International Journal of Phytomedicine 5 (2013) 510-518
http://www.arjournals.org/index.php/ijpm/index

Original Research Article

ISSN: 0975-0185

In vitro antioxidant and antibacterial activities of extracts from Yemen aromatic plants

Riyadh AS Thabit[1], Xiang-Rong Cheng[1], Nabil Al-Hajj[1], Xue Tang[1] , Guo-Wei Le[1*]

***Corresponding author:**

Guo-Wei Le

[1]State Key Laboratory of Food Science and Technology, School of Food Science and Technology, Jiangnan University, Wuxi 214122, P. R. China

Abstract

The aim of this work was to (i) determine the antioxidant activity in the extracts of five Yemeni plants: Zizyphus vulgaris, Thymus vulgaris, Ruta graveolens, *Lavandula* officinalis, and Salavia officinalis by two different antioxidant test 2,2-Diphenyl-1-picrylhydrazyl (DPPH) and β-Carotene bleaching (BCB); (ii) determine the total phenolic (TP), total flavonoids (TF), total tannins (TT), and total anthocyanins (TA) of the extracts by Folin-Ciocalteu method, aluminium chloride method, Folin and Ciocalteu method, and pH-differential method, respectively; (iii) investigate the effectiveness of the extracts on the growth inhibition of some indicators of foodborne illness bacteria by agar well diffusion assay. There is a great variability in the TP, TF, TT, TA, inhibition of DPPH and BCB, and antibacterial effectiveness of the extraction obtained from the five plants. S. officinalis and L. officinalis best inhibition of DPPH (60.42%, 43.97%), BCB (80.77%, 67.77%), possessed the highest TP (75.2, 64.9 mg/g) and TT (0.70, 0.66 mg/g), while the Z. vulgaris possessed the highest TF (48.1 mg/g) and TA (0.49 mg/100g). The extracts of R. graveolens exhibited inhibitory effects against most tested bacteria at all added doses.

Keywords: Antioxidant; antibacterial; total Phenolic; *Salavia officinalis*; *Lavandula officinalis*

Introduction

Aromatic plants have been used for centuries as spices and condiments to confer aroma and flavor to food and beverages. Additionally, due to their constituents, medicinal and aromatic plants can act as stabilizer agents, playing an important role in the shelf-life of foods and beverages [1]. Recently, various extracts of plants have gained special interest as sources of natural antioxidant and antibacterial agents because of the possible toxicities of the synthetic antioxidants and the resistance to antibiotics that some microorganisms have acquired [2]. Natural antioxidants are compounds from plant or animal sources. Phenolic phytochemicals are a large group of substances and found in significant quantities in vegetables, fruits, spices, and seeds. Since they have been regarded as possible antioxidants, their roles in food industry and in chemoprevention of diseases have become an area of active research in many fields [3]. It was shown that oxidation reactions are involved in aging and progression of several diseases. It was proposed that antioxidant molecules may slow down the aging process, disease progression, and prolong the life span [4].

Preservation of food materials from degradation during production, storage and marketing is an important issue in the food industry. Chemical and/or microbial of industrially produced foods can inversely affect their quality and even lead to the formation of toxic materials in food. Herbs and spices have been employed since ancient times as flavouring and storing agents for food. But only in the last decade, scientific research has focused its interest on plant extracts as natural sources of antibacterial and antioxidant compounds [5]. They have gained a special attention as safer alternative additives for food preservation. The antibacterial properties of plant and their constituents from a wide variety of plants have been assessed. It is clear from these studies that these secondary plant metabolites have potential uses in medical procedures and applications in the cosmetic, pharmaceutical and food industries [6].

The objective of the research is to compare and assess the antioxidant and the antibacterial activities of five important plants grown in Yemen, including *Zizyphus vulgaris*, *Thymus vulgaris*, *Ruta graveolens*, *Lavandula officinalis*, and *Salavia officinalis,* and the total contents of phenolic (TPC), total flavonoids (TF), total tannins (TT), and total anthocyanins (TA) in the extracts of these plants.

Material and Methods

Chemicals and Machine

Tert-Butylhydroquinone (TBHQ), 2,2-Diphenyl-1-picrylhydrazyl (DPPH), ascorbic acid (VC), Folin-Ciocalteu reagent, gallic acid, quercetin reagent, β-carotene, and linoleic acid were from Sigma (USA). While ethanol, dimethylsulfoxide (DMSO), aluminium chloride, potassium acetate, potassium chloride, sodium acetate and Tween 20 were from Sinopharm chemical regent co.ltd.

Spectrophotometer (Shanghai-Techcomp, UV 2300), balance (Shanghai-Mettle Toledo, AB 204–N), rotary evaporator (Shanghai-Biochemical Equipment), water bath (Shanghai-Hengzi), pH metter (Shanghai-Mettler Toledo), incubation (Shanghai-Hengzi), and ultrasonic (Wuxi-Kejie Ultrasonic Electronic Equipment Co. Ltd, KJ-300).

Plant materials

Z. vulgaris, T. vulgaris, R. graveolens, L. officinalis, and *S. officinalis* were collected in September 2012 from Taiz region (Yemen). Identification was carried out by the Agricultural Research Authority (Taiz, Yemen) (Table1).

Table 1. Characteristics of the plants

Scientific name	Common name	Local name	Family	Part of the plant used
Z. vulgaris	Jujube	Al - seedr	Rhamnaceae	Leaves
T. vulgaris	Thyme	Al - zatar	Lamiaceae	Leaves & Stalk
R. graveolens	Rue	Al - shathab	Rutaceae	Leaves & Stalk
L.officinalis	Lavender	Al - suneb	Lamiaceae	Leaves & Stalk
S. officinalis	Sage	Al - marmaria	Lamiaceae	Leaves

Preparation of extracts

The plant samples were air-dried in shade, 20 grams from each plant was extracted with 600 ml of 90% ethanol in an ultrasonic device at room temperature. The ethanol extract was filtered and the residues were re-percolated for three times and solvent was removed using a rotary evaporator. Dried extracts were kept refrigerated until use.

Total Phenolic (TP)

This experiment was carried out according to the Folin–Ciocalteu's method [7]. Briefly, using a six-point calibration curve, the total phenolics were determined by a comparison of the values obtained with the calibration curve of gallic acid from (Figure. 1). The sample extract (0.2 mL) was mixed with 5 ml of deionized water, 10 mL of 7% (w/v) Na_2CO_3, and 1 ml of the Folin-Ciocalteu reagent. After incubation at room temperature for 90 min, the absorbance of the reaction mixture was measured at λ 765 nm against a blank containing the same mixture solution without the sample extract. The results were expressed as mg gallic acid equivalents (GAE)/g dry extract.

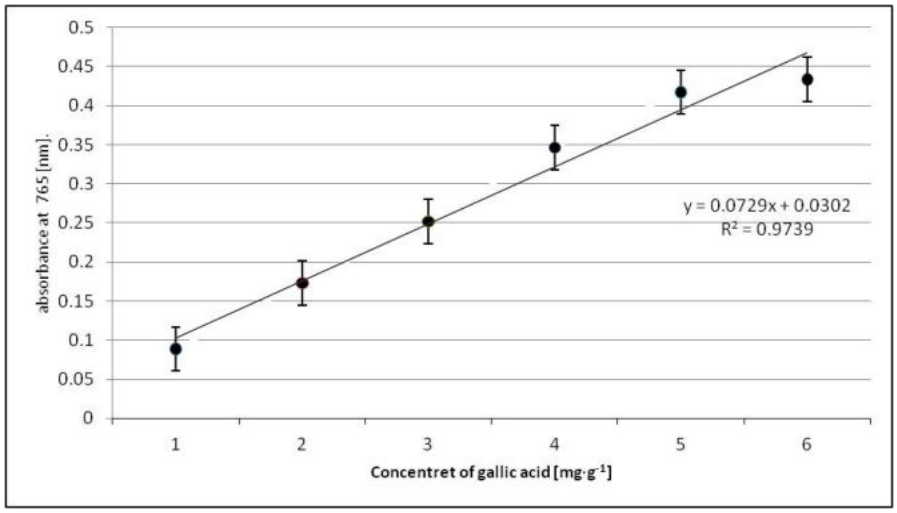

Figure. 1. Calibration curve for gallic acid (mg/g of dry extract)

Total Flavonoids (TF)

Flavonoids in the examined plant extracts were determined using spectrophotometrically using aluminium chloride according to method [8]. Each plant extracts (1 mL, 5 mg/mL) in methanol were mixed with 0.1 mL of 10% aluminium chloride hexahydrate, 0.1 mL of potassium acetate and 2.8 mL of deionized water. After the 40 minutes incubation at the room temperature, the absorbance of the reaction mixture was determined spectrophotometer at λ_{max} 415 nm. Quercetin was chosen as a standard. The concentration of flavonoids was read (mg/ml) on the calibration line and the total flavonoid content was expressed as mg g^{-1} of dry extracts (Figure. 2).

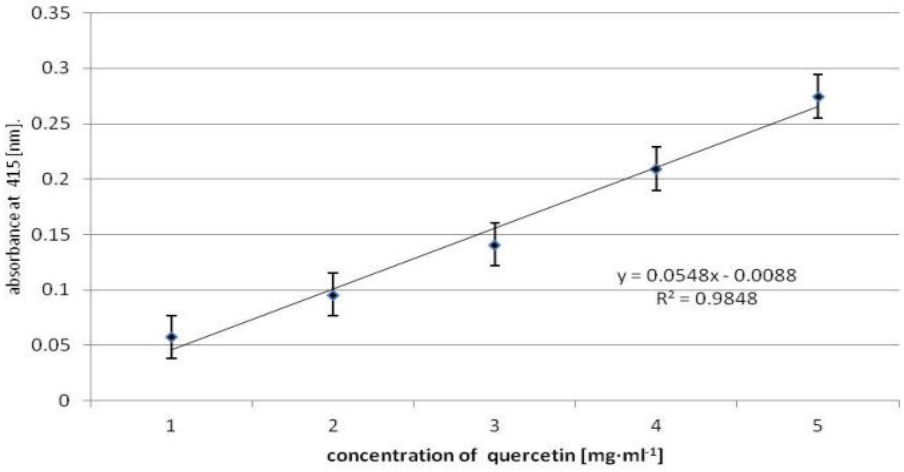

Figure. 2. Calibration curve for quercetin (mg/g of dry extracts

Total Tannins (TT)

The TT were estimated by folin's and Ciocalteu method [9]. Using a five-point calibration curve, the total tannins were determined by a comparison of the values obtained with the calibration curve of gallic acid (Figure. 3), total tannins values are expressed in terms of gallic acid equivalent (mg/g of dry extracts). The sample extract 0.1 mL was added with 7.5 mL of distilled water and adds 0.5 mL of Folin Phenol reagent, 1 mL of 35% sodium carbonate solution and dilute to 10 mL with distilled water. The mixture was shaken well, kept at room temperature for 30 min and absorbance was measured at λ 725 nm.

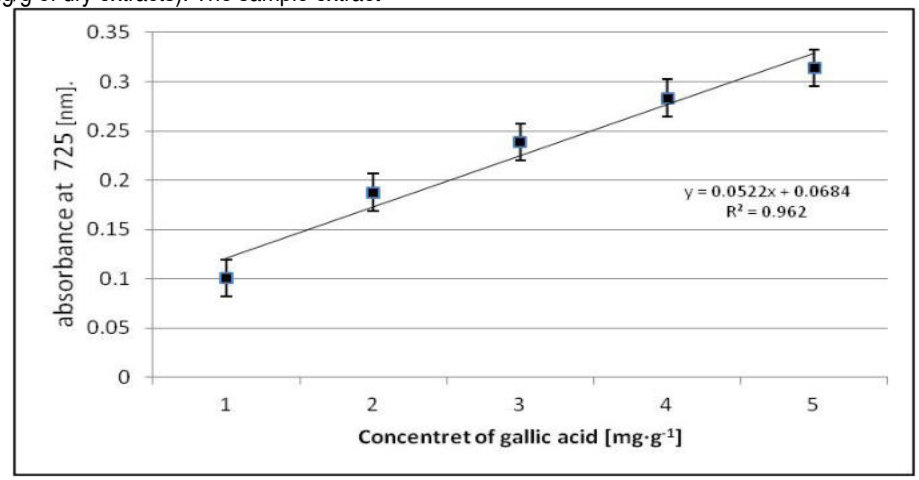

Figure. 3. Calibration curve for gallic acid (mg/g of dry extracts)

Total anthocyanin (TA)

The TA was determined by the pH-differential method [10]. Briefly, transfer 1 ml extracted solution into 10 mL volumetric flask for preparing two dilutions of the sample, one adjust volume with potassium chloride buffer, pH 1.0, and the other with sodium acetate buffer, pH 4.5, diluting each. Let these dilutions equilibrate for 15 min. Measure the absorbance of each dilution at the 510 and 700 nm to correct for haze, against a blank cell filled with distilled water. All measurements should be made between 15 min and 1 h after sample preparation, since longer standing times tend to increase observed readings. Absorbance readings are made against water blanks. The samples to be measured should be clear and contain no haze or sediments; however, some colloidal materials may be suspended in the sample, causing scattering of light and a cloudy appearance (haze). Calculate the absorbance (A) of the diluted sample as follows:

$A = (A_{510}–A_{700})$ pH 1.0 – $(A_{510}–A_{700})$ pH 4.5

Calculate anthocyanin pigment concentration, expressed as cyanidin-3-glucoside equivalents, as follows:

Monomeric anthocyanin pigment (mg/L) = (A × MW × DF × 1000)/(× 1)

and it was converted to mg of total anthocyanin content per 100 g dry extracts. Where MW (molecular weight) is 449.2 g/mol for cyanidin-3-glucoside (cyd-3-glu); DF is dilution factor; and is the molar absorptivity (26.900).

Antioxidant activity of extracts

In this assay, the antioxidant activity of plant extracts was evaluated by measuring the bleaching of the purple-colored ethanolic solution of DPPH [11]. The antioxidant activity of six different concentrations 0.2, 0.1, 0.05, 0.02, 0.01, and 0.005 mg/ml of plant extracts was measured in terms of hydrogen donating or radical scavenging ability [12]. The mixtures were well shaken and kept at room temperature in the dark for 30 min, than absorbance (A) was measured at 517 nm using a spectrophotometer. Vitamin C and TBHQ were used as positive controls, while ethanol was the blank. Inhibition (I %) of DPPH radical was calculated using the equation:

$$I\ (\%) = A_{blank} - A_{sample} / A_{blank}\ \ 100$$

The inhibitory concentration (IC_{50}) value represented the concentration of the plant extracts that caused 50% inhibition.

Determination of antioxidant activity using β-Carotene bleaching (BCB) assay

The antioxidant activity (AA) of extracts was evaluated by the β-carotene [13]. One milliliter of β-carotene solution (0.2 mg/ml chloroform) was pipetted into a round-bottom flask containing 0.02 ml of linoleic acid and 0.2 ml of Tween 20. The mixture was then evaporated at 40 C for 10 min using a rotary evaporator to remove the chloroform. Then, the mixture was immediately diluted with 100 mL of distilled water and agitated vigorously to form an emulsion. 5 mL aliquot of the emulsion was subsequently transferred into test tube containing 0.2 mL of extract. The tube was then gently mixed and placed in a water bath for 2 h at 50 C. The absorbance was measured at 470 nm at initial time (t = 0) against a blank, consisting of an emulsion with β-carotene. Solvent in the 5 ml of emulsion was used as control. The measurement was carried out at every 30 min intervals. AA was calculated based on a formula:

$$AA = [1- (A_0 – A_t)/ (A^o_0 - A^o_t)] \times 100$$

Where (A_0) and (A^o_0) are the absorbance values measured at initial time of the incubation for samples or standard and control respectively, while (A_t) and (A^o_t) are the absorbance values measured in the samples or standard and control at t = 120 min.

Microbial strains and media

Shigella dysenteriae CMCC 51302, *Escherichia Coli* ATCC 25922, *Salmonella typhimurium* CMCC 50013, *Streptococcus pyogenes* ATCC 12344 and *Staphylococcus aureus* ATCC 25923 were provided by the Microbiology Lab in the Nutrition and Food Functions of Jiangnan University, Wuxi, Jiangsu, China. Each culture was activated by transferring a loopful into nutrient broth (4 ml) followed by incubation at 37 ºC ± 1 ºC for 16 h. The optical density of each active culture was adjusted at 615 nm using fresh broth to give a standard inoculums of 10^8 (CFU)/ml.

Determination of antibacterial activity

The antibacterial activity was studied by the agar well diffusion method [14]. The bacterial suspension was spread uniformly on the agar surface. Agar surface was perforated with 6 mm-diameter holes, aseptically cut and filled with 100 µL of plant extracts. The extracts were used in the concentration of 5, 10, and 20 mg/ml from extract of DMSO. Streptomycin and Penicillin were used as a reference antibacterial, whereas DMSO (100 µL) was the negative control. The plates were incubated at 37 ºC ± 1 ºC for 21 h and then examined to verify inhibition. A positive result was defined as inhibition zone (halo size) of 9 mm or more around the holes.

Statistical analysis

Statistical methods were used to calculate means and standard deviations of three simultaneous assays carried out. Statistical analysis (SPSS, 16) was applied to the data to determine differences ($P < 0.05$) performed by ANOVA.

Results and Discussion

Total phenolics and total flavonoids

It has been suggested that the phenolic content of plant materials is correlated with their antioxidant and antibacterial activity. It is considered that the antioxidant activity of phenolic compounds is due to their high redox potentials, which allow them to act as reducing agents [15].

The content of polyphenols was 75.2, 64.9, 58.3, 39.9, and 33 mg/g in *S. officinalis*, *L. officinalis*, *T. vulgaris*, *R. graveolens*, and *Z. vulgaris*, respectively (Table 2). The results indicate significant differences among extracts (P 0.05), where the plant extracts of *S. officinalis* and *L. officinalis* contained higher phenol content compared with other extracts.

There is a strong correlation between total phenols and antioxidant activity in Table 3. These findings suggest that total phenols are a good predictor of *in vitro* antioxidant activity. The variability of total phenolic in this study could be partially attributed to differences in geographic sources of samples and varieties.

Table 2. Total phenolics, flavonoids , tannin and anthocyanins of some plants extracts (means ± S.D.).

Extraction	Total phenols [mg (GAE)/g]	Flavonoids [mg (quercetin)/g]	Tannins [mg (GAE)/g]	Anthocyanins [mg/100g]
Z. vulgaris	33.0 ^{e} ± 7.04	48.1 ^{a} ± 1.61	0.48 ^{d} ± 0.04	0.49 ^{a} ± 0.03
T. vulgaris	58.3 ^{c} ± 3.85	45.4 ^{b} ± 1.13	0.50 ^{c} ± 0.02	0.44 ^{b} ± 0.03
R. graveolens	39.9 ^{d} ± 0.16	23.9 ^{d} ± 0.81	0.51 ^{c} ± 0.01	0.02 ^{d} ± 0.01
L. officinalis	64.9 ^{b} ± 1.07	37.9 ^{c} ± 0.67	0.66 ^{b} ± 0.07	0.08 ^{c} ± 0.03
S. officinalis	75.2 ^{a} ± 7.85	39.9 ^{c} ± 6.01	0.70 ^{a} ± 0.01	0.12 ^{c} ± 0.01

The values are means of three independent analyses.

The TF content in the plant extracts ranged from 23.9 to 48.4 mg/g. There were significant differences (P 0.05) as the plant extract has higher flavonoid content *Z. vulgaris* and *T. vulgaris*, while the extract of *R. graveolens* contains less flavonoids. The differences in the amounts of TF in different extracts and fractions could be explained by the fact that presence of phenolics is affected by plant species, maturity at harvest, growing conditions, soil conditions and post harvest treatments [16].

Total tannins and total anthocyanins

Vegetable tannins are natural polyphenols ubiquitously distributed in plants, frequently occurring in a variety of foods such as vegetables, fruits, seeds, and plant-derived beverages. Commercially tannins are used in the wine industry for a multitude of reasons: to stabilize the color of red wines, to ensure palate balance and complexity in wines, to inhibit lacasse in botrytis-infected fruit and to serve as fining agents to reduce protein concentrations [17].

From the data presented in Table 2, it is apparent that tannin content varied significantly (P 0.05) in some plants extract, where tannin was low in *Z. vulgaris* (0.48 mg/g) which also had the lowest level of total phenolics and high in *S. officinalis* (0.70 mg/g).

Anthocyanins are water-soluble pigments responsible for the red and blue colors in some plants. The present study found that there were significant differences evident between study samples (P < 0.05). The highest anthocyanins content in *Z. vulgaris* and *T. vulgaris* were 0.49 and 0.44 mg/100 g, where the smallest values of total anthocyanins (0.02 mg/100g) in *R. graveolens* due to their low total and colored anthocyanins content, have weakly expressed in color.

Antioxidant activity

Owing to the complex reactive facets of phytochemicals, the antioxidant activities of plant extracts cannot be evaluated by only a single method, but at least two test systems have been recommended for the determination of antioxidant activity to establish authenticity [18]. There are many different methods for determining antioxidant function each of which depends on a particular generator of free radicals, acting by different mechanisms [19]. For this reason the antioxidant activity of thyme, black cumin, lavender, parsley and fennel essential oil was determined by three spectrophotometric methods, DPPH and BCB tests.

Antioxidant activity using DPPH radical scavenging

The extracts of *S. officinalis* and *L. officinalis* retained their antioxidant activity of 60.42% and 43.97% scavenging respectively at a concentration of 0.05 mg/ml, while those of the other three plants are less effective (Table 3). There are significant differences (P 0.05) among the study samples (Fig. 4). The extracts of *S. officinalis* and *L. officinalis* exhibited strong DPPH radical scavenging in Table 3 with IC_{50} values at 23.23 and 31.18 μg/ml (Figure. 4).

Table 3. Antioxidant activity by DPPH scavenging and β -carotene bleaching of plants extracts (means ± S.D.).

Extraction	Inhibition of DPPH/% [0.05 mg/ml]	β -carotene bleaching [5 mg/ml]
Z. vulgaris	26.76 ^{e} ± 0.11	41.03 ^{d} ± 6.35
T. vulgaris	31. 07 ^{d} ± 0. 06	53.33 ^{c} ± 1.69
R. graveolens	32.09 ^{d} ± 0.09	57.00 ^{c} ± 3.32
L. officinalis	43.97 ^{c} ± 0. 18	67.77 ^{b} ± 2.44
S. officinalis	60.42 ^{b} ± 0. 08	80.77 ^{b} ± 2.84
VC	95.01 ^{a} ± 0.12	6.37 ^{e} ± 3.12
TBHQ	95.12 ^{a} ± 0.09	84.60 ^{a} ± 2.25

The values are means of three independent analyses

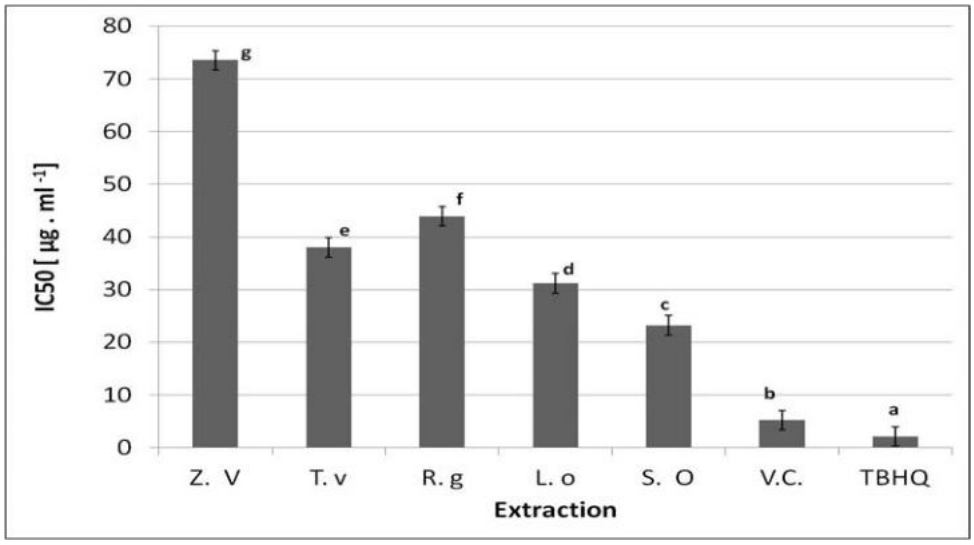

Figure. 4. IC_{50} of extracts from some plants and reference antioxidants (VC and TBHQ)

The extracts of *S. officinalis* and *L. officinalis* show higher scavenging values than *T. vulgaris*, *Z. vulgaris*, and *R. graveolens* with statistically significant differences ($p < 0.05$). These data may be due to the fact that radical-scavenging capacity is directly related to the hydrogen atom donating ability of a compound and not correlated to the redox potentials alone [6]. This also probably due to the presence of high polyphenolic content. There similar correlation of antioxidant activity and phenolic contents [20, 21]. The observed antioxidant potential for these extracts can be related to the presence of various functional groups, such as hydroxyl and carbonyl groups [22, 23].

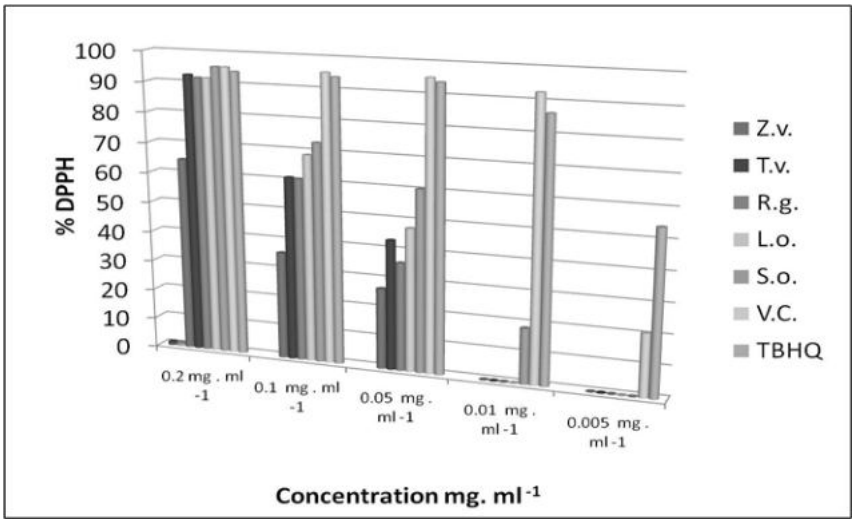

Figure. 5. DPPH radical scavenging activities of extracts from some plants and reference antioxidants (VC and TBHQ)

Some extracts tested in the DPPH assay had good antioxidant properties (Figure. 5). The antioxidant efficiency of the various extracts tested was basically dependent on their concentrations. Their free radical-scavenger effectiveness was in the following ascending order: *Z. vulgaris* *T. vulgaris* *R. graveolens* *L. officinalis* *S. officinalis*.

Antioxidant activity using β-Carotene bleaching assay

The antioxidant activity has also been assessed as ability to prevent β-Carotene from oxidation by linoleic acid. This method usually is used to evaluate the antioxidant activity of compounds in emulsions, accompanied with the coupled oxidation of β-carotene

and linoleic acid. In the BCB assay, the oxidation of linoleic acid generates peroxyl free radicals due to the abstraction of a hydrogen atom from diallylic methylene groups of linoleic acid [24]. There are significant differences (P 0.05) among the study samples (Table 3). The antioxidant activity expressed as AA values decreased in the order *Z. vulgaris* > *T. vulgaris* > *R. graveolens* > *L. officinalis* > *S. officinalis*. Their results showed that antioxidants of *S. officinalis* and *L. officinalis* were better inhibitors of β - carotene bleaching than reference antioxidants like TBHQ.

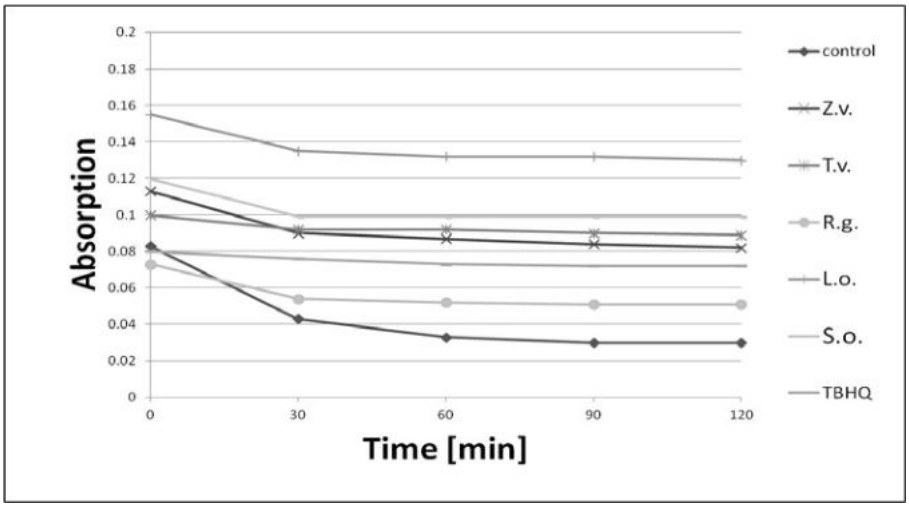

Figure.6. Antioxidant activity of extracts from some plants and standard antioxidants in β-carotene-linoleate bleaching system

The BCB absorbance of *S. officinalis* and *L. officinalis* for the 120 min were 0.130 and 0.099, whereas they were 0.155 and 0.120 at zero time (Figure. 6), indicating their BCB antioxidant activity are stable with time. The extracts of *S. officinalis* and *L. officinalis* exhibited stronger BCB activities than other plants, possibly due to their oil components, such as tocopherols, phytosterols, and phenolic compounds. The synthetic antioxidant, TBHQ has a stronger antioxidant activity when compared to *S. officinalis*, *L. officinalis*, *R. graveolens*, *T. vulgaris*, and *Z. vulgaris* (Fig. 5).
For comparison of DPPH and BCB antioxidant activity methods, *S. officinalis* and *L. officinalis* showed significantly higher (P < 0.05) antioxidant activity than *R. graveolens, T. vulgaris*, and *Z. vulgaris*.

Antibacterial activity

The scientific literatures in food chemistry and technology have emphasized the significance of antibacterial activity of extracts with respect to their application in food conservation which is geared towards providing extension of shelf life of food and their microbial safety. This study investigated the antibacterial activities of seven plant extracts using agar well diffusion method. All tested extracts showed some antibacterial activities against *S. aureus*, *S. pyogenes*, *S. dysenteriae*, and *S. typhimurium*. The results may support the use of *R. graveolens* and *S. officinalis* and *L. officinalis* in traditional medicines.

Presents antibacterial activities of the five plants tested at the concentration of 5, 10, and 20 mg/ml (Table 4).They showed that gram positive bacteria (*Staphylococcus* and *Streptococcus*) were susceptible to extracted materials from *R. graveolens*, *L. officinalis*, *T. vulgaris*, and *S. officinalis*. *S. aureus* was the most sensitive organism to *R. graveolens* and *L. officinalis* extracts at 20 mg/ml than *S. pyogenes.* Gram negative bacteria were susceptible to extracted materials from *R. graveolens.* Extracts of *R. graveolens* and *S. officinalis* were inhibitory for *S. typhimurium* and S. *dysenteriae*. *E.coli* was more inhibited by extract of *S. officinalis* than those of *L. officinalis* and *T. vulgaris* at 20 mg/ml.
Gram negative bacteria were more resistant to extracts of *T. vulgaris* and *L. officinalis* than gram- positive bacteria [25]. The resistance towards antibacterial substances by gram-negative bacteria was related to the lipopolysaccharides in their cell wall [26, 27]. That may explain the different inhibition pattern by R. *graveolens* and *Z. vulgaris* against *S. aureus*.
The results of the present study are encouraging as extracts of *R. graveolens* showed significant antibacterial activity against many enteric pathogens tested. The extract of *R. graveolens* showed pronounced inhibition against *S. aureous*, *S. typhimurium*, *S. pyogenes*, and *S. dysenteriae* with inhibitory zone of 15.0, 15.1, 13.7, and 12.6 mm, respectively.

Table 4. Antibacterial activity of the ethanolic extraction of some plants by agar diffusion assay.

Extraction	Inhibition zone (mm)[1]														
	Escherichia coli (mg/ml)			*Shigella dysenteriae* (mg/ml)			*Salmonella typhimurium* (mg/ml)			*Streptococcus pyogenes* (mg/ml)			*Staphylococcus aureus* (mg/ml)		
	5	10	20	5	10	20	5	10	20	5	10	20	5	10	20
Z. vulgaris	NA*	NA	NA	NA	NA	9.5 ± 0.9	NA	NA	NA	NA	NA	NA	NA	NA	NA
T. vulgaris	NA	9.2 ± 0.4	10.2 ± 0.8	NA	NA	NA	NA	NA	NA	NA	10.0 ± 1.9	11.0 ± 0.9	11.0 ± 1.0	11.3 ± 0.6	12 ± 0.1
R. graveolens	NA	NA	9.3 ± 0.6	10.3 ± 0.6	11.3 ± 0.7	12.6 ± 1.0	10.7 ± 0.6	13.3 ± 1.2	15.0 ± 1.0	11.7 ± 1.0	12.2 ± 0.4	13.7 ± 0.7	11.8 ± 0.3	13.2 ± 2.0	15 ± 0.5
L. officinalis	NA	9.3 ± 0.6	10.2 ± 0.3	NA	NA	NA	NA	NA	9.7 ± 0.6	12.0 ± 0.1	13.0 ± 0.1	11.7 ± 0.6	10.8 ± 1.3	11.7 ± 0.8	11.7 ± 2.0
S. officinalis	NA	NA	11.0 ± 0.1	9.7 ± 0.6	10.8 ± 0.3	12.2 ± 0.3	NA	9.8 ± 0.3	10.2 ± 0.8	NA	9.3 ± 0.1	10.7 ± 0.3	NA	9.8 ± 1.0	11.3 ± 0.6
Reference [2]	5 µg/mL	50 µg/mL	100 µg/mL	5 µg/mL	50 µg/mL	100 µg/mL	5 µg/mL	50 µg/mL	100 µg/mL	5 µg/mL	50 µg/mL	100 µg/mL	5 µg/mL	50 µg/mL	100 µg/mL
Streptomycin	12.3 ± 0.1	15.7 ± 0.9	18.9 ± 1.3	12.0 ± 0.1	15.3 ± 0.6	17.7 ± 0.6	11.3 ± 0.6	13.7 ± 1.2	17.0 ± 2.0	ND	ND	ND	ND	ND	ND
Penicillin	ND**	ND	ND	ND	ND	ND	ND	ND	ND	16.7 ± 0.5	17.3 ± 1.2	18.7 ± 1.0	11.7 ± 1.2	15.7 ± 0.6	15.7 ± 1.0

[1]Values (diameter in mm, including diameter of 6 mm) are expressed as mean ± standard deviation, analyzed individually in triplicate.
[2] Reference streptomycin (gram-negative bacteria), penicillin (gram-positive bacteria)
* NA = not activity; ND** = not done
Inhibition zones:
< 9 mm no active, 9 –12 mm less active, 13–18 mm active, > 18 mm very active.

Conclusions

This study showed that some plants used in traditional medicine in Yemen have antioxidant and antibacterial activities. The types and contents of bioactive components varied among different plants. The characterization of the active components of those plants may lead to full utilization of these plants by the local folks.

Acknowledgements

The study was supported by the 12th Five-Year Plan for Science and Technology Development (No. 2012BAD33B05), Chinese Nature Science Foundation (21403601 and 31201805), Fundamental Research Funds for the Central Universities (JUSRP111A36, JUSRP1052), and Priority Academic Program Development of Jiangsu Higher Education Institutions.

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