributed to absence of thymol, chlorothymol, and carvacrol proven to be potent against *A. tumefaciens* and *Erwinia carotovora (P. carotovorum)* and quite close to the effect of the standard (El-Zemity et al., 2008) also carbamate derivatives of  $\beta$ -citronellol and borneol improved the bactericidal activity against *E. carotovora*, but decreased it against *A. tumefaciens*. On the other hand, the oil from *S. cumini* showed good active against the growth *A. tumefaciens* and *P. carotovorum. S. lutea* presented the lowest MIC values (<250 µg/mL). The strains which presented the biggest inhibition zones (diffusion method) are not always the most sensitive (value of MIC was lower) because the diameter of IZs does not reflect the antibacterial activity of a compound (Hernández et al., 2005). Moreover, the IZ values could be affected by the solubility of the oil, the diffusion range in the agar, the evaporation (it can affect the dose), etc. (Kim et al., 1995; Cimanga et al., 2002). In addition, the antibacterial activities of the essential oils suggest its usefulness in the treatment of various infectious diseases caused by the tested bacteria.

# 3.3 Antioxidant Capacity / DPPH Radical-scavenging Assay

Essential oils, as natural sources of phenolic components, attract investigators to evaluate their activity as antioxidants or free radical scavengers. The DPPH is a stable free radical, which has been widely accepted as a tool for estimating free radical-scavenging activities of antioxidants (Edris, 2007). Free radical scavenging activity using DPPH results presented as TAA% were  $11.13\pm0.2$ ,  $9.13\pm0.1$  and  $3.13\pm0.2\%$  for *S. cumini*, *L. camara* and *C. sempervirens*; respectively. The chemical complexity of essential oils, often a mixture of dozens of compounds with different functional groups, polarity and antioxidant activity which may lead to scattered results, depending on the test employed (Sacchetti et al., 2005), that's why we preferred using DPPH which is widely used and geographic comparisons could be made. In *C. sempervirens* similar result was achieved (Sacchetti et al., 2005). In addition, the phenolic compounds extracted from leaves of *C. sempervirens* grown in Egypt showed high antioxidant activity (Ibrahim et al., 2007).

The highest antioxidant activity was found here is *S. Cumini* (11.13%) which might be attributed to the presence of high percentage  $\alpha$ -terpineol (16.67%) and  $\alpha$ -pinene (17.53%) as previously found in *Melaleuca alternifolia* oil (Kim et al., 2004) with the inherent antioxidant activity attributed mainly to terpinene and terpinolene contents. In general, *Syzygium* sp. is well documented as natural antioxidants in ancient civilizations like Thai in Thailand where they used the seeds as antioxidant (Maisuthisakul et al., 2007) but for our knowledge, no studies were conducted on leaf oil. A higher antioxidant activity is related to the presence of phenolic compounds (Choudhary & Swarnkar, 2011) like carvacrol, a monoterpenoid phenol (Tepe et al., 2005) which is available in *C. sempervirens* (0.87%) but with very low amounts. The cineol also was found to be strong antioxidant in *Mentha aquatica* (Mimica-Dukic et al., 2003) which was found in low amount in *L. camara* (1.43%). Additionaly, *L. camara* leaf extracts was reported to have antioxidant potential (Sathish et al., 2011). With a similar trend, *C. sempervirens* leaves essential oil had antioxidant activity comparable to our results (Sacchetti et al., 2005). Other specie *L. ukambensis* showed a strong antioxidant activity measured by DPPH method (Sawadogo et al., 2012).

# 4. Conclusions

The use of essential oils from *S. cumini, L. camara* and *C. sempervirens* as antibacterial material or as antioxidants might be of great value for pharmaceutical industry were they showed either high antibacterial or antioxidant activities agreed with or differed than other species or cultivars in the world. Indeed further work should be done to elucidate the future of the economic exploitation of their essential oils as antibacterial or antioxidants for human and plant health.

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