

## Fatty Acid, Tocopherol, Sterol Compositions and Antioxidant Activity of Three *Garcinia* Seed Oils

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**Abstract:** The fatty acid, tocopherol, and sterol contents of the extracted seed oils from three *Garcinia* species, including *G. hanburyi* Hook. f., *G. multiflora* Champ. ex Benth., and *G. gaudichaudii* Planch. & Triana, were analyzed. The results showed that the *Garcinia* seeds contained a high amount of oil (16.28–29.91%). The major monounsaturated and saturated fatty acids were oleic acid (82.97 % in *G. multiflora* and 45.37 % in *G. hanburyi*) and stearic acid (58.87 % in *G. gaudichaudii* and 50.12 % in *G. hanburyi*). In addition, three extracted oils exhibited moderate antioxidant activity with *G. hanburyi* displayed the strongest activity with IC<sub>50</sub> value of 6.34± 0.43 µg /mL or EC<sub>50</sub> = 11.71 ± 0.22 µg /mL.

**Keywords:** Seed oil; fatty acid; tocopherol; sterol; *Garcinia*; antioxidant activity. © 2018 ACG Publications. All rights reserved.

### 1. Introduction

*Garcinia* is a large genus that occurs mainly in the tropical countries. The fruits of most *Garcinia* species are eatable for human and animals, and some are highly esteemed in some region. Seed of some species in this genus are sources of edible fat known as "kokam butter" or "kokam fat" for *G. indica* and "tamal" for *G. morella*. Species in the family are mainly wood or shrubs, characterized by yellow latex, branches, usually horizontal. Some species are trees fruits and many species contain active compounds such as xanthenes, benzophenones, flavonoids, and tannins [1]. Although there are lot of works in Vietnam related to the economic values and preliminary chemical research of some other species [2-6], three *Garcinia* species mentioned herein have not been studied for their chemical compositions extensively. *G. hanburyi* was reported to show anti-inflammatory, analgesic, antibacterial, and antipyretic activities [7,8]. *G. multiflora* was also known for its antioxidant and antibacterial, especially anti-HIV activity [9,10]. Comparatively, the third species *G.*

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*gaudichaudii* was lack of bioactivity report. Plant seeds are important oil sources of nutritional and pharmaceutical industries, and therefore detailed knowledge of the chemical composition of seed oil is very important for their application [11,12]. However, the comprehensive constitution of *Garcinia* seed oils is still poorly investigated. The present research is the first report of fatty acid, tocopherol and sterol compositions and antioxidant activity of seed oils from three *Garcinia* species, *G. hanburyi* Hook f., *G. multiflora* Champ. ex Benth., and *G. gaudichaudii* Planch. & Triana. Hopefully, it would provide systematic information regarding the further application of these species.

## 2. Materials and Methods

### 2.1. Plant Materials

Three *Garcinia* seeds including *G. hanburyi* Hook f., *G. multiflora* Champ. ex Benth., and *G. gaudichaudii* Planch. & Triana were collected from Phu Quoc province, Vietnam in 2016. Voucher specimens VNMN-B 2015.121, VNMN-B 2015.123 and VNMN-B 2015.130 were kept at Department of Organic Biochemistry, Institute of Natural Products Chemistry, VAST and identified and set herbarium number by Dr. Nguyen Quoc Binh (Department of Biology, Vietnam National Museum of Nature, VAST), stored at -4 °C for further experiments.

### 2.2 Oil Extraction

The oils were extracted from three *Garcinia* seeds by Soxhlet extraction according to method [13] with some modifications. In brief, 10 g sample of each seed was ground in a ball mill and extracted with 200 mL of petroleum ether in a Twisselmann apparatus for 6 hrs. The solvent was removed with a rotary evaporator at 40 °C and 25 torr. The oil contents obtained by Soxhlet extraction were 17.73 % for *G. hanburyi*, 29.91 % for *G. multiflora*, and 16.28 % for *G. gaudichaudii* respectively. The oils was dried by a gentle stream of nitrogen and stored at -20 °C until use.

### 2.3. Analysis of Fatty Acid, Tocopherol and Sterol Compositions

Fatty acid was determined by gas chromatography followed the ISO draft standard method [14]. In brief, 10 mg of oil was dissolved in 1 mL of petroleum ether. Then, 25 $\mu$ L of 2M sodium methanolate methanol solution was added, and the closed vial was agitated vigorously for 1 min. About 20  $\mu$ L water was added, and after centrifugation the aqueous phase was removed. Then 20  $\mu$ L methyl orange in 0.1 N HCl was added as a pH indicator. The mixture was agitated carefully and different derivatives was analyzed by a Hewlett-Packard Gas Chromatography Instrument Model 5890 Series II/5989 A, equipped with a 0.25 mm ZB-1 fused-silica capillary column (30 m x 0.25  $\mu$ m i.d.; Phenomenex, Torrance, CA). The carrier gas was helium at a flow rate of 1.0 mL/min.

Tocopherol determination was accomplished by HPLC analysis [15]. A solution of 250 mg oil in 25 mL heptane was used for the HPLC analysis conducted on a Merck-Hitachi low-pressure gradient system, fitted with a L-6000 pump, a Merck-Hitachi F-1000 fluorescence spectrophotometer (detector wavelengths at 295 nm for excitation, and at 330 nm for emission) and Chemstation integration software. About 20  $\mu$ L of the sample was injected by a Merck 655- A40 autosampler onto a Diol phase HPLC column (250 mm  $\times$  4.6 mm i.d.; Merck), which was used at a flow rate of 1.3 mL/min. The mobile phase used was heptane/*tert*-butyl methyl ether (99 + 1, v/v). The results are given as mg vitamin E/100g oil.

Sterol was determined according to the official IOOC (the International Olive Oil Council) method [16]. Oil sample (15 g) was saponified by 50 mL of 2 N ethanolic potassium hydroxide solution. The unsaponifiable fraction was dissolved in chloroform, and approximately 20 mg were loaded on a basic silica TLC plate. The sterol and triterpenediol fraction was separated by eluent mixture with hexane and diethyl ether 65:35 (v/v). The corresponding band was visualized under UV light after being sprayed with a 2,7-dichlorofluorescein in 0.2 % ethanolic solution, then scraped off with a spatula, and extracted with chloroform. After the extract was evaporated to dryness, sterols and

triterpenediols were converted into trimethylsilyl ethers by the addition of pyridine hexamethyldisilazane-trimethylchlorosilane (9:3:1, v/v/v). The resulting mixture was kept for 15 min and then centrifuged. The analysis of sterols was performed on a fused silica capillary column coated with 2 % isopropanol/98 % hexane (30 m x 0.32 mm i.d., film thickness 0.25  $\mu\text{m}$ ; Rtx-5: Restek Corporation, Bellefonte, PA, USA; or HP-5: Agilent Technologies Inc., Little Falls, DE, USA) with a Hewlett Packard series 6890 GC (Waldbronn, Germany) equipped with a split/splitless injector, an autosampler, and a flame ionization detector (FID). An aliquot of 1.0 mL of derivatized sample solution was introduced into the column via split injection (300  $^{\circ}\text{C}$ , split ratio 1:50). The components were separated isothermally at 300  $^{\circ}\text{C}$  and detected with the FID (at 310  $^{\circ}\text{C}$ ). The carrier gas was helium. To identify the individual peaks of sterols, the determination of relative retention times (RRT) for sterols was carried out according to the majority compound of sterols ( $\beta$ -sistosterol), knowing that RRT ( $\beta$ -sitosterol) equal to 1 as described by COI [17].

#### 2.4. Determination of Antioxidant Activity with the DPPH Free Radical Scavenging Method

For determination of the antioxidant activity of different extracts, three different antioxidant assay had been utilized.

##### 2.4.1 DPPH Free Radical Scavenging Method [18]

An aliquot (0.5 mL) of the DPPH solution (about 50 mg/100 mL) was diluted in 4.5 mL of methanol, and 0.1 mL of a methanol solution of the extract was added. The mixture was shaken vigorously and allowed to stand for 45 min in the dark. The decrease in absorbance was measured at 515 nm against a blank (without extract) with a spectrophotometer. From a calibration curve obtained with different amounts of extracts, the  $\text{IC}_{50}$  which was required to quench 50 % of the initial DPPH radicals under the experimental conditions given was calculated.

##### 2.4.2 FRAP Ferric Reducing Ability of Plasma [19]

The FRAP reagent contained 2,4,6-tripyridyl-s-triazine (TPTZ) solution (20 mM) in HCl (40 mM),  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  (20 mM) and 0.3 M acetate buffer with pH 3.6. FRAP reagent (1.8 mL) mixed with 0.2 mL of test sample was incubated at 37 $^{\circ}\text{C}$  for 10 min in a water bath. After incubation, the absorbance was measured immediately at 593 nm. The calibration curve was plotted with absorbance vs concentration of  $\text{FeSO}_4$  in the range of 0-1 mM and the total antioxidant activity was expressed as  $\mu\text{mol Fe (II)}/\text{g extract}$ .

##### 2.4.3 TEAC Trolox Equivalent Antioxidant Capacity [20]

The green radical cation  $\text{ABTS}^+$  was generated by oxidation of ABTS [2,2'-azinobis(3-ethylbenzothiazoline-sulfonic-6-acid) diammonium salt] 7 mM with  $\text{K}_2\text{S}_2\text{O}_8$  2.45 mM and allowing the mixture in the dark at room temperature for 16 h. The  $\text{ABTS}^+$  solution was diluted to an absorbance of  $0.7 \pm 0.02$  at 734 nm. After addition of 1900  $\mu\text{l}$  of diluted  $\text{ABTS}^+$  solution to 100 $\mu\text{l}$  of test substances or Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) standards, the absorbance was taken 1–6 min after initial mixing. The percentage inhibition of absorbance of  $\text{ABTS}^+$  at 734 nm was then calculated and plotted towards concentrations of test/standard substances as a function of time or concentration. The TEAC value was finally calculated as the ratio of the slopes of the linear regression of the concentration–response curves of the test substances towards the reference substance (Trolox).

### 3. Results and Discussion

The oil contents obtained by Soxhlet extraction were 17.73 % for *G. hanburyi*, 29.91 % for *G. multiflora*, and 16.28 % for *G. gaudichaudii*, respectively. Comparatively, the oil contents reported for *G. indica* is about 50 %, for *G. xanthochymus* is 16.9 %, and for *G. gummi-gutta* is 11.21 % [21,22]. The oil contents of three seeds studied herein were lower than those of seeds used in commercial oils,

such as rapeseed and sesame oils (about 40 %); and were similar to that of soybean oil (about 20 %). Regarding the economic aspects of oilseed production, a high oil content is important for the utilization of seeds [23,24]. Therefore, the oils from three *Garcinia* species were further investigated for the chemical compositions including the fatty acids, tocopherols and sterols, to elucidate the bioactive potentials of these seeds.

The fatty acid compositions of three seed samples were summarized in Table 1. Despite of three studied species belong to the same genus, the fatty acids composition of them are different significantly. It is determined that the seed oil of all three species contain common fatty acids in different proportions, for example: stearic acid (18:0) represent for saturated fatty acids and in unsaturated fatty acids, there are oleic acid [18:1(n-9)] and linoleic acid [18:2(n-6)]. The literature reported that the higher portions of total saturated fatty acids increase the level of low density cholesterol in the blood serum, which leads to a higher risk for heart diseases and arteriosclerosis [25,26]. Oleic acid is the predominant monounsaturated fatty acids identified in *G. multiflora* (83.46 %) which reveals that the seed oil of *G. multiflora* are a rich source of unsaturated fatty acids. In *G. hanburyi* seed oil, the content of oleic acid is 45.50 %, however, the content of this acid in *G. gaudichaudii* is only 7.82 %. It is reported that the contents of oleic acid in two other species *G. indica* and *G. morella* were 53 % and 50 %, respectively [27]. Various reports have also shown higher oleic acid contents in *G. gummi-gutta* (63.74 %), *G. indica* (67.06 %), *G. xanthochymus* (51.68 %) [27], and *G. mangostana* (45.38 %) [28]. Oleic acid is the most common dietary monounsaturated fatty acid, found in most animal fats, including poultry, beef and lamb, as well as olives, nuts, seeds, and corn. It is reported that oleic acid is effective to lower low-density lipoprotein (LDL) cholesterol level [29,30]. Therefore, the high quantity of oleic acid in the *Garcinia* seed oils may be good for health. The other major fatty acid identified in *G. multiflora* was stearic acid (11.49 %). Comparatively, it was found to be a major fatty acid in seed oils of *G. gaudichaudii* (58.87 %) and *G. hanburyi* (50.12 %). Fats or oils abundant with stearic acid are more common in animal fats (up to 30 %) than in vegetable oils (typically < 5 %). The important exceptions are cocoa butter and shear butter, where the stearic acid contents (as a triglyceride) are 28–45 % [31]. It also indicated the potential of these *Garcinia* seed oils. Linoleic acid was found to be the highest concentration in *G. gaudichaudii* (30.46%) meanwhile in *G. hanburyi* and *G. multiflora* were only 0.77 and 0.37 % respectively. Similar amounts of linoleic acid were also found in the seed oils like corn, soybean, safflower, sunflower [26,32]. In contrast, linolenic acid (C18:3(9, 12, 15)) was not found in genus *Garcinia*.

**Table 1.** Fatty acid composition (%) of three *Garcinia* seed oils

Fatty acid	KI	<i>G. hanburyi</i>	<i>G. multiflora</i>	<i>G. gaudichaudii</i>
C16:0	1984	2.11 ± 0.04	4.31 ± 0.01	1.71 ± 0.02
C18:0	2114	50.12 ± 0.01	11.49 ± 0.01	58.87 ± 0.04
C18:1(n-9)	2141	45.50 ± 0.03	83.46 ± 0.10	7.82 ± 0.10
C18:1(n-7)	2161	0.25 ± 0.02	< LOQ	0.21 ± 0.03
C18:2(n-6)	2173	0.77 ± 0.01	0.37 ± 0.01	30.46 ± 0.20
C18:3(n-3)	2162	0.12 ± 0.00	< LOQ	0.20 ± 0.01
C20:0	2327	0.43 ± 0.00	< LOQ	0.57 ± 0.00
C20:1(n-9)	2356	0.14 ± 0.00	0.36 ± 0.01	< LOQ
C24:0	2400	0.45 ± 0.00	< LOQ	< LOQ
Saturated		53.11 ± 0.01	15.80 ± 0.01	61.15 ± 0.05
Unsaturated		46.78 ± 0.01	84.19 ± 0.03	38.69 ± 0.05

\*KI: Kovats Index.

\*LOQ: Limit of quantitation.

Tocopherols are naturally occurred in most vegetable oils. There are four types of tocopherols, including  $\alpha$ -,  $\beta$ -,  $\gamma$ - and  $\delta$ -tocopherols, with  $\alpha$ -tocopherol being the most abundant in foods and considered to have the highest nutritional value. On the other hand, studies revealed that various tocopherols exhibit diverse health-promoting properties. Tocopherols are natural antioxidants, and the antioxidant and anti-inflammatory activities are more significant with higher  $\gamma$ -tocopherol contents [33,34]. The compositions of tocopherols of three seed oils are listed in Table 2. The seeds with high

contents of tocopherols were regarded as rich sources for tocopherols and tocotrienols for the stabilization of fats and oils against oxidative deterioration and for extensive application in dietary, pharmaceutical or biomedical products. The main ingredients were  $\gamma$ -T- (31.88 mg/kg) and  $\delta$ -T3-tocopherols (110.70 mg/kg) presented in the seeds of *G. hanburyi*;  $\gamma$ -T3- (42.59 mg/kg) and  $\gamma$ -T-tocopherols (35.19 mg/kg) in *G. multiflora*; and  $\alpha$ - (80.86 mg/kg) and  $\delta$ -T3-tocopherols (45.72 mg/kg) in *G. gaudichaudii*, respectively.

**Table 2.** Tocopherol content (mg/kg) of three *Garcinia* seed oils

Compound	Content (mg/kg)		
	<i>G. hanburyi</i>	<i>G. multiflora</i>	<i>G. gaudichaudii</i>
$\alpha$ -T	< LOQ	20.12 $\pm$ 0.05	80.86 $\pm$ 0.03
$\alpha$ -T3	3.82 $\pm$ 0.03	1.31 $\pm$ 0.01	2.12 $\pm$ 0.04
$\beta$ -T	< LOQ	0.43 $\pm$ 0.04	1.54 $\pm$ 0.02
$\gamma$ -T	0.45 $\pm$ 0.01	35.19 $\pm$ 0.04	6.30 $\pm$ 0.02
$\beta$ -T3	< LOQ	< LOQ	1.68 $\pm$ 0.03
P8	< LOQ	7.15 $\pm$ 0.04	< LOQ
$\gamma$ -T3	21.68 $\pm$ 0.02	42.59 $\pm$ 0.01	1.86 $\pm$ 0.05
$\delta$ -T	1.08 $\pm$ 0.04	3.60 $\pm$ 0.03	2.19 $\pm$ 0.03
$\delta$ -T3	110.70 $\pm$ 0.03	< LOQ	45.72 $\pm$ 0.01
<b>Total</b>	137.73 $\pm$ 0.02	110.37 $\pm$ 0.02	142.27 $\pm$ 0.02

\*LOQ: Limit of quantitation.

**Table 3.** Sterol composition (mg/kg) of three *Garcinia* seed oils

Compound	Content (mg/kg)		
	<i>G. hanburyi</i>	<i>G. multiflora</i>	<i>G. gaudichaudii</i>
Cholesterol	8.51 $\pm$ 0.02	9.33 $\pm$ 0.01	13.18 $\pm$ 0.02
Brassicasterol	144.42 $\pm$ 0.03	10.13 $\pm$ 0.05	< LOQ
24-Methylencholesterol	6.78 $\pm$ 0.02	8.29 $\pm$ 0.04	< LOQ
Campesterol	355.55 $\pm$ 0.02	197.41 $\pm$ 0.02	124.96 $\pm$ 0.03
Campestanol	594.45 $\pm$ 0.04	17.46 $\pm$ 0.05	< LOQ
Stigmasterol	1871.53 $\pm$ 0.03	379.90 $\pm$ 0.04	118.15 $\pm$ 0.01
$\Delta^7$ -Camersterol	140.59 $\pm$ 0.02	4.55 $\pm$ 0.01	< LOQ
$\Delta^{5,23}$ -Stigmastadienol	24.25 $\pm$ 0.01	7.18 $\pm$ 0.02	< LOQ
Chlerosterol	89.31 $\pm$ 0.05	1.92 $\pm$ 0.03	< LOQ
$\beta$ -Sitosterol	339.57 $\pm$ 0.03	459.38 $\pm$ 0.05	312.56 $\pm$ 0.01
Sitostanol	21.67 $\pm$ 0.02	27.68 $\pm$ 0.04	< LOQ
$\Delta^5$ -Avenasterol	84.19 $\pm$ 0.01	44.91 $\pm$ 0.03	< LOQ
$\Delta^{5,24}$ -Stigmastadienol	86.76 $\pm$ 0.03	2.82 $\pm$ 0.04	< LOQ
$\Delta^7$ -Stigmastenol	149.27 $\pm$ 0.04	4.23 $\pm$ 0.02	< LOQ
$\Delta^7$ -Avenastenol	81.45 $\pm$ 0.02	1.04 $\pm$ 0.03	< LOQ
<b>Total</b>	3998.33 $\pm$ 0.02	1176.25 $\pm$ 0.03	568.86 $\pm$ 0.02

\* Cholestanol is used as Internal standard (ISTD)

Sterols are important constituents of oils corresponding to the oil quality and are widely used to check the authenticity of oils [35]. Phytosterols are usually of interest due to their antioxidant activity and impact on health. This fraction has been considered as the major unsaponifiable fraction in many

oils. The sterol composition in the *Garcinia* seed oils are presented in Table 3. From the data, three seeds contain the typical phytosterol compounds identified as campesterol, stigmasterol and sitosterol. Campestanol only occurred in the seeds of *G. hanburyi* (594.45 mg/kg). The most abundant sterol in the seeds of *G. hanburyi* was stigmasterol (1871.50 mg/kg) followed by campesterol (355.55 mg/kg) and  $\beta$ -sitosterol (339.57 mg/kg). Comparatively, the predominant sterols were  $\beta$ -sitosterol (459.38 mg/kg) and campesterol (197.41 mg/kg) in the seeds of *G. multiflora*; and  $\beta$ -sitosterol (312.56 mg/kg), campesterol (124.96 mg/kg), and stigmasterol (118.15 mg/kg) in the seeds of *G. gaudichaudii*. sitosterol is reported to have anti-hypercholesterolemic, anti-inflammatory, antibacterial, antifungal, and anti-hyperlipoproteinemic activities, as well as inhibiting carcinogenesis [36], and decreasing glycated hemoglobin and serum glucose levels, while increasing serum insulin levels [37]. Stigmasterol is used as a precursor in the manufacturing of semi-synthetic progesterone, a valuable human hormone that plays an important physiological role in the regulatory and tissue rebuilding mechanisms related to estrogen effects, as well as acting as an intermediate in the biosynthesis of androgens, estrogens, and corticoids. It is also used as the precursor of vitamin D<sub>3</sub>. The Upjohn company used stigmasterol as the starting raw material for the synthesis of cortisone [38]. Briefly, the high sterol contents of these three *Garcinia* seed oils may extend their application in the future.

**Table 4.** DPPH radical scavenging percentage (%) of three *Garcinia* seed oils

Concentration ( $\mu\text{g/mL}$ )	<i>G. hanburyi</i>	<i>G. multiflora</i>	<i>G. gaudichaudii</i>	ascorbic acid (Control)
100	85.20	81.37	84.54	86.11
50	83.62	79.07	71.36	78.84
25	81.55	71.61	46.92	39.15
12.5	78.49	38.41	31.78	19.31
6.25	44.16	-	-	9.42
IC <sub>50</sub> ( $\mu\text{g/mL}$ )	6.34 $\pm$ 0.43	14.27 $\pm$ 0.30	26.23 $\pm$ 1.64	6.63 $\pm$ 0.57

**Table 5.** The ferric reducing ability of plasma FRAP ( $\mu\text{M/g}$ ) of three *Garcinia* seed oils

Concentration ( $\mu\text{g/mL}$ )	<i>G. gaudichaudii</i>	<i>G. hanburyi</i>	<i>G. multiflora</i>	ascorbic acid (Control)
2000	1475.37	2803.97	2701.40	-
1000	873.79	2563.37	1640.17	-
500	468.31	1727.18	931.26	3053.17
250	-	-	-	2391.51
125	-	-	-	1248.66

**Table 6.** The Trolox equivalent antioxidant capacity TEAC (%) of three *Garcinia* seed oils

Concentration ( $\mu\text{g/mL}$ )	<i>G. gaudichaudii</i>	<i>G. hanburyi</i>	<i>G. multiflora</i>	Trolox (Control)
100	85.40	88.13	88.29	-
50	54.90	84.85	82.35	91.73
25	35.65	79.14	51.34	90.02
12.5	21.93	57.58	22.99	89.44
6.25	7.31	24.60	10.70	47.77
3.125	-	-	-	17.48
EC <sub>50</sub> ( $\mu\text{g/mL}$ )	41.44 $\pm$ 1.57	11.71 $\pm$ 0.22	24.70 $\pm$ 2.26	6.68 $\pm$ 0.83

DPPH radical scavenging activity and two other methods FRAP and TEAC were used to evaluate the antioxidant potentials of three extracted oils and the results demonstrated that all samples show moderate antioxidant potentials (Table 4, 5 and 6). At high concentration, all three-tested species do not show significant difference in the scavenging percentages with approximately 85 % at 100  $\mu\text{g/mL}$  and 80 % at 50  $\mu\text{g/mL}$ , except *G. gaudichaudii* (71.36 %) according to DPPH assay and the results were the same for TEAC method with 87 % at 100  $\mu\text{g/mL}$  and 83 % at 50  $\mu\text{g/mL}$  and only *G. gaudichaudii* showed lower antioxidant capacity (54.90 %). At the tested concentration of 25  $\mu\text{g/mL}$ ,

two species *G. hanburyi* and *G. multiflora* remained the high percentages of scavenging, while *G. gaudichaudii* and ascorbic acid decreased rapidly, however, in FRAP and TEAC assays at lower concentration showed that *G. gaudichaudii* and *G. multiflora* antioxidant activity weaken rapidly in compared to *G. hanburyi* and the control. At the concentration of 12.5  $\mu\text{g/mL}$  (DPPH and TEAC) or 500  $\mu\text{g/mL}$  (FRAP) only *G. hanburyi* still exhibited high scavenging percentage (78.49 % and 57.58 %) and 1727.18  $\mu\text{M/g}$  respectively. In summary, in all three tested methods, the species *G. hanburyi* showed the strongest antioxidant activity ( $\text{IC}_{50} = 6.43 \mu\text{g/mL}$ ,  $\text{EC}_{50} = 11.71 \pm 0.22 \mu\text{g/mL}$ ) which is quite similar to the value of ascorbic acid ( $\text{IC}_{50} = 6.63 \mu\text{g/mL}$ ) and Trolox ( $\text{EC}_{50} = 6.68 \pm 0.83 \mu\text{g/mL}$ ). The two other species *G. multiflora* and *G. gaudichaudii* are slightly less active. The antioxidant activity could be due to the presence of  $\alpha$ -tocopherol (20.12 and 80.86 mg/kg) in *G. multiflora* and *G. gaudichaudii* and also other phenolic compounds and unsaturated fatty acids.

#### 4. Conclusion

In this work, the fatty acid, tocopherol, and sterol compositions from three *Garcinia* seeds were studied for the first time. The *Garcinia* seeds contained a high amount of oil (11.71–29.91%). The major monounsaturated and saturated fatty acids were oleic acid (82.97 % in *G. multiflora*, 45.37 % in *G. hanburyi*) and stearic acid (58.87 % in *G. gaudichaudii* and 50.12 % in *G. hanburyi*). Although the three studied seeds were belong to the same genus, the contents of fatty acids were significantly different among the samples, especially in *G. multiflora*. The seed oil of this species contained mainly oleic acid up to more than 80%, but 45.50% in *G. hanburyi* and 7.82% in *G. gaudichaudii* comparatively. Linoleic acid was presented in the seed oil of *G. gaudichaudii* (30.46%), however, its concentration in the two other species was lower than 1%. Both two species *G. gaudichaudii* and *G. hanburyi* contained stearic acid C18:0 at high concentrations (more than 50%). These two species were only different in the contents of linoleic acid (30.46% in *G. gaudichaudii* and 0.77% in *G. hanburyi*, respectively). Steroids were predominant in these oils, including several typical sterols such as campesterol, stigmasterol and sitosterol. The most abundant sterol in the seeds of *G. hanburyi* was stigmasterol (1871.53 mg/kg), whereas it was  $\beta$ -sitosterol in the seeds of *G. multiflora* (459.38 mg/kg) and *G. gaudichaudii* (312.568 mg/kg). The seed oils from three *Garcinia* species showed moderate antioxidant activity evaluated using three antioxidant assays DPPH, FRAP and TEAC and *G. hanburyi* displayed the strongest activity with  $\text{IC}_{50}$  value of  $6.34 \pm 0.43 \mu\text{g/mL}$  or  $\text{EC}_{50} = 11.71 \pm 0.22 \mu\text{g/mL}$ .

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