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Chemical ingredients and antioxidant activities of underutilized wild fruits

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ABSTRACT

This study evaluated nutritional values, bioactive constituents and antioxidant activities of the five wild underutilized fruits in the mountains of southwest Saudi Arabia (*Coccinia grandis* (L.) Voigt, *Diospyros mespiliformis* Hochst. Ex A.Dc., *Cissus rotundifolius* (L.), *Ephedra foeminea* Forssk., and *Grewia villosa* Willd.). The moisture content, crude fibers, total protein, total lipids, total hydrolyzable carbohydrate, total soluble sugars, and total free amino acids were analyzed. The results showed varying amounts among fruits of the five study species. In addition, the mineral composition, amino acid content, phenolic compounds, vitamins, and antioxidant activity were assessed. The highest content of total phenolic and total tannins was measured in *D. mespiliformis* (20.69 mg/ g and 3.84 mg/g) and the lowest in *E. foeminea* (10.83 mg/g and 1.44 mg/g), respectively. The methanol extract (1 mg/ml) of *D. mespiliformis* displayed higher percentage of DPPH radical scavenging activity (87.36%). The sufficient nutritional and antioxidant activity and hydrogen peroxide scavenging activity (71.53%). The sufficient nutritional and antioxidant value of these wild fruits provide healthy food source for the local residents, much the same as many cultivated fruits and vegetables.

1. Introduction

Antioxidants play an important role in health-promoting biochemical pathways. Oxidative stress, resulting from imbalance among the reactive oxygen species including free radicals and antioxidant defenses in living organisms produces oxidative changes to proteins, fatty acids, and DNA molecules in the living cells, which encourage the initiation of ailments, e.g., inflammation, liver cirrhosis and vascular diseases (Aruoma, 1998).

The consumption of healthy components is a regular way of preventing different ailments and increases the use of natural ingredients. Wild edible plants with its high content of bioactive constituents comparing to different cultivated species represent good nutritional and healthy sources for native and rural regions (Ruiz-Rodríguez et al., 2014). Wild underutilized fruits have important economic and nutritional value since they represent also an important part of the diet for native people (Hegazy and Lovett-Doust, 2016); being a rich source of vitamins, carbohydrates, fats, proteins, fibers, and minerals (Hegazy et al., 2013). Wild fruits, distributed in different geographical areas including *Balanites aegyptiaca* L., *Ziziphus oenophia, Adansonia digitata, Aegle marmelos, Phyllanthus emblica, Diospyros decandra* Lour., and *Spon-dias pinnata* contain different bioactive compounds including phenolics, flavonoids, carotenoids, tannins, and vitamins which hold potent anti-oxidant activity, which could supply the main nutraceuticals, dietary supplements and functional foods for the native people of such areas, as reviewed by Fernández-Ruiz et al. (2017).

The five wild fruits investigated in this study include: (1) *C. grandis* (L.) Voigt (Cucurbitaceae) fruits are used as a foodstuff which may be eaten raw in vegetable salad or cooked. *Coccinia* fruits contain several chemical ingredients such as taraxerone, cryptoxanthin, taraxerol, β -carotene, (24R)-24-ethylcholest-5-en-3 β -ol glucoside, and β -sitosterol, and it exhibited varied biological effects like anti-inflammatory, analgesic, antidiabetic, antipyretic, antioxidant, hypoglycemic, hep-atoprotective, and anticancer (Pekamwar et al., 2013; Doka et al., 2014). (2) *D. mespiliformis* Hochst. Ex A.Dc. (Ebanaceae) fruits are edible as

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nutritive food. Tannins, alkaloids, saponins, flavonoids, glycosides, and Steroids are the major phytochemicals identified in D. mespiliformis extracts (Ebbo et al., 2014). It used in traditional medicine for wound dressing, syphilis, anthelmintic and fever (Dangoggo et al., 2012). (3) C. rotundifolia L. (Vitaceae) is commonly used as food thickeners (Korish, 2016). Flavonoids, quinolizidine alkaloids, tritepenes, stilbenes, tannins, steroids, coumarin, and saponins are the main chemical contituents identified in Cissus etracts (Fernandes and Banu, 2012), and it has shown anti-diabetic, antioxidant and anti-parasitic activity (Onyechi et al., 1998; Nagani et al., 2011). (4) E. foeminea Forssk. (Ephedraceae) fruits contain several phytochemical compounds such as ephedrine, methylephedrine, methyl pseudoephedrine, ephedroxane, and macrocyclic spermidines, beside kynurenates, citric, malic and oxalic acid phenolic compounds which are used as medicine for different diseases (Ibragic and Sofic, 2015). (5) G. villosa Willd. (Tiliaceae) ripe fruits are either eaten fresh or dry and also used in sorbet. Preliminary phytochemical analysis of different Grewia species revealed that their different extracts generally contain alkaloids, triterpenoids, flavonoids, fatty acids, saponins, tannins and steroids and are used as medicine for dysentery, cholera, wounds, and sores (Goyal, 2012).

In this study, we assessed the nutritional value, chemical composition and *in vitro* antioxidant activities of ripe fruits of the five wild species, which are consumed as edible fruits by the mountain dwellers in the western side of Saudi Arabia.

2. Materials and methods

2.1. Fruit collection

Ripe fruits of *C. grandis*, *D. mespiliformis*, *C. rotundifolius*, *E. foeminea*, *and G. villosa* were collected from Shada Mountain, southwest Saudi Arabia during the fruit ripening season (May 2014). The collected fruits were kept in tight plastic bags in ice box and carried to the laboratory for freeze-drying, and stored at -20 °C until analyzed.

2.2. Chemicals

2,2-diphenyl-1-picrylhydrazyl (DPPH), Folin-Ciocalteu reagent, sodium phosphate, ammonium molybdate, β -carotene, ascorbic acid, 2,6dichlorophenolindophenol, and gallic acid were purchased from Sigma-Aldrich (St. Louis, Missouri, United States). All other chemicals and solvents were of analytical grade.

2.3. Proximate composition

The proximate analysis of fruit samples including the moisture content, crude fiber, total proteins, total lipids, total hydrolyzable carbohydrate, total soluble sugars, and total free amino acids were assessed. Moisture content was determined by drying the fruits at 80 °C in an oven until constant weight was obtained (AOAC, 2002).

Crude fiber was determined by the loss in weight on ignition of dried residue following the digestion of fat-free samples with 1.25% each of sulfuric acid and sodium hydroxide solutions (AOAC, 2002). The total protein content (N \times 6.25) was estimated by the macro-kjeldahl nitrogen assay method using a digestion apparatus combined with the photo-colorimetric method described by Baethgen and Alley (1989). The total lipids content was determined according to AOAC (1990), by n-hexane extraction using an automatic Soxhlet analyser (Soxtherm 2000 Automatic, C. Gerhardt UK). Total hydrolyzable carbohydrate was determined colorimetrically according to Dubois et al. (1956). Total soluble sugars was extracted by boiling in 80% ethanol for 6 h and determined using the phenol-sulfuric acid method according to Dubois et al. (1956). Total free amino acids were hydrolyzed by acid hydrolysis and measured according to Etsushiro et al. (1981).

2.4. Mineral composition

The mineral composition was determined on the dry weight basis. The mineral ions Ca, Mg, Fe, Zn, and Cu were measured after wetdigestion by atomic absorption (Model Varian, spectra AA220- Atomic Absorption Spectrophotometer). The Na and K were measured by using flame photometry, P was measured colorimetrically (UV-visible spectrophotometer) using potassium dihydrogen phosphate standard (AOAC, 2000).

2.5. Amino acid composition

The amino acids composition was estimated following the methods of Spackman et al. (1958) and expressed as mg/g protein. Amino acids were determined by reaction with ninhydrin using Biochrom 20 amino acid analyzer (Pharmacia Biotech, Cambridge, England) equipped with a 90 \times 4.6 mm PEEK sodium pre-wash column and 250 \times 4.6 mm Bio PEEK sodium high performance column (Pharmacia Biotech, Cambridge, England) after acid hydrolysis (6 M HCl, 110 °C, 24 h).

2.6. Total phenolic content

Total phenolic content was determined in methanol extracts of freezedried fruits using Folin-Ciocalteu reagent by spectrophotometer at 750 nm, according to the method of Singleton and Rossi (1965). The results were expressed as mg Gallic acid equivalents (GAE)/g, dry-weight basis.

2.7. Total tannin content

Total tannins were determined by copper acetate gravimetric method, which depends on quantitative precipitation of tannins with copper acetate solution, then ignition of copper tannate to copper oxide and weighing the residual copper oxide. The amount of tannins (mg/g, dryweight basis) was calculated as each one gram copper oxide is equivalent to 1.305 g tannins (Ali et al., 1991).

2.8. Total anthocyanin content

The freeze-dried fruits were blended in 100 ml 0.1 N hydrochloric acid in ethanol (15:85, v/v) and kept overnight at 43 °C then filtered through Whatman No. 1 filter paper. The absorbance of an aliquot of filtrate was measured at 535 nm and converted to anthocyanin contents (mg/g) using the formula proposed by Fuleki and Francis (1968).

2.9. Total carotenoid content

The carotenoid content of fruits was measured in 85% aqueous acetone extract of the freeze-dried fruits according to Fahmy et al. (1990). The optical density of the extract was measured at 453, 505, 645 and 663 nm. The content of total carotenoid was calculated according to the following equation:

Total carotenoid (mg β -carotene equivalents/g) = (0.216 \times A663) – (1.220 \times A645) – (0.304 \times A505) + (0.452 \times A453)

where A is the absorbance of the sample.

2.10. Vitamin C content

The freeze-dried fruits were extracted with 10 ml metaphosphoric acid (1%) for 45 min at room temperature and filtered. One milliliter of filtrate was mixed with 9 ml of 2,6-dichlorophenolindophenol and the absorbance was measured spectrophotometrically at 515 nm (Klein and Perry, 1982). The results were expressed as μ g L-ascorbic acid equivalents (AAE)/100 mg fresh-weight.

2.11. Vitamin A content

Vitamin A was determined spectrophotometrically according to AOAC (2002). Five grams of the freeze-dried fruits were homogenized with 100 ml of diethyl ether for 10 min in a shaker and filtered. Additional 100 ml of 95% ethanol was added to the residue and shaken for 30 min and filtered. The reaction colors were recorded and its absorbance measured at 452 nm. The results were expressed as $\mu g \beta$ -carotene/100 mg fresh-weight.

2.12. Antioxidant activities

2.12.1. DPPH scavenging activity

The capacity of different concentrations (0.2-1.0 mg/ml) of fruit methanolic extracts to scavenge the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical was evaluated following Oliveira et al. (2011). The reduction of DPPH radical was determined by measuring absorbance at 517 nm against a blank without DPPH. The DPPH scavenging effect (%) was calculated by using the following equation:

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

where A_{DPPH} is the absorbance of the DPPH solution without the extract and A_S is the absorbance of the sample extract solution. The antioxidant activity of standard concentration of vitamin E was assayed as positive control.

2.12.2. Total antioxidant activity

The total antioxidant activity of fruit methanolic extract was assayed following Pan et al. (2007). One ml of the extract was combined with 3 ml reagent solution (0.6 M H₂SO₄, 28 mM sodium phosphate and 4 mM ammonium molybdate). The reaction mixture was incubated at 95 °C for 150 min, and after cooling at room temperature; the absorbance was measured at 695 nm against blank or the reaction mixture without the extract. The absorbance was measured every 30 min for a total of 150 min. The results were expressed as absorbance (nm) either for the samples and positive control. The catechol (0.5 mg/ml) was used as positive control.

2.12.3. Hydrogen peroxide scavenging activity

The ability of fruit methanolic extracts to scavenge H2O2 was estimated according to Zhao et al. (2006). The relative activity to scavenge hydrogen peroxide was expressed as percentage of the titer volume change as following:

$$H_2O_2$$
 Inhibition (%) = [(A_{control} - A_{sample}) / A_{control}] × 100

where A_{control} is the titer volume of the mixed solution without the extract and Asample is the titer volume of the solution with added extract.

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2.13. Statistical analysis

All the data were recorded in five replicates and expressed as mean \pm standard error (SE) using Microsoft Excel 2010.

3. Results and discussion

3.1. Nutritional value

The results of Table 1 revealed significant variation in the nutritional contents of the investigated fruits. The moisture content of these fruits ranged from 61.03% in G. villosa to 88.15% in C. rotundifolius. The physical properties of the fruits such as size, viscosity, weight, and bulk density are affected by moisture content. The physical properties assessment could be helpful in fruit harvesting, transportation, storage, and processing operations. The moisture content of 61.03% in G. villosa in the present study was higher than the result obtained by Elhassan and Yagi (2010) for fruits of Grewia tenax (13%), G. flavescens (15%) and G. villosa (14%). In the present study, the moisture content in C. grandis (86.76%), C. rotundifolius (88.15%) and E. foeminea (85.04%) was higher than the moisture content of some cultivated fruits including Banana (74.91%), Mango (81%) and Morinda tinctoria (78.34%), as reported by Ajay et al. (2012), however it is lower than the moisture contents (95.4%, 94.42% and 92.86%) of wild fruits Ziziphus mauritiana, Lannea schimperi, and Gardenia aqualla, respectively (Lockett et al., 2000).

Total carbohydrates, soluble sugars, and protein are the vital nutrients in many fruits as they are the main source of energy. The highest content of total protein, hydrolyzable carbohydrates, total soluble sugars, and free amino acids were found in D. mespiliformis as 9.28, 15.88, 9.82 and 2.95%, respectively, followed by C. grandis (7.6, 12.26, 8.14 and 2.4%), while the lowest contents (2.04, 5.04, 3.49 and 0.61%) were found in G. villosa, respectively (Table 1). Proteins are well known as bodybuilding biomolecules because it contains essential amino acids which are important for human growth and health. The carbohydrate and protein content in the present study is higher than that reported by Tripathy et al. (2014) in C. grandis (4.42% and 1.26%) and Luffa acutangula (5.1% and 1.37%) respectively. Additionally, crude protein content of the studied wild fruits was higher than other cultivated fruits, for example, grapes, mango, papaya, orange and banana which have a range of 0.5-1.2 g/100 g crude proteins (Rathore, 2009), however it is lower than the protein contents (15.21%, 11.70% and 7.76%) of wild fruits Ximenia americana, Strychnos spinosa, and Lannea schimperi, respectively (Lockett et al., 2000). The content of protein (9.28%) in D. mespiliformis in the present study is lower than that of D. mespiliformis (12.44%) fruits collected from Nigeria (Akinyemi and Kayode, 2012). The protein, carbohydrates and soluble sugars content of G. villosa in the present study were lower than that reported by Elhassan and Yagi (2010) for G. tenax

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	C. grandis	D. mespiliformis	C. rotundifolius	E. foeminea	G. villosa
Nutritional content					
Moisture (%)	86.76 ± 2.73	80.29 ± 02.69	88.15 ± 2.16	85.04 ± 2.08	61.03 ± 1.51
Fibers (%)	6.84 ± 0.84	4.81 ± 01.08	3.65 ± 0.84	$\textbf{3.44} \pm \textbf{0.34}$	5.38 ± 1.03
Total protein (%)	7.60 ± 2.60	9.28 ± 1.14	5.06 ± 1.13	4.30 ± 0.87	2.04 ± 0.27
Total lipids (%)	5.51 ± 0.15	4.58 ± 0.5	2.23 ± 1.06	2.18 ± 0.69	0.63 ± 0.12
Total hydrolyzable carbohydrates (%)	12.26 ± 2.86	15.88 ± 3.42	9.85 ± 1.58	8.73 ± 1.62	5.04 ± 0.83
Total soluble sugars (%)	8.14 ± 1.34	9.82 ± 2.07	6.94 ± 1.19	5.82 ± 0.85	$\textbf{3.49} \pm \textbf{0.66}$
Total free amino acids (%)	2.40 ± 0.46	2.95 ± 0.77	0.92 ± 0.24	1.86 ± 0.28	0.61 ± 0.15
Phytochemical content					
Total phenolic (mg/g)	18.25 ± 3.25	20.69 ± 1.62	14.06 ± 2.07	10.83 ± 1.40	17.39 ± 2.06
Total tannin (mg/g)	3.15 ± 0.35	3.84 ± 0.32	2.68 ± 0.45	1.44 ± 0.21	2.06 ± 0.38
Total anthocyanin (mg/g)	0.28 ± 0.06	0.26 ± 0.04	0.22 ± 0.04	0.14 ± 0.03	0.16 ± 0.02
Total carotenoid (mg/g)	35.19 ± 3.19	23.82 ± 3.12	22.06 ± 2.73	18.33 ± 2.28	$\textbf{7.49} \pm \textbf{1.58}$
Vitamin C (µg/100 mg)	896.41 ± 42.81	709.52 ± 23.84	612.14 ± 40.16	339.15 ± 16.41	241.70 ± 20.74
Vitamin A (µg/100 mg)	856.74 ± 46.14	597.41 ± 28.69	525.36 ± 33.17	501.42 ± 47.37	306.06 ± 18.4

Each value is a mean of five replicates \pm SE (standard error).

(7.7, 66 and 13.8%), G. flavescens (8.7, 75 and 10%) and G. villosa (6.7, 84 and 10.4%) fruits, respectively. The investigated fruits may play an important role in providing energy for the rural and tribal communities as content. The highest percentage content of fiber and lipid (6.84 and 5.51%) were found in C. grandis (Table 1). These values were higher than that found in fruits of different wild cucurbits especially C. grandis (Tripathy et al., 2014). The content of fiber and lipid (5.38 and 0.63%) in G. villosa in the present study were lower than that reported by Elhassan and Yagi (2010) for G. tenax (20.5 and 1.7%), G. flavescens (42.8 and 1.3%) and G. villosa (25.5and 1.5%) fruits, respectively.

3.2. Mineral composition

The macro and micro elements of the investigated wild fruits are shown in Table 2. Potassium was the prominent macro element in the investigated fruits with the highest value 418.42 mg/100 g in C. rotundifolius followed by 412.61 mg/100 g in C. grandis. These values of potassium are, lower than (2308-2392 mg/100 g) of wilf fruit Adansonia digitata (Lockett et al., 2000), and superior to banana (348-370 mg/100 g), the common potassium enriched fruit (Yahia et al., 2011). In this context, Korish (2016) reported 8.09 mg/g potassium in the leaves of C. rotundifolia, also Akinyemi and Kayode (2012) reported 16.31% potassium content in Nigerian D. mespiliformis fruits. The potassium content in the present study (153.08 mg/100 g) in G. villosa was lower than (817–966 mg/100 g) that reported in the fruits of different Grewia spp. by Elhassan and Yagi (2010).

The highest amount of calcium, 102.58 mg/100 g (Table 2), was found in C. rotundifolius followed by C. grandis (102.46 mg/100 g) and G. villosa (92.61 mg/100 g). These values were lower than 296–790 mg/ 100 g that was reported in the fruits of different Grewia spp. (Elhassan and Yagi, 2010); and (58.62%) in D. mespiliformis fruits (Akinyemi and Kayode, 2012). The amount of magnesium and sodium in the five wild fruits studied was in the ranges of 12.08-31.15 and 8.05-26.92 mg/100 g, respectively (Table 2), which were lower than that of D. mespiliformis fruits (Akinyemi and Kayode, 2012) and C. rotundifolia leaves (Korish, 2016). The high potassium and low sodium contents found in all five wild fruits can help minimize potassium deficiency and maintain optimum sodium-potassium balance in the body (Siddig and Greiby, 2013). The studied wild fruits contained significant amount of phosphorus and iron (Table 2). The iron and zinc contents were higher than that of the tropical and sub-tropical fruits (Yahia et al., 2011) whereas they are lower than of D. mespiliformis fruits (Akinyemi and Kayode, 2012) and C. rotundifolia leaves (Korish, 2016).

The detected amount of iron in the investigated fruits (Table 2) also is higher than that reported in Satkara fruit and Taikor fruit (Islam et al., 2015). Iron plays an essential role in preservation of normal functioning of the central nervous system, hemoglobin formation and in the oxidation of carbohydrates, fats, and proteins (Sajib et al., 2014). Deficient consumption of minerals in the food may increase susceptibility to infectious diseases due to the weakening of the immune system (Sajib et al., 2014). Therefore, the five wilds fruits investigated in the present study may be

considered as a good source for both essential macro and microelements which are important to improve the human body immune system to resist many diseases.

3.3. Amino acid composition

The amino acid composition of the wild fruits, including total contents of amino acids and 14 free amino acids are presented in Table (3). The highest amount of total amino acids (114.61 mg/g) was found in C. grandis fruit that is higher than (77.36 mg/g) the measured amounts in C. grandis raw fruits from Sudan (Doka et al., 2014). The total amino acid content in C. rotundifolius (80.89 mg/g) and G. villosa (25.95 mg/g) fruits in the present study were lower than that reported by Korish (2016) and Elhassan and Yagi (2010) in the same fruits. Among the detected amino acids, seven essential amino acids (EAAs) and seven non-essential amino acids (non-EAAs) were detected. Most of the study fruits contained sufficient amounts of EAAs with high amount of threonine, lysine, leucine, and isoleucine. Aspartic acid represented the major component of the non-EAAs, followed by tyrosine in the study fruits except in the G. villosa, where the tyrosine showed values higher than aspartic acid (Table 3). The highest amount of aspartic acid (26.84 mg/g), glycine (10.95 mg/g), and serine (8.58 mg/g) were observed in C. grandis as compared to the other fruits, it was higher than that estimated in C. grandis raw fruit from Sudan (Doka et al., 2014) and lower than that reported in C. rotundifolius leaves by Korish (2016). The amounts of non-EAAs in G. villosa fruits in the present study were relatively lower when compared with results of Elhassan and Yagi (2010) for the same fruits collected from Sudan and this variation may be due to the different environmental conditions and production practices in these different geographical regions.

3.4. Phytochemical components

The total phenolic content reached the highest value (20.69 mg/g) in D. mespiliformis and the lowest (10.83 mg/g) in E. foeminea (Table 1). The content of total phenolic (18.25 mg/g) in C. grandis in the present study is higher than that content (15.47 mg/g) reported by Meenatchi et al. (2017) in the same fruit from India. Tannins are generally defined as naturally occurring polyphenolic compounds of high molecular weight to form complexes with the proteins. As appears in Table 1, the highest tannins content was measured in D. mespiliformis (3.84 mg/g) and the lowest in *E. foeminea* (1.44 mg/g). The total tannins content (2.06 mg/g) in G. villosa fruit in this study is lower than that tannin content (2.46%) reported by Elhassan and Yagi (2010) for the same fruit collected from Sudan. Total anthocyanin contents (mg/g) ranged from 0.14 in E. foeminea to 0.28 in C. grandis (Table 1).

For total carotenoids, the C. grandis showed the highest content of 35.19 mg/g among the fruits analyzed. Simonne et al. (1993) observed a wide range of β -carotene (1–190 mg/g dry weight) among various plants. Carotenoids may act as a singlet oxygen quencher and can transfer one electron to the radicals, giving rise to a stable carotenoid radical cation regenerating the original molecule (Mortensen and Skkibsted, 1997).

Mineral	composition	(mg/100)	g) of i	nvestigated	wild	fruits.

Mineral	C. grandis	D. mespiliformis	C. rotundifolius	E. foeminea	G. villosa
Macroelements					
Sodium	13.18 ± 0.59	26.92 ± 1.18	14.05 ± 0.52	13.41 ± 0.86	8.05 ± 0.65
Potassium	412.61 ± 40.06	$\textbf{389.38} \pm \textbf{18.36}$	418.42 ± 34.16	358.07 ± 30.41	153.08 ± 8.14
Calcium	102.46 ± 6.39	89.50 ± 5.61	102.58 ± 4.73	$\textbf{72.16} \pm \textbf{5.26}$	92.61 ± 5.28
Magnesium	31.15 ± 3.15	23.95 ± 2.06	$\textbf{27.25} \pm \textbf{2.42}$	28.06 ± 3.36	12.08 ± 1.84
Phosphorus	58.33 ± 4.18	$\textbf{42.69} \pm \textbf{4.45}$	46.59 ± 3.15	$\textbf{47.19} \pm \textbf{4.63}$	18.06 ± 1.07
Microelements					
Iron	4.60 ± 0.37	4.59 ± 0.59	4.47 ± 0.33	3.11 ± 0.48	2.61 ± 0.58
Zinc	1.20 ± 0.08	0.24 ± 0.03	0.92 ± 0.05	0.47 ± 0.02	0.16 ± 0.03
Copper	0.35 ± 0.03	0.28 ± 0.02	0.33 ± 0.01	0.22 ± 0.01	0.14 ± 0.03

Each value is a mean of five replicates \pm SE (standard error).

Table 3

Amino acids composition (mg/g) of investigated wild fruits.

Amino acids	C. grandis	D. mespiliformis	C. rotundifolius E. foeminea		G. villosa	
Essential Amino Acids (EA	As)					
Threonine	11.18 ± 2.15	9.48 ± 1.20	3.92 ± 0.17	3.88 ± 0.16	1.39 ± 0.31	
Lysine	8.66 ± 0.92	6.62 ± 1.24	3.46 ± 0.41	3.17 ± 0.41	2.04 ± 0.27	
Leucine	$\textbf{7.01} \pm \textbf{0.08}$	$\textbf{7.84} \pm \textbf{1.72}$	6.85 ± 0.06	$\textbf{4.17} \pm \textbf{0.04}$	2.07 ± 0.38	
Isoleucine	6.71 ± 1.07	2.81 ± 0.63	$\textbf{7.08} \pm \textbf{0.82}$	3.14 ± 0.08	2.06 ± 0.58	
Valine	5.43 ± 0.51	3.94 ± 0.62	5.04 ± 0.47	4.82 ± 0.60	1.44 ± 0.17	
Phenylalanine	1.71 ± 0.07	$\textbf{2.49} \pm \textbf{0.54}$	2.16 ± 0.26	1.03 ± 0.07	1.01 ± 0.13	
Histidine	-	1.08 ± 0.25	1.05 ± 0.06	0.08 ± 0.01	-	
Total EAAs	40.7	33.9	29.56	20.29	10.01	
Non-Essential Amino Acids	(Non-EAAs)					
Aspartic acid	26.84 ± 2.71	20.75 ± 03.16	20.15 ± 1.83	16.34 ± 1.05	2.32 ± 0.85	
Tyrosine	11.23 ± 1.03	11.18 ± 1.70	12.48 ± 1.14	10.62 ± 1.10	6.02 ± 1.05	
Alanine	9.57 ± 1.07	13.08 ± 2.15	$\textbf{2.76} \pm \textbf{0.16}$	-	0.54 ± 0.08	
Glycine	10.95 ± 1.69	8.55 ± 1.08	8.14 ± 1.07	5.09 ± 0.59	3.81 ± 0.68	
Serine	8.58 ± 1.07	$\textbf{7.92} \pm \textbf{1.64}$	1.86 ± 0.25	-	2.19 ± 0.66	
Glutamine	5.38 ± 0.63	8.50 ± 1.86	4.72 ± 0.64	0.61 ± 0.04	0.53 ± 0.07	
Arginine	1.36 ± 0.08	1.50 ± 0.18	1.22 ± 0.06	1.15 ± 0.08	0.53 ± 0.07	
Total Non-EAAs	73.91	71.48	51.33	33.81	15.94	
Total amino acids	114.61	105.38	80.89	54.1	25.95	

Each value is a mean of five replicates \pm SE (standard error).

The high or low β -carotene content observed in the present study could be attributed to various factors such as physiological and morphological characteristics of the plant and environmental conditions. Vitamin C and Vitamin A also exhibited similar trends. The *C. grandis* showed the highest content (896.41 ± 42.81 µg/100 mg and 856.74 ± 46.14 µg/100 mg) of vitamin C and vitamin A, respectively (Table 1). The content of vitamin C in the ripe fruit of *C. grandis* in this study is higher than that content (122.4 mg/kg) in unripe (green) fruit of *C. grandis* collected from Sudan as reported by Doka et al. (2014).

3.5. Antioxidant activities

3.5.1. Free radical scavenging activity by DPPH

The scavenging activity of five wild fruits extracts against DPPH was concentration-dependent (Fig. 1). Among different investigated wild fruits, the *D. mespiliformis* displayed higher percentage of DPPH radical scavenging activity (87.36%), at 1 mg/ml concentration. The methanol extract of *C. grandis* also showed high DPPH scavenging effect (82.46%) at 1 mg/ml concentration. Corresponding to this result, Meenatchi et al. (2017) reported that *C. grandis* exhibited DPPH scavenging effect (60%)

at 250 μ g/ml concentration. It is interesting to note that the methanol extracts of these wild fruits displayed antioxidant activity higher than 50% even at low concentration of 0.6 mg/ml.

3.5.2. Total antioxidant activity

The total antioxidant activity method is based on the reduction of Mo (VI) to Mo (V) by the antioxidant compounds, with the formation of a green phosphate complex with a maximal absorption at 695 nm (Pan et al., 2007).

The total antioxidant activity of the extract (equivalent to catechol standard) ranged between 0.510 to 0.350 Abs, at 150 min (Fig. 2). The *C. grandis* showed the highest total antioxidant activity (0.510 Abs), whereas *G. villosa* showed the lowest antioxidant activity (0.350 Abs). In addition, total antioxidant activity of all extracts was found to increase in a dose-dependent manner.

3.5.3. Hydrogen peroxide scavenging activity

The scavenging activity of hydrogen peroxide, as measured in concentration-dependent manner, was comparable to that of the standard vitamin E (Fig. 3). The activity of the different concentrations of

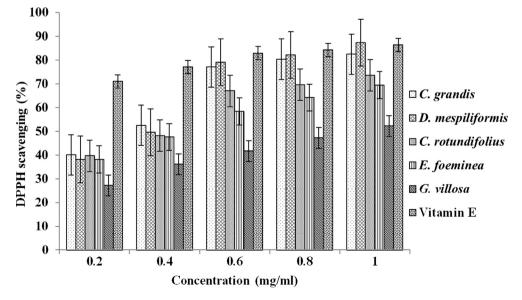


Fig. 1. DPPH scavenging effect (%) of different concentrations of methanol extracts of investigated wild fruits. Each value is a mean of five replicates ±SE (standard error).

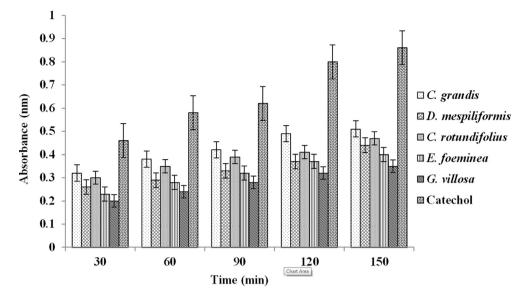


Fig. 2. Total antioxidant activity of methanol extracts of investigated wild fruits at different times. Each value is a mean of five replicates ±SE (standard error).

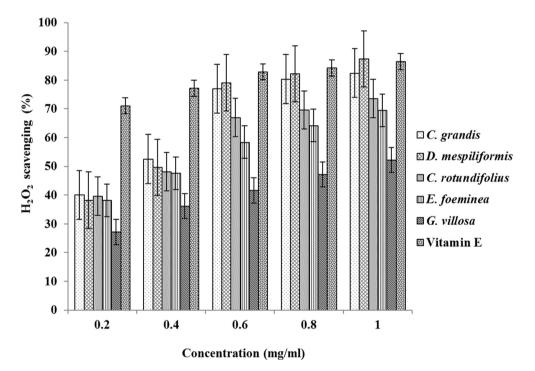


Fig. 3. H₂O₂ scavenging activity (%) of different concentrations of methanol extracts of investigated wild fruits. Each value is a mean of five replicates ±SE (standard error).

methanol extracts of *C. grandis* and *D. mespiliformis* exhibited higher hydrogen peroxide scavenging activity than the other studied species. The hydrogen peroxide scavenging activity of *C. grandis* were 54.18, 63.29, 69.04 and 71.53% at 0.4, 0.6, 0.8 and 1 mg/ml concentration, respectively. Analogous to this result, Meenatchi et al. (2017) stated that *C. grandis* showed hydrogen peroxide scavenging activity (59.64%) at 250 µg/ml concentration.

On the whole, the significant antioxidant activities of the ripe fruits in this study may be due to its high contents of antioxidant components including total phenolic, tannins, carotenoids, anthocyanin, vitamin C and vitamin A as showed in Table 1. These results are in accord to earlier studies attributed the antioxidant activity of different plant extracts to their contents of phenolic and flavonoid compounds and others (Mohamed et al., 2016; Ali et al., 2018).

4. Conclusions

The ripe fruits of *C. grandis, D. mespiliformis, C. rotundifolius, E. foeminea, and G. villosa* have high nutritive value and possess significant amounts of pharmaceuticals and active constituent compounds which exhibit high antioxidant activity. It represent good nutritional and healthy sources for native and rural regions especially *C. grandis* fruits as it contain adequate amounts of fibers (6.84%), total lipids (5.51%), total anthocyanin (0.28 mg/g), total carotenoid (35.19 mg/g), vitamin C (896.41 µg/100 mg), vitamin A (856.74 µg/100 mg), total amino acids (114.61 mg/g), and good amount of macro and micro minerals, beside to its high DPPH and hydrogen peroxide scavenging effect (82.46% and 71.53%, respectively), and the highest total antioxidant activity. Also, *D. mespiliformis* fruits contain pleasing amounts of total protein (9.28%),

total hydrolyzable carbohydrates (15.88%), total soluble sugars (9.82%), total free amino acids (2.95%), total phenolic (20.69 mg/g), and total tannin (3.84 mg/g), beside to it highest antioxidant activity. Future studies are necessary to screen the individual chemical compounds besides more medicinal properties of these wild underutilized fruits for supporting their nutritive and pharmaceutical value. The primary results of this study propose these wild fruits to be used in a variety of food and pharmaceutical applications.

Declarations

Author contribution statement

Ahmad Hegazy: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Amal Mohamed: Conceived and designed the experiments; Analyzed and interpreted the data; Wrote the paper.

Sami Ali: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Nasser Alghamdi Contributed reagents, materials, analysis tools or data. Amal M. Abdel-Rahman: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data.

Sanad Al-Sobeai: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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