VIETNAM JOURNAL OF CHEMISTRY 54(2) 256-261 APRIL 2016
DOI: 10.15625/0866-7144.2016-00272

NATURAL COMPOUNDS FROM SEEDS OF *CASSIA GRANDIS* L.f

Ngo Quoc Luan[1*], Nguyen Cong Tao[1], Vu Thi Hoai Tien[1], Nguyen Ba Nghia[1], Le Tien Dung[2], Mai Dinh Tri[2], Nguyen Ngoc Hanh[2], Nguyen Kim Phi Phung[3]

[1]*School of Education, Cantho University*

[2]*Institute of Chemical Technology, Vietnam Academy of Science and Technology*

[3]*University of Natural Science of Hochiminh City*

Received 22 July 2015; Accepted for publication 25 April 2016

Abstract

For the first time, four known natural compounds from extracts of seeds of *Cassia grandis* L.f including *o*-anisic acid **(1)**, chrysophanol (**2**), pulmatin (**3**) and (-)-epiafzelechin (**4**) have been isolated. Their structures were elucidated by MS, NMR spectra and comparison with published data. Three in isolated compounds were tested for anticancer, antioxidant and antimicrobial activities, only pulmatin could inhibit to *E. coli, B. subtillis and S. aureus*.

Keywords. *Cassia grandis* L.f, *o*-anisic acid, chrysophanol, pulmatin, (-)-epiafzelechin.

1. INTRODUCTION

Cassia grandis L.f was introduced in the previous reported [1]. Chemical investigation on this plant has been going from 2011 to present, and has successfully detected more than 30 natural compounds. In this paper we continue to report 4 compounds as the first time detected in the seeds of *Cassia grandis* L.f.

2. EXPERIMENTAL

2.1. Plant material

Ripe fruit of *Cassia grandis* L.f were collected in Can Tho City on September, 2014. A voucher specimen was identified by MSc. Dang Minh Quan - Cantho University. After separating and cleaning, the poor quality seeds were removed, good material was dried at 50 °C to decrease humidity to 0-2%, followed by crushing into fine powder.

2.2. General experimental procedures

2.2.1. Extraction and purification

Solid-liquid, liquid-liquid extraction were used with solvents as ethanol 96%, *n*-hexane, EtOAc, BuOH, MeOH. Solvent evaporating was applied by Buchi R-210 rotary evaporator system.

Thin layer chromatography (TLC) was carried out on pre-coasted silica gel $60F_{254}$ (0.2 mm) aluminum sheet (Merck) and compounds were detected under UV (254/365 nm) fluorescence or spraying 10 % H_2SO_4 solution in EtOH, followed by heating at 105 °C for 1-2 min on electric stove.

For common phase column chromatography (CP-CC), silica gel 60 (0.040-0.063 mm, Merck) with increasing polarity solvent systems including *n*-hexane (H), EtOAc (E), $CHCl_3$ (C), MeOH (M) and H_2O (W) were used. Sections have similar traces in TLC were collected into a fraction.

Compound purification was applied by re-crystallization.

2.2.2. Structural elucidation and identification

Melting point (mp.) was recorded on a melting point meter (Electrothermal 9100-UK), using with capillary, uncorrected. ^{1}H-NMR, ^{13}C-NMR, DEPT, HSQC, HMBC spectra were recorded on a Bruker AM500 FT-NMR spectrometer. Mass spectrum (MS) was recorded on mass spectrometer (HP 1100 series, LC/MSD Trap, Agilent). These equipment are available at Vietnam Academy of Science and Technology.

2.3. Extraction and isolation

The dried powdered seeds (4.0 kg) was exhausted extracted with ethanol 96% to gain

ethanol extracts (283 g). The ethanol extracts was consecutively distributed into *n*-hexane, ethyl acetate, *n*-butanol, acetone and methanol.

The ethyl acetate extracts (SE, 70 g) was subjected to CP-CC with H:E solvent systems (10:0 to 0:10) as eluent to give 6 fractions (SE1-SE6).

The fraction SE4 (3.01 g) was continued CP-CC with C:M (10:0 to 7:3) to give 5 fractions (SE41-SE45). The fraction SE42 (489 mg) was recrystalized two times to result in compound **1** (301 mg).

The fraction SE5 (8.12 g) was continued CP-CC with C:M (10:0 to 5:5) to afford 7 fractions (SE51-SE57). The fraction SE52 (692 mg) was recrystalized to gain compound **2** (402 mg).

The acetone extracts (SA, 27 g) was subjected to CP-CC with E:M solvent systems (10:0 to 0:10) as eluent to give 7 fractions (SA1-SA7).

The fraction SE2 (2.67 g) was continued CP-CC with C:M (10:0 to 5:5) to get 6 fractions (SA21-SA26). The fraction SA22 (521 mg) was recrystalized three times to result in compound **3** (302 mg).

The fraction SE3 (2.83 g) was continued CP-CC with C:M (95:5 to 5:5) to get 6 fractions (SA31-SA36). The fraction SA33 (682 mg) was recrystalized many times to obtain compound **4** (218 mg).

2.4 Arranged 1D-NMR spectral data

***o*-Anisic acid (1)**: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ_H ppm, *J* Hz): 8.16 (1H, *dd*, *J* = 8.0, 1.5, H-6); 7.57 (1H, *qd*, *J* = 7.0, 1.0, H-4); 7.13 (1H, *p*, *J* = 7.0, 1.0, H-5); 7.07 (1H, *d*, *J* = 8.5 H-3); 4.08 (3H, *s*, H-8. ^{13}C-NMR ($CDCl_3$, 125 MHz, δ_C ppm): 165.7 (C-7); 158.1 (C-2); 135.1 (C-4); 133.6 (C-6); 122.0 (C-5); 117.5 (C-1); 111.6 (C-3); 56.6 (C-8).

Chrysophanol (**2**): ^{1}H-NMR (DMSO-d_6, 500 MHz, δ_H ppm, *J* Hz): 11.88 (2H, *s*, 1-OH and 8-OH); 7.78 (1H, *t*, *J* = 8.0, H-6); 7.67 (1H, *d*, *J* = 8.0, H-5); 7.50 (1H, *s*, H-2); 7.35 (1H, *d*, *J* = 8.0, H-7); 7.18 (1H, *s*, H-4); 2.42 (3H, *s*, H-3a). ^{13}C-NMR (DMSO-d_6, 125 MHz, δ_C ppm): 191.6 (C-9); 181.4 (C-10); 161.5 (C-8); 161.3 (C-1); 149.1 (C-3); 137.3 (C-6); 133.2 (C-10a); 132.9 (C-4a); 124.3 (C-7); 124.0 (C-2); 120.5 (C-4); 119.3 (C-5); 115.8 (C-8a); 113.7 (C-9a); 21.6 (C-3a).

Pulmatin (**3**): ^{1}H-NMR (DMSO-d_6, 500 MHz, δ_H ppm, *J* Hz): 12.94 (1H, *s*, 8-OH); 7.74 (1H, *t*, *J* = 8.0, H-6); 7.71 (1H, *s*, H-2); 7.66 (1H, *dd*, *J* = 8.0, 1.0, H-5); 7.54 (1H, *s*, H-4); 7.35 (1H, *dd*, *J* = 8.0, 1.0, H-7); 2.47 (3H, *s*, H-3a); Sugar moiety: 5.16 (1H, *d*, *J* = 7.5, H-1'); 5.12 (1H, *s*, 6'-OH); 5.09 (1H, *s*, 2'-OH); 5.08 (1H, *s*, 3'-OH); 4.63 (1H, *s*, 4'-OH); 3.71-3.74 (1H, *m*, H-6'a); 3.47-3.51 (1H, *m*, H-6'b); 3.44-3.46 (1H, *m*, H-5'); 3.41-3.43 (1H, *m*, H-2'); 3.30-3.32 (1H, *m*, H-3'); 3.22-3.25 (1H, *m*, H-4'). ^{13}C-NMR (DMSO-d_6, 125 MHz, δ_C ppm): 187.7 (C-9); 182.1 (C-10); 161.4 (C-8); 158.4 (C-1); 147.4 (C-3); 136.2 (C-6); 134.5 (C-4a); 132.5 (C-10a); 124.3 (C-7); 122.8 (C-2); 121.3 (C-4); 118.34 (C-9a); 118.31 (C-5); 116.8 (C-8a); 21.8 (C-3a); Sugar moiety: 100.5 (C-1'); 77.3 (C-5'); 76.6 (C-3'); 73.4 (C-2'); 69.6 (C-4'); 60.7 (C-6').

(-)-Epiafzelechin (**4**): ^{1}H-NMR (DMSO-d_6, 500 MHz, δ_H ppm, *J* Hz): 9.28 (1H, *s*, 5-OH); 9.10 (1H, *s*, 7-OH); 8.89 (1H, *s*, 4'-OH); 7.22 (2H, *d*, *J* = 8.5, H-2' and H-6'); 6.71 (2H, *dd*, *J* = 6.5, 2.0, H-3' and H-5'); 5.89 (1H, *d*, *J* = 2.0, H-8); 5.72 (1H, *d*, *J* = 2.0, H-6); 4.80 (1H, *s*, H-2); 4.67 (1H, *d*, *J* = 4.5, 3-OH); 4.02 (1H, *t*, *J* = 4.0, H-3); 2.68 (1H, *dd*, *J* = 4.5, 16.0, H-4a); 2.48 (1H, *d*, *J* = 3.0, H-4b). ^{13}C-NMR (DMSO-d_6, 125 MHz, δ_C ppm): 156.5 (C-5 and C-4'); 156.2 (C-7); 155.7 (C-9); 129.9 (C-1'); 128.2 (C-2' and C-6'); 114.4 (C-3' and C-5'); 98.4 (C-10); 95.1 (C-6); 94.1 (C-8); 78.0 (C-2); 64.8 (C-3); 28.2 (C-4).

3. RESULTS AND DISCUSSION

3.1 *o*-Anisic acid (1)

Compound **1** was a white amorphous powder, mp. 102-103 ^{o}C. UV spectrum of **1** gave two maximum absorbance at 235 and 322 nm, it proved that the structure of **1** existed conjugated double bonds.

The ^{1}H-NMR spectrum of **1** revealed 4 signals of 4 aromatic methine *ortho* coupling protons at δ_H 8.16 (1H, *dd*, *J* = 8.0, 1.5); 7.07 (1H, *d*, *J* = 8.5); 7.57 (1H, *dt*, *J* = 7.0, 1.0); 7.13 (1H, *dt*, *J* = 7.0, 1.0) and one signal of a methoxyl group at δ_H 4.08 (3H, *s*). The ^{1}H-NMR spectroscopic data showed that **1** has two substituents locating at *ortho* position each other, because if they were at *para* position, the benzene ring would have symmetry agent, and if they were at *meta* position, then at least one proton would show *meta* coupling only (*J* < 4).

The ^{13}C-NMR and DEPT spectra of **1** showed total 8 carbon signals, including 4 methine (δ_C 135.1, 133.6, 122.0, 111.6 ppm) and 2 quaternary carbon (δ_C 158.1, 117.5 ppm) of an aromatic two substitued ring; one carboxylic carbon at δ_C 165.7 ppm (C-7) and only one methoxyl carbon at δ_C 56.6 ppm (C-8).

The molecular formula of **1** was determined to be $C_8H_8O_3$ (calc. for 152.05) on the basis of the ESI-

MS (*m/z* 134.96 $[M\text{-}OH]^+$, 151.97 $[M]^+$) and 1D-NMR data. This spectra data was the same with those of *o*-anisic acid reported in the literature [2]. The stucture of **1** (figure 1) was also confirmed by HSQC and HMBC spectra of it.

The HSQC spectrum of **1** showed protons δ_H 8.16, 7.57, 7.13, 7.07 and 4.08 ppm sequence correlated with carbons δ_C 133.6, 135.1, 122.0, 111.6 and 56.6 ppm.

Firstly, the HMBC spectrum of **1** (Figure 1a) allowed to determine the carbon δ_C 158.1 ppm was C-2 by protons δ_H 4.08 ppm correlated with it, thus the remain quaternary aromatic carbon δ_C 117.5 ppm was C-1. Then proton δ_H 8.16 ppm and proton δ_H 7.13 ppm correlated with C-7, proton δ_H 8.16 ppm also correlated with C-2 but proton δ_H 7.13 ppm was not, so proton δ_H 8.16 ppm was H-6 and proton δ_H 7.13 ppm was H-5. Similarly, proton δ_H 7.07 ppm correlated with C-1 and was determined to be H-3, while proton δ_H 7.57 ppm did not and was determined to be H-4.

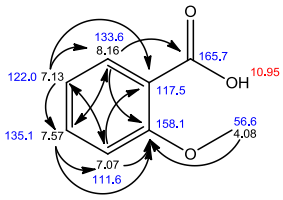

Figure 1a: Selected HMBC correlations of compound **1**

On detailed examination of NMR spectra, the structure of compound **1** was determined as Figure 1b and identified to be *o*-anisic acid.

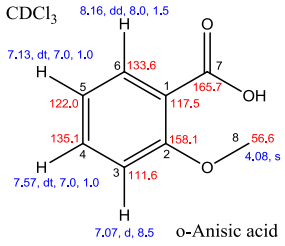

Figure 1b: Chemical structure and arranged 1D-NMR spectra data of o-anisic acid

3.2. Chrysophanol (2)

Compound **2** isolated as yellow amorphous powder, well dissolved in $CHCl_3$, mp. 193-194 ^oC. Compound **2** produced a positive reaction to $FeCl_3$ reagent, so **2** can be a phenolic compound.

The ^1H NMR spectrum of **2** exhibited signals of five asymmetrical benzene ring protons (δ_H 7.78-7.18 ppm), two protons of hydrogen bonded hydroxyl groups at δ_H 11.88 ppm and one methyl group at δ_H 2.42 ppm.

The ^{13}C-NMR and DEPT spectra of **2** showed signals of total 15 carbons, consisted of 2 carbonyl groups at δ_C 191.6 and 181.4 ppm, 2 quaternary oxygenated carbons at δ_C 161.5 and 161.3 ppm, 5 methine carbons (δ_C 119-138 ppm), 5 aromatic quaternary carbons (δ_C 113-150 ppm) and only one methyl carbon at δ_C 21.6 ppm. Excepting methyl group, 14 remain carbons played a typical pattern of anthraquinone skeleton, especially the typical signal of two carbonyl carbons at δ_C 191.6 and 181.4 ppm.

The molecular formula of **2** was suggested to be $C_{15}H_{10}O_4$ (calc. for 254.06) on the basis of the ESI-MS (*m/z* 253.0 $[M\text{-}H]^-$, 166.9 $[C_9H_{11}O_3]^+$).

The HSQC spectrum of **2** showed protons δ_H 7.78, 7.67, 7.50, 7.35, 7.18 and 2.42 ppm in turn correlated with carbons δ_C 137.3, 119.3, 120.5, 124.3, 124.0 and 21.6 ppm.

Because of two hydrogen bonded hydroxyl groups (1-OH and 8-OH), carbonyl carbon δ_C 191.6 was assigned to C-9 and thus the other carbonyl carbon δ_C 181.4 was C-10. Think the only methyl substituent linked at A-ring that means the A-ring contained 2 protons and the B-ring with 3 protons.

On HMBC spectrum of **2** (figure 2a), proton methyl group δ_H 2.42 ppm correlated with carbons δ_C 149.1, 124.0 and 120.5, that proved these carbons were depended on A-ring. Moreover, the signals of protons δ_H 7.50 and 7.18 ppm were all singlet (it can infer that these proton located at *meta* position each other), this is an evidence of methyl group contacted with C-3 (149.1 ppm). Proton δ_H 7.50 ppm was assigned to H-4 by it also correlated with C-10, therefore, proton δ_H 7.18 ppm was H-2. The aromatic quaternary carbon δ_C 113.7 ppm which was assigned to C-9a because two protons δ_H 7.50 and 7.18 ppm also correlated with it.

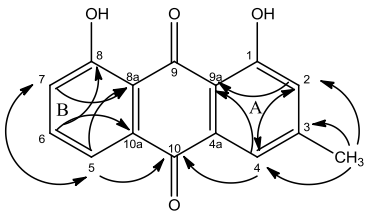

Figure 2a: Selected HMBC correlations of compound **2**

In the B-ring, proton δ_H 7.67 ppm correlating with C-10 was assigned to H-5, the *triplet* δ_H 7.78 ppm was H-6 and the last *doublet* δ_H 7.35 was H-7. That was confirmed by H-5 correlated with C-7 and H-7 correlated with C-5. The aromatic quaternary

carbon δ_C 115.8 ppm was assigned to C-8a because H-5 and H-7 correlated with it. H-6 correlated with the quaternary oxygenated carbons δ_C 161.5 which determined to be C-8, therefore, the other quaternary oxygenated carbons δ_C 161.3 was C-1. H-6 also correlated with the aromatic quaternary carbon δ_C 133.2, so which was C-10a, of course, the last one 132.9 was C-4a.

Moreover, the spectral data of **2** were similar to those of chrysophanol previously reported in the literature [3]. To sum up, the structure of **2** was suggested as Figure 2b and identified to be chrysophanol.

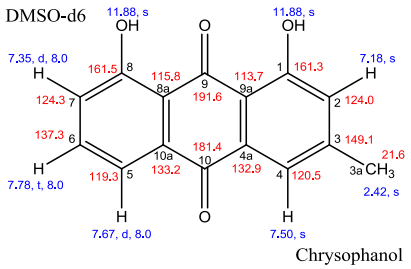

Figure 2b: Chemical structure and arranged 1D-NMR spectra data of chrysophanol

3.3. Pulmatin (3)

Compound **3** were isolated as orange amorphous powder, mp. 252-254 °C, which produced a positive reaction to $FeCl_3$ reagent. UV spectrum of **3** gave three maximum absorbance at 222, 280 and 407 nm.

The ^{1}H-NMR spectrum of **3** showed typical signals of 9 protons of a chrysophanol aglycon like **2** and proton of a sugar moiety. With 5 methine aromatic protons, 3 protons of a methyl group and only one hydrogen bonded hydroxyl, it proved that the sugar moiety linked at another hydrogen bonded hydroxyl of chrysophanol compound.

The ^{13}C-NMR and DEPT spectra of **3** exhibited typical signals of total 15 carbons like a chrysophanol aglycon and 6 carbons of a sugar moiety. The presence of hydroxymethyl carbon δ_C 60.7 ppm, other hydroxymethine carbons δ_C 100.5, 77.3, 76.6, 73.4, 69.6 ppm and anomeric proton δ_H 5.16 ppm (1H, *d*, *J* = 7.5) indicated the sugar moiety was β-glucose.

The molecular formula of **3** was determined to be $C_{21}H_{20}O_9$ (calc. for 416.11) on the basis of the ESI-MS (*m/z* 439.02 $[M+Na]^+$) and 1D-NMR spectral data.

All the protons and carbons were assigned to their positions by using the correlations on HSQC and HMBC spectra of **3** as processing for compound **2**. The sugar moiety attached to C-1 of chrysophanol aglycon because anomeric proton H-1' correlated with it. Besides, the spectral data of **3** were very suitable to those of pulmatin given in the literature [4]. Briefly, compound **3** was identified to be pulmatin (figure 3).

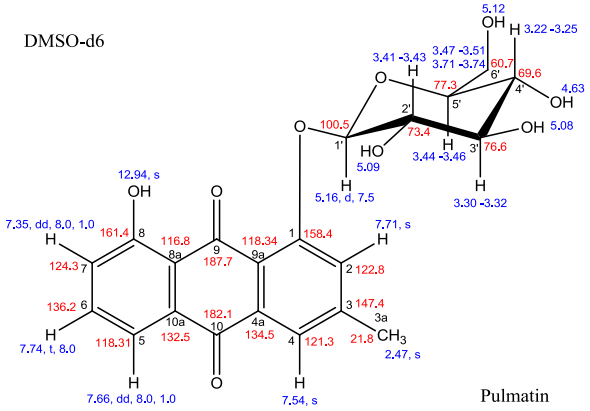

Figure 3: Chemical structure and arranged 1D-NMR spectra data of pulmatin

3.4. (-)-Epiafzelechin (4)

Compound **4** is a white amorphous powder, mp. 239 °C, $[\alpha]_D^{25}$ = -74.2° (c 0.5, MeOH), which produced a positive reaction to $FeCl_3$ reagent.

The ^{1}H NMR spectrum of **4** appeared six signals of aromatic protons in which there were two couples of equivalent protons δ_H 7.22 and 6.71 ppm; two oxygenated methine protons δ_H 4.80 and 4.02 ppm, two methylene protons δ_H 2.68 and 2.46 ppm; 4 protons of hydroxyl groups δ_H 9.28, 9.10, 8.89, 4.67 ppm.

The ^{13}C-NMR and DEPT spectra of **4** showed signals of total 15 carbons including six benzene ring methine carbons, in which there were two overlaped signals of two couples of equivalent carbons δ_C 128.2 and 114.4 ppm, they were suitable to two couples of equivalent protons; two oxygenated methine carbons δ_C 78.0 and 64.8 ppm; four oxygenated quaternary carbons δ_C 156.58, 156.55, 156.2 and 155.7 ppm; two aromatic quaternary carbons δ_C 129.9 and 98.4 ppm; one methylene carbon δ_C 28.2 ppm. The typical signals of protons and carbons in 1D-NMR showed that **4** gave the characteristic of a flavanol with a symmetric benzene ring.

The molecular formula of **4** was determined to be $C_{15}H_{14}O_5$ (calc. for 274.27) on the basis of the HRMS (*m/z* 275.09192 $[M+H]^+$) and ^{13}C-NMR data.

Compound **4** was a flavanol, it was futher confirmed by correlations in HSQC and HMBC spectra of **4** (figure 4a).

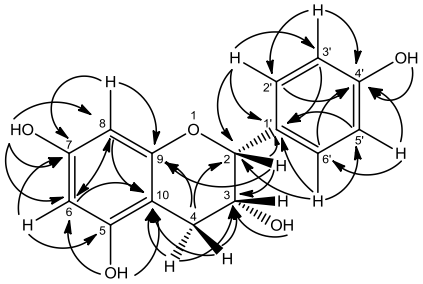

Figure 4a: Selected HMBC correlations of compound **4**

In structure of flavan back-bone, it was easy to identify the only methylene carbon δ_C 28.2 was C-4 and two non-aromatic oxygenated methine carbons δ_C 78.0, 64.8 ppm. Carbons δ_C 78.0 ppm was assigned to C-2 by the main reason that the couple of equivalent protons δ_H 7.22 ppm correlated with it, then carbon δ_C 64.8 ppm was C-3. In symetrical aromatic ring, the couple of equivalent protons δ_H 7.22 ppm which correlated with C-2 was assigned to H-2' and H-6', and the other couple δ_H 6.71 ppm was H-3' and H-5'. The quaternary carbon 129.9 ppm was C-1' because H-3' and H-5' correlated with it. Similarly, the oxygenated quaternary carbon δ_C 156.55 ppm was C-4' because H-2', H-6', H-3' and H-5' correlated with it. In non-symetrical aromatic ring, the non-oxygenated quaternary carbon δ_C 98.4 ppm was C-10, the oxygenated quaternary carbon δ_C 155.7 ppm was C-9 because H-2 correlated with it. Two oxygenated quaternary carbons δ_C 156.58 and 156.2 ppm located at *meta* position each other as a result of *meta* coupling of two aromatic methine protons δ_H 5.89 and 5.72 ppm (J = 2.0 Hz). Carbons δ_C 156.58 ppm was C-5 because of correlations between hydroxyl proton δ_H 9.28 with this carbon, C-10 and C-6 (δ_C 95.1 ppm). Therefore, remain oxygenated quaternary carbons δ_C 156.2 ppm was C-7 and the last aromatic methine carbon δ_C 94.1 ppm was C-8.

H-2 (δ_H 4.80 ppm) appeared as *singlet* by the coupling constant of H-2 and H-3 was negligible, this confirmed that H-2 and H-3 are in a *cis* relationship. In the other hand, $[\alpha]_D^{25}$ = -74.2° < 0, two these points indicated the stereo configuration of **4** was (-)-epi.

Besides, the spectral data of **4** were identical to (-)-epiafzelechin as previously described in the literature [5] (figure 4b).

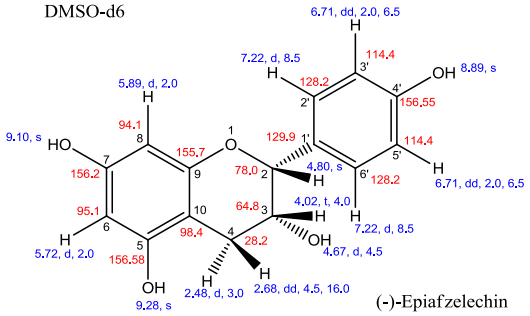

Figure 4b: Chemical structure and arranged 1D-NMR spectra data of (-)-epiafzelechin

3.5. Bioactivity of isolated compounds

o-Anisic acid (CGSE01), chrysophanol (CGSE02) and pulmatin (CGSA01) were tested anticancer, antioxidant and antimicrobial activities. The results showed almost of the samples were negative, except pulmatin could against to *E. coli*, *B. subtillis and S. aureus* (tables 1-3).

Aloe-emodin, a typical anthraquinone, was previously tested for anticancer activity, the results showed that this compound has good activity against cell strains of liver cancer Hep-G2. Above results show that the expectation for this activity to anthraquinone derivatives is not always being met.

Table 1: Anticancer activity of o-anisic acid, chrysophanol and pulmatin

No.	Sample code	Concentration (µg/ml)	Cancer cell survival (%)		Conclusion
			Hep G2	Lu	
	DMSO		100.0±0.0	100.0±0.0	
	Positive evidence (+)	5	**3.12±1.7**	**1.93±0.5**	Positive
1	**CGSA01**	10	62.76±1.3	89.79±1.5	Negative
2	**CGSE01**	10	97.85±2.8	95.88±0.7	Negative
3	**CGSE02**	10	90.29±2.3	98.63±0.8	Negative

Table 2: Antioxidant activity of o-anisic acid, chrysophanol and pulmatin

TT	Sample code	Concentration (μg/ml)	Scavenging capacity (SC, %)	SC_{50} (μg/ml)	Result
	Positive evidence **(+)**	**44**	**80.87±0.13**	**20.7**	**Positive**
	Negative evidence **(-)**	-	0.0±0.0	-	Negative
1	**CGSA01**	200	8.37±0.8	-	Negative
2	**CGSE01**	200	7.86±0.9	-	Negative
3	**CGSE02**	200	0.14±0.8	-	Negative

(-): DPPH/EtOH + DMSO. (+): Ascobic acid.

Table 3: Antimicrobial activity of o-anisic acid, chrysophanol and pulmatin

No.	Sample code	Concentration (μg/ml)	Minimum inhibitory concentration (MIC: μg/ml)								Result
			Bacteria Gr(-)		Bacteria Gr(+)		Fungus		Yeast		
			E. coli	*P. aeruginosa*	*B. subtillis*	*S. aureus*	*A. niger*	*F. oxysporum*	*S. cerevisiae*	*C. albicans*	
1	**CGSE01**	**50**	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	Negative
2	**CGSE02**	**50**	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	Negative
3	**CGSA01**	**50**	**50**	(-)	**50**	**50**	(-)	(-)	(-)	(-)	Antimicrobial

Note: These tests were performed at Experimental Biology Department, Institute of Natural Compounds Chemistry, Vietnam Academy of Science and Technology.

4. CONCLUSION

From ethyl acetate and acetone extracts of the seeds of *Cassia grandis* L.f collected in Can Tho City, four known natural compounds were isolated and identified as *o*-anisic acid **(1)**, chrysophanol (**2**), pulmatin (**3**) and (-)-epiafzelechin (**4**). However, compounds **1**, **2** and **3** isolated herein are reported for the first time from this plant. Compounds **1**, **2** and **3** were negative for anticancer and antioxidant activity tests. Only pulmatin could inhibit to *E. coli*, *B. subtillis and S. aureus* in antimicrobial test.

Acknowledgment. *The authors acknowledge financial support from Cantho University.*

REFERENCES

1. N.Q. Luan et al. *Flavonoid compounds from leaves of Cassia grandis* L.. Journal of Science and Technology, Vietnamese Academy of Science and Technology, **50(3A)**, 296-301 (2012).
2. Liqin Xue, Lijun Shi, Yuan Han, Chungu Xia, Han Vinh Huynh and Fuwei Li. *Pd-carbene catalyzed carbonylation reactions of aryl iodides (Electronic Supplementary Material)*. Dalton Transactions, **40(29)**, 7632-7638(S20) (2011).
3. Shuying Guo, Bo Feng, Ruonan Zhu, Jiankang Ma, and Wei Wang. *Preparative Isolation of Three Anthraquinones from Rumex japonicus by High-Speed Counter-Current Chromatography*. Molecules, **16**, 1201-1210 (2011).
4. T. Annamalai, G. Venkateswara Rao and T. Mukhopadhyay. *Pulmatin from the roots of Rumex acetosa*, Der Pharmacia Lettre, **5(5)**, 116-119 (2013).
5. Kafui Kpegba, Amegnona Agbonon, Ana G. Petrovic, Etchri Amouzou, Messanvi Gbeassor, Gloria Proni, and Nasri Nesnas. *Epiafzelechin from the Root Bark of Cassia sieberiana: Detection by DART Mass Spectrometry, Spectroscopic Characterization, and Antioxidant Properties*. Journal of Natural Products, **74(3)**, 455-459 (2011).

Corresponding author: **Ngo Quoc Luan**
School of Education, Cantho University
Campus 2, 3/2 Street, Xuankhanh Ward, Ninhkieu District, Can Tho City
E-mail: ngoquocluan@ctu.edu.vn; Telephone: 0919.054856.