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## Original Article

# Anti-oxidant activity and major chemical component analyses of twenty-six commercially available essential oils



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## ABSTRACT

This study analyzed 26 commercially available essential oils and their major chemical components to determine their antioxidant activity levels by measuring their total phenolic content (TPC), reducing power (RP),  $\beta$ -carotene bleaching (BCB) activity, trolox equivalent antioxidant capacity (TEAC), and 1,1-diphenyl-2-picrylhydrazyl free radical scavenging (DFRS) ability. The clove bud and thyme borneol essential oils had the highest RP, BCB activity levels, and TPC values among the 26 commercial essential oils. Furthermore, of the 26 essential oils, the clove bud and ylang ylang complete essential oils had the highest TEAC values, and the clove bud and jasmine absolute essential oils had the highest DFRS ability. At a concentration of 2.5 mg/mL, the clove bud and thyme borneol essential oils had RP and BCB activity levels of  $94.56\% \pm 0.06\%$  and  $24.64\% \pm 0.03\%$  and  $94.58\% \pm 0.01\%$  and  $89.33\% \pm 0.09\%$ , respectively. At a concentration of 1 mg/mL, the clove bud and thyme borneol essential oils showed TPC values of  $220.00 \pm 0.01$  and  $69.05 \pm 0.01$  mg/g relative to gallic acid equivalents, respectively, and the clove bud and ylang ylang complete essential oils had TEAC values of  $809.00 \pm 0.01$  and  $432.33 \pm 0.01$   $\mu$ M, respectively. The clove bud and jasmine absolute essential oils showed DFRS abilities of  $94.13\% \pm 0.01\%$  and  $78.62\% \pm 0.01\%$ , respectively. Phenolic compounds of the clove bud, thyme borneol and jasmine absolute essential oils were eugenol (76.08%), thymol (14.36%) and carvacrol (12.33%), and eugenol

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(0.87%), respectively. The phenolic compounds in essential oils were positively correlated with the RP, BCB activity, TPC, TEAC, and DFRS ability.

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## 1. Introduction

Free radicals are highly reactive molecules with unpaired electrons that can cause various oxidative stresses [1,2]. Oxidative stress involves the generation of reactive oxygen and nitrogen species. Such species have been implicated in aging and various pathological processes [3,4] because they damage the structures of cells, lipids, membranes, proteins, and DNA [5]. To reduce the damages caused by reactive species, butylated hydroxyl anisole (BHA) and butylated hydroxyl toluene (BHT) are widely used as antioxidant additives; however, they have been extensively examined because of their potential toxicity [6,7]. Therefore, natural antioxidants have attracted increased interest because natural ingredients may be safer than synthetic ingredients [8].

Essential oils are natural, volatile complex compounds characterized by the odor of their corresponding aromatic plants, which synthesize them as secondary metabolites [9]. Numerous essential oils not only serve as food and cosmetic additives but also exhibit antimicrobial [10,11] and antioxidant properties [12]. In particular, phenolic compounds in essential oils are very effective free radical scavengers [13,14]. Factors such as reducing power (RP) [15], total phenolic content (TPC) [16],  $\beta$ -carotene bleaching (BCB) activity [17–19], trolox equivalent antioxidant capacity (TEAC) [20–22], and 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging (DFRS) ability [23,24] have been evaluated to investigate the antioxidant or free radical scavenging abilities of foods, plant extracts, and essential oils.

The composition of essential oils substantially varies with different aspects, such as the manufacturer, harvesting time, and plant materials. However, because commercially available essential oils are used by people, it is essential to investigate whether these oils have good antioxidant activity (or their orders) as well as to elucidate the chemical components contributing to their observed antioxidant abilities. In this study, we studied more than 200 essential oils from Australian companies [25,26] and compared their antioxidant activities. To explore the sources of essential oils in functional foods, and their applications in cosmetic products and to investigate their TPC and antioxidant activities, factors such as RP, BCB activity, TEAC, and DFRS ability were evaluated. In addition, we assessed the antioxidant activities and analyzed the major chemical components of 26 essential oils obtained from Ayus GmbH (Baden, Germany).

## 2. Materials and methods

### 2.1. Raw materials and chemicals

DPPH, BHT, BHA and eugenyl acetate were purchased from TCI. (Shanghai, China) and 3,4,5-trihydroxybenzoic acid

anhydrous (gallic acid) was purchased from Lancaster (England). The Folin–Ciocalteu phenol reagent (2N), eugenol, borneol, benzyl acetate, and potassium hexacyanoferrate were procured from Merck (Darmstadt, Germany). Moreover, 2,2'-azino-bis (3-ethyl-benzothiazoline-6-sulfonic acid) diammonium salt (ABTS) and  $\beta$ -carotene (type I, synthetic) were purchased from Sigma (St. Louis, MO, USA). Linoleic acid, thymol, L (+)-ascorbic acid, and benzyl benzoate were obtained from Acros Organics (Geel, Belgium). Sodium carbonate, polyoxyethylene sorbitan monopalmitate (Tween-40), sodium dihydrogen phosphate anhydrous, disodium hydrogen phosphate, and iron (III) chloride hexahydrate were procured from Showa (Tokyo, Japan). Carvacrol and *p*-cymene were purchased from SAFC (USA) and Fluka (Buchs, Switzerland), respectively. Trichloroacetic acid (TCA) was obtained from Alfa Aesar (Karlsruhe, Germany). The 26 essential oils were purchased from Ayus GmbH (Baden, Germany) in their origin form. Trichloromethane and all other chemicals and solvents were of standard analytical grade and were procured from Echo Chemical Co. (Miaoqi, Taiwan).

### 2.2. Gas chromatography-mass spectrometry

The volatile compounds were analyzed using a Thermo GC-MS system (GC-MS Trace DSQ-Mass Spectrometer, MSD 201351, Thermo, Minneapolis, MN, USA). An Equity<sup>TM-5</sup> capillary column (length, 30 m; inside diameter, 0.25 mm; film thickness 0.25  $\mu$ m; Supelco, USA) was used. The oven temperature was programmed as follows: isothermal at 40 °C, and then increased to 100 °C at 5 °C/min, and held for 5 min. Subsequently, the temperature was increased to 250 °C at 5 °C/min and held for 20 min. Helium (1 mL min<sup>-1</sup>) was used as carrier gas. The injection port and detector temperatures were maintained at 250 °C. The sample components were ionized in electron ionization mode (70 eV). The injection volume was 1  $\mu$ L of essential oil (100 ppm in ethanol [EtOH] 99.95%). The linear retention indices (RIs) for all compounds were determined by co-injecting the samples with a solution containing a homologous series of C8–C22 n-alkanes [25]. The individual components were identified comparing their RIs with those of known compounds reported in the literature, and by matching their mass spectra with those of the known compounds or the Trace DSQ-MASS spectral database (Thermo, USA).

### 2.3. TPC determination

The TPC was determined using a previously reported method [28] with some modifications involving the Folin–Ciocalteu reagent, and gallic acid was used as the standard. The reaction mixture included 0.5 mL of essential oil (10 mg/mL EtOH), 1 mL of Folin–Ciocalteu reagent, and 1 mL of Na<sub>2</sub>CO<sub>3</sub> (7.5%)

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