Available online on www.ijppr.com
International Journal of Pharmacognosy and Phytochemical Research 2015; 7(3); 502-509

ISSN: 0975-4873

Research Article

Assessment of Antioxidant and Antimicrobial Activities of Essential Oil and Extracts of *Boswellia carteri* Resin

Amal A. Mohamed[1], Sami I. Ali[1*], Hanan F. Kabiel[2], Ahmad K. Hegazy[2], Mimona A. Kord[2], Farouk K. EL-Baz[1]

[1]*Plant Biochemistry Dept., National Research Centre, 33 El Bohouth St., Dokki, Giza, Egypt. P.O. 12622, ID: 60014618.*
[2]*Botany Dept., Faculty of Science, Cairo University, Giza, Egypt.*

Available Online: 22nd April, 2015

ABSTRACT

The present study aimed to investigate the chemical composition of essential oil (EO) and the antioxidant and antimicrobial activities of methanol extract (Me-OH), ethyl acetate extract (ETOAC) and EO of *B. carteri* resin. Using gas chromatography/mass spectrometry (GC/MS), 40 components were identified in *B. carteri* resin essential oils, and the most abundant components were, verticiol (14.48 %), isobutylcyclopentane (12.25 %) and n-octyl acetate (9.20 %). The Me-OH extract exhibited the highest antioxidant activity using DPPH radical scavenging, Fe^{2+} chelating and the reducing power assays compared to ETOAC extract and EO. Also, the Me-OH extract showed the highest antimicrobial activity against all the tested bacterial and yeast microorganisms compared to ETOAC extract and EO. The Me-OH and ETOAC extracts exhibited the highest antimicrobial activity against *Streptococcus faecalis, Bacillus subtilis and Bacillus circulans* with MIC value 25µg/ml. In conclusion, Me-OH and ETOAC extracts as well as EO of *B. carteri* resin may be considered as a natural source for antimicrobial and antioxidant agents and could be used for application in pharmaceutical and food industries.

Keywords: Antimicrobial activity, essential oil, Fe^{2+} chelation, GC/MS, Boswellia resin.

INTRODUCTION

Essential oils and extracts of medicinal plants have been known as a great source of antioxidant agents and food additives[1]. Crude extracts of different plant materials rich in antioxidant compounds are progressively of interest in the food industry. They can block, prevent or inhibit the oxidative degradation of lipids that cause rancidity, discoloration and produce unpleasant flavors in food and therefore improve the quality and nutritional value of food[2]. For many years, different synthetic preservatives have been widely used as antioxidants and antimicrobial agents in the food industry to increase the storage and marketing shelf life of food[3]. The excessive use of synthetic preservatives, some of which are suspected because of their toxicity[4], making the way for the search of new safe substances for food preservation especially those derived from plants[5]. There are many plants which produce bioactive compounds that can be considered as good alternatives to synthetic antimicrobial and antioxidant agents[6]. In this concern, Mohamed et al.[7] confirmed the *in vitro* antioxidant, antibacterial, and antifungal efficacy of plant based essential oils in biological systems.

Boswellia carteri Birdwood., (Family: Burseraceae) is a moderate to large sized branching tree that grows in dry mountainous regions in Somalia, Southern Arabia, Sudan and, in rare cases, in Yemen[8]. *B. carteri* trees produce a natural oleogum resin called Frankincense through incisions made in their trunks[9]. Chemical analysis of *B. carteri* resin extracts revealed a group of abundant pentacyclic triterpenes, termed boswellic acids and tirucallic acids. Several types of α- and β- boswellic acids besides to tirucallic acids were isolated from *B. carteri*[10,11]. Moreover, several neutral terpenic compounds were isolated from Boswellia resins including verticillane diterpenes, cembrane diterpenes and olibanumols as terpenic alcohols[11,12]. Different compounds isolated from Boswellia resins especially boswellic acids and their derivatives have shown various biological activities[13,14]. Moreover, Ali et al.[11] evaluated the anti-inflammatory and anticancer activities of five bioactive compounds named; 3α-O-acetyl-8,24-dien-tirucallic acid, verticilla-4(20),7,11-triene, cembrene A, incensole acetate and incensole which isolated from *B. carteri* resin.

The present study aimed to investigate the chemical composition of *B. carteri* resin essential oil by using GC/MS. As well as to evaluate the antioxidant and antimicrobial activities of methanol extract, ethyl acetate extract and the essential oil of *B. carteri* resin.

MATERIALS AND METHODS

Plant materials

B. carteri resin was purchased from Harraz Herbs Company (http://www.harrazherbs.com), Cairo, Egypt and authenticated as resin of *Boswellia carteri* Birdwood

**Author for Correspondence*

(Somalia) by Prof. Dr. Fathy M. Soliman by comparison with a genuine sample kept in the Drug Museum of Pharmacognosy Dept., Faculty of Pharmacy, Cairo University, Egypt.

Preparation of resin extracts

The dried powder from *B. carteri* resin (5 g) was macerated separately in methanol and ethyl acetate for 24 h using orbital shaker (150 rpm) at room temperature. The extracts were filtered through Whatman No.4 filter paper. Residues were re-extracted twice with fresh aliquots of the same solvents. The filtrates of each solvent were pooled and evaporated using a rotatory vacuum evaporator (Heidolph, Germany) at 40ºC to obtain methanolic (Me-OH) extract (3.4 g, 68%, w/w) and ethyl acetate (ETOAC) extract (3.1 g, 62%, w/w).

Isolation of B. carteri resin essential oil

The pulverized resin of *B. carteri* (200 g) underwent water distillation for 5 h using all-glass Clevenger apparatus[15]. The essential oil (EO) was dried over anhydrous sodium sulfate to obtain average yield 2.44% (v/w) on a dry weight basis and relative density of 0.97. The oil was stored at 4ºC until further analysis.

Gas Chromatography/Mass Spectrometry (GC/MS) analysis of the essential oil

The essential oil analysis was performed on a Thermo Scientific capillary gas chromatography (model Trace GC ULTRA) directly coupled to ISQ Single Quadruple MS and equipped with TG-5MS non polar 5% Phenyl Methylpolysiloxane capillary column (30 m x 0.25 mm ID x 0.25 µm). The operating condition of GC oven temperature was maintained as: initial temperature 40ºC for 3 min, programmed rate 5ºC/min up to final temperature 280ºC with isotherm for 5 min. For GC/MS detection, an electron ionization system with ionization energy of 70 eV was used. Helium was used as a carrier gas at a constant flow rate of 1.0 ml/min. One microliter of diluted essential oil (1:1, v/v, in diethyl ether) was injected automatically in the splitless mode. Detection was performed in the full scan mode from 40 to 500 m/z. The identification of the chemical components was carried out based on the retention time (R_t) of each component compared with those of the Wiley9 and NIST08 mass spectra libraries[16].

Antioxidant activities

DPPH free radical scavenging activity

The DPPH free radical scavenging activity of Me-OH, ETOAC extracts and EO was measured from the bleaching of purple-colored solution of 2, 2-diphenyl-1-picrylhydrazyl (DPPH) according to our previously published work[17]. One milliliter of DPPH methanol solution (0.1 mM) was added to 3 ml of crude extracts and essential oil at various concentrations (0.5, 1.0, 1.5 and 2.0 mg/ml). Discoloration of the mixture was measured at 517 nm after 30 min incubation in dark at room temperature. BHT was used as positive control. The ability to scavenge the DPPH radical was calculated using the following equation:

DPPH scavenging effect (%) = $[A_C - A_S / A_C] \times 100$

where, A_C is the absorbance of the control reaction (containing all reagents except the sample) and A_S is the absorbance when the sample extract is added. The extract concentration providing 50% inhibition of radical scavenging activity (IC_{50}) was calculated and expressed as mg/ml.

Ferrous ion chelating activity

The ferrous ion chelating activity of Me-OH extract, ETOAC extract and EO was estimated following the procedure of Zhu et al.[18]. Three milliliters of Me-OH and ETOAC extracts at various concentrations (0.2, 0.3, 0.4, 0.5 and 1.0 mg/ml) and essential oil at various concentrations (0.5, 1.0, 1.5 and 2.0 mg/ml), were added to 60 µl of $FeSO_4.7H_2O$ (2 mM). The reaction was started by adding 100 µl of ferrozine (5mM). The mixture was shaken vigorously and kept back to stand at room temperature for 10 min. Absorbance of the solution was measured spectrophotometrically at 562 nm. EDTA was used as positive control. The inhibition percentage of ferrozine-Fe^{2+} complex formation was calculated according to the following equation:

Ferrous ion chelating activity (%) = $(1-A_1/A_0) \times 100$

where, A_0 was the absorbance of the control reaction (containing all reagents except the extract) and A_1 was the absorbance in presence of extract.

Ferric reducing power activity

The reduction capacity of both Me-OH and ETOAC extracts and EO was determined according to our previously published work[19]. One milliliter of Me-OH and ETOAC extracts and EO at different concentrations (1.0, 1.5 and 2.0 mg/ml) was mixed with 2.5 ml of sodium phosphate buffer (200 mM, pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixtures were incubated at 50ºC for 20 min, after that 2.5 ml of 10% trichloroacetic acid was added and the mixtures were centrifuged at 10,000 rpm for 10 min. the upper layer of the solution (5.0 ml) was mixed with 5.0 ml distilled water and 1.0 ml of 0.1% ferric chloride. The absorbance of the reaction mixtures was measured at 700 nm against blank. The BHT was used as positive control and results expressed as absorbance reading.

Antimicrobial activity

Microbial strains

The microorganisms were obtained from the American type culture collection (ATCC; Rockville, MD, USA). The Gram-positive bacteria; *Streptococcus faecalis* (ATCC- 47077), *Bacillus subtilis* (ATCC- 12228), *Bacillus circulans* (ATCC- 4513), *Listeria monocytogenes* (ATCC- 35152), Gram-negative bacteria; *Escherichia coli* (ATCC- 25922), *Pseudomonas aeruginosa* strain OS4 and two yeasts; *Saccharomyces cerevisiae* (ATCC- 9763) and *Candida albicans* (ATCC-10231) were used.

Culture medium and inoculums

The stock cultures of microorganisms used in this study were maintained on plate count agar slants at 4ºC. Inoculum was prepared by suspending a loop full of bacterial cultures into 10 ml of nutrient agar broth and was incubated at 37ºC for 24 h. About 60 µl of bacterial suspensions adjusted to 10^6-10^7 colony-forming units (CFU)/ml were taken and poured into Petri plates

containing 6 ml sterilized nutrient agar medium. Bacterial suspensions were spread to get a uniform lawn culture.

Antimicrobial activity assay

The agar well diffusion method was applied to detect antimicrobial activity[20]. Wells of 6 mm diameter were dug on the inoculated nutrient agar medium using sterilized cork borer and 60 µl of Me-OH extract, ETOAC extract and EO, dissolved in dimethylsulfoxide (DMSO) at concentration (400 µg/ml), were added in each well. The wells introduced with 60 µl of DMSO were used as a negative control. The plates were allowed to stand at 4ºC for 2 h before incubation to prevent evaporation of tested extracts. The plates were incubated at 37ºC overnight and examined for the inhibition zone. The diameter of the inhibition zone was measured in mm.

Minimum inhibitory concentration

A bacterial suspension (10^6-10^7 CFU/ml) of each tested microorganism was spread on the nutrient agar plate. The wells (6 mm diameter) were dug on the agar plate, and 60 µl of tested samples, dissolved in DMSO at different concentrations (25, 50, 100, 200, 300, 400 and 1000 µg/ml) were delivered into them. The plates were allowed to stand at 4ºC for 2 h before incubation to prevent evaporation of tested extracts. The plates were incubated at 37ºC for 24 h under aerobic conditions then followed by the measurement of the diameter of the inhibition zone expressed in millimeter. MIC was taken from the concentration of the lowest dosed well visually showing no growth after 24 h.

Statistical analysis

Data are reported as the mean ± SD of three measurements.

RESULTS AND DISCUSSION

Chemical composition of essential oil

The hydrodistillate of *B. carteri* resin was pale yellow oil with average yield 2.44 % (v/w) on a dry weight basis. The GC/MS investigation led to the identification of 40 constituents (Table 1). Generally, the components of the *B. carteri* essential oil were divided into seven classes in different ratios, which were monoterpene hydrocarbons (6.42%), oxygenated monoterpenes (13.31%), sesquiterpene hydrocarbons (2.09%), oxygenated sesquiterpenes (0.51%), diterpenes hydrocarbons (3.41%), oxygenated diterpenes (21.04%) and other components (53.22%). The major constituents were identified as verticiol (14.48 %), isobutylcyclopentane (12.25%), n-octyl acetate (9.20%) and 9-oxabicyclo-[6.1.0]-non-3-yne (9.12%).

The present results revealed some differences in the concentrations and types of the essential components with other reports of GC/MS analysis of *B. carteri* resin essential oil. For instance, in the present study, verticiol (14.48%), α-pinene (3.09%) and cembrene (2.07%) were presented at a high concentrations compared to the previous report of Chen et al.[21] who found low concentrations of the same compounds which were presented as (1.25, 0.07 and 0.24%), respectively. On the other hand, n-octyl acetate was presented at low concentration (9.20%) in the present study compared to (13.4% and 34.66%) in previous reports of Mikhaeil et al.[22] and Chen et al.[21], respectively. These results indicate that the compositions of essential oils of *B. carteri* resins are influenced by several factors such as climatic, seasonal and experimental conditions. The different biological activities such as; anticancer, antibacterial and antifungal activities of Boswellia essential oils have been reported[21,23].

Antioxidant activity

DPPH free radical scavenging activity

The effect of antioxidants on DPPH radical scavenging is thought to be due to their hydrogen donating ability. Figure 1 shows DPPH scavenging activity (%) of Me-OH, ETOAC extracts and EO of *B. carteri* resin at different concentrations (0.5-2.0 mg/ml). A concentration dependent scavenging activity was clearly demonstrated where DPPH scavenging activity increased with increasing concentrations. At 2.0 mg/ml, the value percent of the activity of Me-OH, ETOAC extracts and EO were 18.6, 17.8 and 7.3%, respectively. The lowest scavenging activity of the essential oil in this study is in agreement with the results of Mothana et al.[24] who reported that the essential oils of different Boswellia species exhibited weak antioxidant abilities in DPPH reduction. The same authors demonstrated that the three essential oils of *B. elongate*, *B. dioscorides* and *B. socotrana* exhibited weak radical scavenging effects (21%, 22% and 28%, respectively) at a concentration of 1mg/ml compared to ascorbic acid (which used as positive control) that gave 96% antioxidant activty.

The values of IC_{50} were in the ascending order BHT > Me-OH extract > ETOAC extract > EO with values of 0.07, 5.78, 7.66, and 15.21 mg/ml, respectively. However, Ali et al.[25] reported that *B. ameero, B. socotrana* and *B. elongata* resin essential oils exhibited low DPPH scavenging activity with an IC_{50} value of 175.2, 121.4 and 211.2 µg/ml, respectively. The present results indicated that Me-OH extract exhibited the highest DPPH radical scavenging activity compared to the ETOAC extract and the EO, but it still gave low DPPH radical scavenging activity compared to BHT. The results of the present study are in similarity with the previous results of Mohamed et al.[7] who demonstrated that the methanolic extract of *Commiphora myrrha* resin exhibited DPPH radical scavenging activity higher than thier ethyl acetate extract and essential oils. The highest DPPH radical scavenging activity of Me-OH and ETOAC extracts compared to EO seems to be attributed to the presence of sesquiterpenoids, diterpenes and triterpenes especially boswellic acids and other tirucallic and lupeolic acids in crude extracts of *B. carteri* resin[10,26], and these terpenoids could be act as electron donors, and they can react with free radicals to convert them to more stable products and terminate radical chain reaction.

Ferrous ions chelating activity

The Fe^{2+} chelating activity of Me-OH, ETOAC extracts and EO of *B. carteri* resin showed a concentration dependent manner where it increased with increasing concentrations (Figure 2). The Me-OH extract was found to be the most potent Fe^{2+}chelator, as it caused 75.1 %

Table 1. GC/MS chemical profile of *B. carteri* resin essential oil.

No.	[a]R_t	Compounds name	Area (%)[b]	Molecular formula
1	9.67	α-Phellandrene	0.25	$C_{10}H_{16}$
2	9.89	α-Pinene	3.09	$C_{10}H_{16}$
3	11.20	Sabinene	0.77	$C_{10}H_{16}$
4	11.78	α-Myrcene	0.86	$C_{10}H_{16}$
5	12.16	Octanal	0.28	$C_8H_{16}O$
6	12.53	1-Hexyl acetate	1.11	$C_8H_{16}O_2$
7	13.05	9-Oxabicyclo[6.1.0]non-3-yne	9.12	$C_8H_{10}O$
8	13.29	Trans-á-Ocimene	0.63	$C_{10}H_{16}$
9	13.63	1,4-Cyclononadiene	2.86	C_9H_{14}
10	14.61	Isobutylcyclopentane	12.25	C_9H_{18}
11	14.67	Tetrahydro- 5- hydroxymethyl-2- furaldehyde-3-one	5.91	$C_6H_8O_4$
12	14.77	1-Butanol	4.28	$C_4H_{10}O$
13	15.48	Cis-á-Terpineol	1.82	$C_{10}H_{18}O$
14	16.20	trans,trans-3,5-Heptadien-2-one	2.63	$C_7H_{10}O$
15	18.60	n-Octyl acetate	9.20	$C_{10}H_{20}O_2$
16	19.42	1-Decanol	2.07	$C_{10}H_{22}O$
17	19.88	Carvone	0.22	$C_{10}H_{14}O$
18	20.32	3,5-Dimethoxytoluene	0.25	$C_9H_{12}O_2$
19	22.45	(Z)-3-Methyl-1,3-nonadiene	0.82	$C_{10}H_{18}$
20	22.75	Geranyl acetate	0.89	$C_{12}H_{20}O_2$
21	23.25	Lavandulyl acetate	1.44	$C_{12}H_{20}O_2$
22	23.94	3-Methyl-7-oxabicyclo-[4.1.0]-heptane	2.11	$C_7H_{12}O$
23	26.17	(E)2-decenyl acetate	0.38	$C_{12}H_{22}O_2$
24	26.53	Butylated hydroxytoluene	0.21	$C_{15}H_{24}O$
25	27.93	Dodecanoic acid	0.33	$C_{12}H_{24}O_2$
26	28.09	Octanoic acid, hexyl ester	0.57	$C_{14}H_{28}O_2$
27	30.76	Z-4-Tridecen-1-yl acetate	0.30	$C_{15}H_{28}O_2$
28	34.13	Geranyl-à-terpinene	0.28	$C_{20}H_{32}$
29	35.77	Cembrene	2.07	$C_{20}H_{32}$
30	36.02	Germacrene-D	0.58	$C_{15}H_{24}$
31	36.42	Verticellol	6.05	$C_{20}H_{34}O$
32	36.62	Cycloheptane,4-methylene-1-methyl-2-(2-methyl-1-propen-1-yl)-1-vinyl	0.44	$C_{15}H_{24}$
33	37.03	Cembrene C	0.24	$C_{20}H_{32}$
34	37.54	Verticiol	14.48	$C_{20}H_{34}O$
35	37.66	Cembrene A	0.82	$C_{20}H_{32}$
36	38.08	Trans-Geranylgeraniol	0.51	$C_{20}H_{34}O$
37	38.29	Azulene,1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl), [1R(1à,3aá,4à,7á)]-	0.69	$C_{15}H_{24}$
38	39.74	à-Humulene	0.38	$C_{15}H_{24}$
39	40.02	Benzo[f]chromen-3-one,Perhydro-2-acetyl-4-a,-7,7,10b-tetramethyl	2.69	$C_{19}H_{30}O_3$
40	40.46	2,5-dioxa-8,12-dimethyl-13-acetoxytetracyclo- [6.11.0.0-(9,17).0(12,16)]-nonadecane	6.12	$C_{21}H_{32}O_4$
		Total identified compounds	100	
		Monoterpenes hydrocarbons	6.42	
		Oxygenated monoterpenes	13.31	
		Sesquiterpene hydrocarbons	2.09	
		Oxygenated sesquiterpenes	0.51	
		Diterpenes hydrocarbons	3.41	
		Oxygenated diterpenes	21.04	
		Other constituents	53.22	

[a] Retention time (as minutes).
[b]The percentage composition was computed from the gas chromatography peak areas.

chelation, followed by ETOAC extract which caused 62.2% chelation at concentration 0.5 mg/ml. At the same concentration, the EO caused 24.6% as the lowest chelation activity. The present results are in agreement with Mohamed et al.[7] who reported that *Commiphora myrrha* resin essential oil showed low metal chelating activity compared to its methanol and ethyl acetate extracts. These results are in parallel trend with that of DPPH-scavenging activities (Figure 1). The Me-OH extract Fe^{2+} chelating activity was less than that of the positive control EDTA.

The IC_{50} of the Fe^{2+} values for EDTA, Me-OH, ETOAC extracts and EO were 0.028, 0.303, 0.419 and 0.868 mg/ml, respectively. In the present study the lowest Fe^{2+} chelating activity of *B. carteri* resin essential oil might be due to its low content of monoterpenes hydrocarbons. The present results are in agreement with previous reports of Nanyonga et al.[27] and Tepe et al.[28]; they confirmed that the essential oils having low content of monoterpenes hydrocarbons have poor antioxidant activity.

Ferric reducing power activity

The reducing power of Me-OH and ETOAC extracts as well as EO of *B. carteri* resin showed a concentration dependent activity where it increased with increasing concentrations (Figure 3). The Me-OH extract was superior to ETOAC extract and EO. At the highest concentration (2.0 mg/ml) the reducing powers were in the following order, Me-OH extract > ETOAC extract > EO with absorbance values of 0.153, 0.134 and 0.118, respectively. However Me-OH extract still gave low reducing power compared to BHT which used as positive control, it gave (1.377) absorbance at 200 µg/ml concentration.

The present results are in partial agreement with that of Jallali et al.[29] who reported that acetone crude extracts of *Crithmum maritimum* and *Inula crithmoïdes* showed higher ferric reducing power activity than their essential oils**.** It is extremely important to point out that, reductive potentials of the extracts and/or essential oils are strictly related with the polarities of their phytochemicals.

Parallel with the previously discussed results of DPPH scavenging and Fe^{2+} chelating activity (Figures 1 and 2) the essential oil of *B. carteri* showed the lowest ferric reducing power activity compared to Me-OH and ETOAC extracts and this might be attributed to low content of monoterpenes hydrocarbons and oxygenated sesquiterpenes in its essential oil.

Antimicrobial activity

The results of antimicrobial activities of Me-OH, ETOAC extracts and EO of *B. carteri* resin against both Gram-positive and Gram-negative bacteria as well as two yeasts were presented in Table (2). The Me-OH extract at 400 µg/ ml concentration exhibited the highest *in vitro* antimicrobial activity against all the tested microorganisms; it gave inhibition zones ranged from 8.5 to 31.8 mm. However the ETOAC extract gave the largest zone (11.8 mm) against *E. coli*. In general, all tested extracts have no effect on *C. albicans* growth at

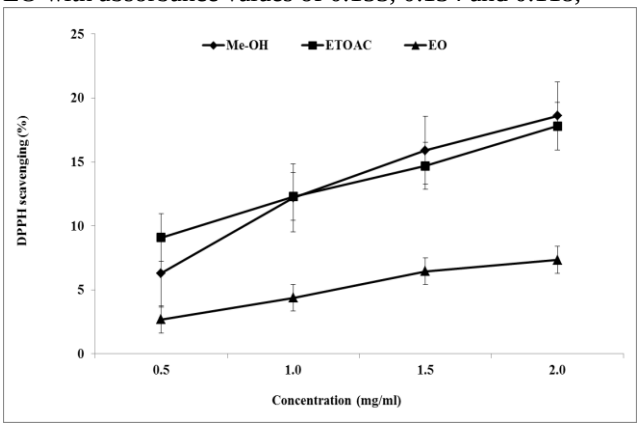

Figure 1. DPPH scavenging activity (%) of Me-OH, ETOAC extracts and EO of *B. carteri* resin at different concentrations. (n= 3, value= mean ± SD).

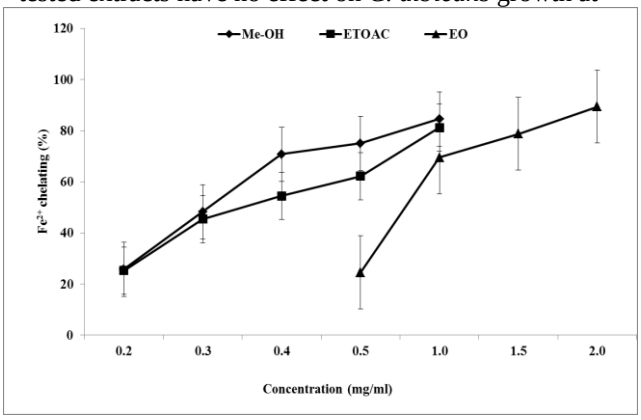

Figure 2. Fe^{2+} chelating activity (%) of Me-OH, ETOAC extracts and EO of *B.carteri* resin at different concentrations. (n= 3, value= mean ± SD).

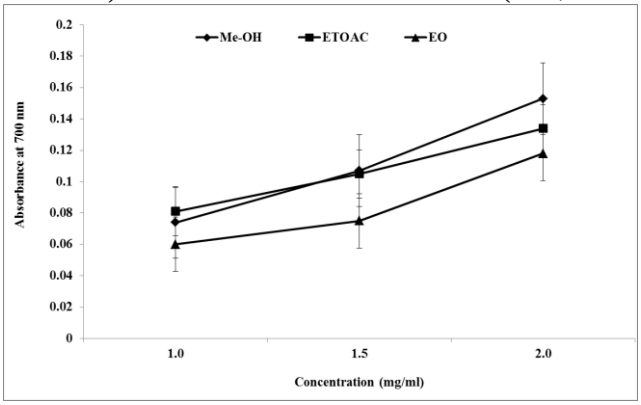

Figure 3. Ferric reducing power of Me-OH, ETOAC extracts and EO of *B. carteri* resin at different concentrations. (n= 3, value= mean ± SD).

Table 2. Antimicrobial activity of Me-OH, ETOAC extracts and EO of *B. carteri* resin at 400µg/ml concentration by agar well diffusion method.

Microorganisms	Inhibition zone (mm)[a]		
	Me-OH	ETOAC	EO
Gram-positive			
S. faecalis	13.5±1.3	12±0.8	9.5±1.2
B. subtilis	11.8±1.5	11.5±1.3	9.7±1.5
B. circulans	11.3±1.9	9.8±0.9	8.5±0.7
L. monocytogenes	31.8±2.1	28±1.4	9±1.4
Gram-negative			
E. coli	9.8±0.9	11.8±2.3	9.5±1.5
P. aeruginosa	8.5±0.2	7.1±0.1	NI
Yeast			
S. cerevisiae	13.3±1.6	12.5±1.2	9±0.8
C. albicans	NI	NI	NI

[a] Values are mean inhibition zone (mm) ±SD of three replicates. The diameter of the well (6mm) is included. NI: No inhibition zone.

Table 3. Minimal Inhibitory Concentration (MIC) of Me-OH, ETOAC extracts and EO of *B. carteri* resin.

Microorganisms	MIC (µg/ml)		
	Me-OH	ETOAC	EO
Gram-positive			
S. faecalis	25	25	50
B. subtilis	25	25	200
B. circulans	25	25	200
L. monocytogenes	400	100	400
Gram-negative			
E. coli	25	50	50
P. aeruginosa	300	200	>1000
Yeast			
S. cerevisiae	25	25	50
C. albicans	>1000	>1000	>1000

400 µg/ ml concentration. Also, EO at the same concentration showed no effect on *P. aeruginosa* growth. The highest antibacterial activity of *B. carteri* Me-OH extract might be attributed to the high polarity of Me-OH which is effective for more consistent extraction of different types of sesquiterpenoids, diterpenes and triterpenes especially boswellic acid and its derivatives. In this circumstance, Raja et al.[30] stated that β-boswellic acid exhibited antimicrobial activity against 112 pathogenic bacterial isolates including most of the American Type Culture Collection strains. It has been demonstrated that crude extracts and the essential oils exercise their antimicrobial activity by causing structural and functional damages to the microbial cell membrane[31]. In this context, Raja et al.[30] reported that 3-acetyl-11-keto-β-boswellic acid exhibited antibacterial activity against *S. aureus*, *S. epidermidis* and *S. mutans* by inhibiting the formation of bacterial cell biofilms generated by these bacteria. The present findings indicated that the highest activity was observed against *L. monocytogenes* followed by *S. faecalis* with the widest inhibition zones (31.8 and 13.5 mm), respectively, in contrast to *P. aeruginosa* showed the less zone (7.1 mm) with ETOAC extract (Table 2). This result is in dissimilarity with the previous study of Camarda et al.[23] who reported that *B. carteri* resin exhibited antimicrobial activity against *E. coli*, *P. aeruginosa*, and three strains of *S. aureus*, and the highest activity was found against *P. aeruginosa***.**

The results of MIC values obtained from antimicrobial tests ranged from 25 to >1000 µg/ml were presented in Table (3). The results showed that the Gram-positive bacterial strains such as, *S. faecalis, B. subtilis and B. circulans* were the most sensitive to both Me-OH and ETOAC extracts with MIC value 25µg/ml (Table 3). These results are in agreement with the findings of Mohamed et al.[32] who stated that Gram-positive bacteria were found to be more susceptible to plant extracts than Gram-negative bacteria**.** Furthermore, 3-acetyl-11-keto-β-boswellic acid which isolated from *B. serrate* resin was found to be an active compound showing MIC range of 2-8 µg/ml against the entire tested Gram positive bacteria[30]. The weaker antimicrobial activity against Gram-negative compared to Gram-positive bacteria is ascribed to the structure of their cellular walls, mainly with regard to the presence of lipoproteins and lipopolysaccharides in Gram-negative bacteria that form a barrier to hydrophobic compounds[33].

However, EO showed the lowest activity against *P. aeruginosa* with MIC value >1000 µg/ml (Table 3). The Me-OH, ETOAC extracts and EO exhibited the most activity against the yeast *S. cerevisiae* with MIC values 25, 25 and 50 µg/ml, respectively (Table 3). However, the Me-OH, ETOAC extracts and EO showed the lowest activity against *C. albicans* with MIC value >1000 µg/ml. These results are disagree with those of Camarda et al.[23] who reported that *B. rivae* essential oils showed the lowest MIC value of 2.6 µg /ml against *C. albicans*. According to Salvat et al.[34], plant extracts with MIC's less than/or around 0.5 mg/ml indicate good antibacterial activity. Accordingly, the Me-OH and ETOAC extracts as well as EO of *B. carteri* resin exhibited good antimicrobial activity against most of the tested microorganisms.

CONCLUSION

GC/MS analysis revealed the presence of verticiol as the major oxygenated diterpene compound in the EO of *B. carteri* resin. However, the Me-OH, ETOAC extracts and EO of *B. carteri* resin exhibited interesting antioxidant and antimicrobial activities. The Me-OH extract exhibited the highest antioxidant and antimicrobial activity as compared to ETOAC extract and EO. Further investigations are recommended including *in vitro* and *in vivo* studies to establish which components of the extracts or essential oil are responsible for the antioxidant and antimicrobial activity. Our investigation recommended *B. carteri* resin as a natural antioxidant and antimicrobial agents and it could be considered for use in food and pharmaceutical industries.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

REFERENCES

1. Joy B, Rajan A, Abraham E. Antimicrobial activity and chemical composition of essential oil from *Hedychium coronarium*. Phytotherapy Research 2007; 21:439-443.
2. Prakash A, Mathur K, Vishwakarma A, Vuppu S, Mishra B. Comparative assay of antioxidant and antibacterial properties of Indian culinary seasonal fruit peel extracts obtained from Vellore, Tamilnadu. International Journal of Pharmaceutical Sciences Review and Research 2013; 19:131-135.
3. Dai J, Zhu L, Yang L, Qiu J. Chemical composition, antioxidant and antimicrobial activities of essential oil from *Wedelia prostrate*. EXCLI Journal 2013; 12:479-490.
4. Seneviratne K, Kotuwegedara R. Antioxidant activities of the phenolic extracts of seed oils and seed hulls of five plant species. Food Science and Technology International 2009; 15:419-425.
5. Magnuson B, Munro I, Abbot P, Baldwin N, Lopez-Garcia R, Ly K, Mc Girr L, Roberts A, Socolovsky S. Review of the regulation and safety assessment of food substances in various countries and jurisdictions. Food Additives & Contaminants: Part A 2013; 30:1147-1220.
6. Silva-Espinoza BA, Ortega-Ramirez LA, Gonzalez-Aguilar GA, Olivas I, Ayala-Zavala JF. Proteccion antifungica y enriquecimiento antioxidante de fresa con aceite de hoja de canela. Revista Fitotecnia Mexicana 2013; 36:217-224.
7. Mohamed AA, Ali SI, EL-Baz FK, Hegazy AK, Kord MA. Chemical composition of essential oil and *in vitro* antioxidant and antimicrobial activities of crude extracts of *Commiphora myrrha* resin. Industrial Crops and Products 2014; 57:10-16.
8. Martinetz D, Lohs K, Janzen J. Weihrauch und Myrrhe, Kulturgeschichte und wirtschaftliche Bedeutung. Botanik-Chemie- Medizin, Wissenschaftliche Verlagsgesellschaft Stuttgart, 1989.
9. Blaschek W, Hänsel R, Keller K, Reichling J, Rimpler H, Schneider G. *Hagers Handbuch der Pharmazeutischen Praxis.* 5 Auflage, Springer, Verlag, Berlin-Heidelberg, 1998.
10. Banno N, Akihisa T, Yasukawa K, Tokuda H, Tabata K, Nakamura Y, Nishimura R, Kimura Y, Suzuki T. Anti-inflammatory activities of the triterpene acids from the resin of *Boswellia carteri*. Journal of Ethnopharmacology 2006; 107:249-253.
11. Ali SI, Zhang C-R, Mohamed AA, EL-Baz FK, Hegazy AK, Kord MA, Nair MG. Major constituents of *Boswellia carteri* resin exhibit cyclooxygenase enzyme inhibition and antiproliferative activity. Natural Product Communications 2013; 8:1365-1366.
12. Yoshikawa M, Morikawa T, Oominami H, Matsuda H. Absolute stereostructures of Olibanumols A, B, C, H, I, and J from Olibanum, gum- resin of *Boswellia carterii*, and inhibitors of nitric oxide production in lipopolysaccharide-activated mouse peritoneal macrophages. Chemical and Pharmaceutical Bulletin 2009; 57:957-964.
13. Ganzera M, Khan IA. A reversed phase high performance liquid chromatography method for the analysis of boswellic acids in *Boswellia serrata*. Planta Medica 2001; 67:778-780.
14. Poeckel D, Werz O. Boswellic acids: biological actions and molecular targets. Current Medicinal Chemistry 2006; 13:3359-3369.
15. European Pharmacopoeia. Pharmacopée Européene, Conseil de l' Europe 1996. Vol.1, Maisonneuve S.A., Sainte Ruffine, 1997.
16. National Institute of Standards and Technology (NIST). 2010; http://webbook.nist.gov/chemistry/name-ser.html Accessed 15.05.13.
17. Mohamed AA, Ali SI, El-Baz FK. Antioxidant and antibacterial activities of crude extracts and essential oils of *Syzygium cumini* leaves. PLoS One 2013; 8:e60269.
18. Zhu K-X, Su C-Y, Guo X-N, Peng W, Zhou H-M. Influence of ultrasound during wheat gluten hydrolysis on the antioxidant activities of the

resulting hydrolysate. International Journal of Food Science and Technology 2011; 46:1053-1059.
19. El-Baz FK, Mohamed AA, Ali SI. Callus formation, phenolics content and related antioxidant activities in tissue culture of a medicinal plant colocynth (*Citrullus colocynthis*). Nova Biotechnologica 2010; 10:79-84.
20. Albayrak S, Aksoy A, Sagdic O, Hamzaoglu E. Compositions, antioxidant and antimicrobial activities of Helichrysum (Asteraceae) species collected from Turkey. Food Chemistry 2010; 119:114-122.
21. Chen Y, Zhou C, Ge Z, Liu Y, Liu Y, Feng W, Li S, Chen G, Wei1 T. Composition and potential anticancer activities of essential oils obtained from myrrh and frankincense. Oncology Letters 2013; 6:1140-1146.
22. Mikhaeil BR, Maatooq GT, Badria FA, Amer MMA. Chemistry and immunomodulatory activity of frankincense oil. Zeitschrift fur Naturforschung C 2003; 58:230-238.
23. Camarda L, Dayton T, Di Stefano V, Pitonzo R, Schillaci D. Chemical composition and antimicrobial activity of some oleogum resin essential oils from *Boswellia spp.* (Burseraceae). Annali di Chimica 2007; 97:837-844.
24. Mothana RAA, Hasson SS, Schultze W, Mowitz A, Lindequist U. Phytochemical composition and *in vitro* antimicrobial and antioxidant activities of essential oils of three endemic Soqotraen Boswellia species. Food Chemistry 2011; 126:1149-1154.
25. Ali ANA, Wurster M, Arnold N, Lindequist U, Wessjohann L. Chemical composition and biological activities of essential oils from the Oleogum resins of three endemic Soqotraen Boswellia species. Records of Natural Products 2008; 2:6-12.
26. Estrada AC, Syrovets T, Pitterle K, Lunov O, Buchele B, Schimana-Pfeifer J, Schmidt T, Morad SA, Simmet T. Tirucallic acids are novel pleckstrin homology domain-dependent Akt inhibitors inducing apoptosis in prostate cancer cells. Molecular Pharmacology 2010; 77(3):378-387.
27. Nanyonga SK, Opoku A, Lewu FB, Oyedeji AO, Singh M. Chemical composition, antioxidant activity and cytotoxicity of the essential oils of the leaves and stem of *Tarchonanthus camphoratus*. African journal of pharmacy and pharmacology 2013; 7:360-367.
28. Tepe B, Daferera D, Sokmen A, Sokmen M, Polissiou M. Antimicrobial and antioxidant activities of the essential oil and various extracts of *Salvia tomentosa* Miller (Lamiaceae). Food Chemistry 2005; 90:333-340.
29. Jallali I, Zaouali Y, Missaoui I, Smeoui A, Abdelly C, Ksouri R. Variability of antioxidant and antibacterial effects of essential oils and acetonic extracts of two edible halophytes: *Crithmum maritimum* L. and *Inula crithmoïdes* L. Food Chemistry 2014; 145:1031-1038.
30. Raja AF, Ali F, Khan IA, Shaw AS, Arora DS, Shah BA, Taneja SC. Antistaphylococcal and biofilm inhibitory activities of acetyl-11- keto-β-boswellic acid from *Boswellia serrate*. BMC Microbiology 2011; 11:54. doi:10.1186/1471-2180-11-54.
31. Goni P, Lopez P, Sanchez C, Gomez-Lus R, Becerril R, Nerin C. Antimicrobial activity in the vapour phase of a combination of cinnamon and clove essential oils. Food Chemistry 2009; 116:982-989.
32. Mohamed AA, Khalil AA, El-Beltagi HES. Antioxidant and antimicrobial properties of kaff Maryam (*Anastatica hierochuntica*) and doum palm (*Hyphaene thebaica*). Grasas Y Aceites 2010; 61:67-75.
33. Inouye S, Yamaguchi H, Takizawa T. Screening of the antibacterial effects of a variety of essential oils on respiratory tract pathogens, using a modified dilution assay method. Journal of Infection and Chemotherapy 2001; 7:251-254.
34. Salvat A, Antonacci L, Fortunato RH, Suarez EY, Godo HM. Antimicrobial activity in methanolic extracts of several plant species from Northern Argentina Phytomedicine 2004;11:230-234.