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Original Article

Flavonoids from the Rhizomes of *Kaempferia Parviflora* Wall. Ex Baker

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Abstract: *Kaempferia parviflora* Wall. ex Baker, belonging to the Zingiberaeceae family, has a history of traditional medicinal use in Vietnam for treating various conditions such as inflammation, abdominal pain, indigestion, menstrual cramps, and fatigue. From the *n*-hexane fraction of the rhizomes of *Kaempferia parviflora*, six compounds were isolated and identified as 5-hydroxy-3,7-dimethoxyflavone (**1**), 5,7-dimethoxychrysin (**2**), 5-hydroxy-7-methoxyflavone (**3**), 5-hydroxy-3,4',7-trimethoxyflavone (**4**), 4',5,7-trimethoxyflavone (**5**), and 3,4',5,7-tetramethoxyflavone (**6**). The structures of compounds **1-6** were determined using Mass Spectrometry (MS), and 1D and 2D Nuclear Magnetic Resonance (NMR) techniques.

Keywords: Kaempferia parviflora, Zingiberaceae, flavonoid, black ginger.

1. Introduction

Zingiberaceae is a family of diverse genus and rich species comprises about 47-56 genus and 1075-1600 species and widely distributed in tropical countries [1]. *Kaempferia*, a popular genus in the Zingiberaceae family, has 66 accepted species, which distributed in tropical and subtropical climates of Asia [2], mainly in Vietnam, Laos, Cambodia, China, India, and Thailand. The main components of the genus *Kaempferia* were essential oils, diterpenes, phenolic compounds, sterols, and flavonoids [3-5], which exhibited anti-allergy [6], anti-inflammatory [7, 8], anti-stress [9], antibacterial [10], and anti-cancer [11, 12] activities. Black ginger (*Kaempferia parviflora* Wall ex Baker), also known as "Ngải đen" in An Giang province, is considered as an herb are used in folk medicine for the treatment of abdominal pain, indigestion, flatulence, menstrual cramps, fatigue, cold stomach, hepatitis, jaundice, wound swelling and pain [13-15]. Recently, the extract and/or methoxyflavones from *K. parviflora* has been reported with various bioactivities, including anti-plasmodial, antifungal, antimycobacterial [10], NO production inhibitory [8], anti-allergy [6], and prevent cancer cell proliferation [12].

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In this study, we present the extraction, isolation and identification of six flavonoids from the precipitate of *n*-hexane extract of *K. parviflora* rhizomes (Figure 1). The structures of these compounds were elucidated using spectroscopic data and compared to those reported in the literature.

2. Experimental

2.1. Materials and Chemicals

The rhizomes of *K. parviflora* (3.0 kg) was collected in September 2019, in An Giang province, Vietnam. A voucher specimen (KP092019.AG) has been deposited at the University of Medicine and Pharmacy at Ho Chi Minh City (UMP) and was identified by Dr. Vo Van Leo - Department of Pharmacognosy, UMP. The solvents were used for isolation and extraction: ethanol 96%, *n*-hexane, and *n*-butanol (Xilong, China), ethyl acetate and methanol (Fisher Scientific, USA) in analytical reagent grade. Methanol used for semi-preparative HPLC was purchased from Fisher Scientific (USA) and water was distilled by using Aquatron Water Stills A4000 instrument (Bibby Scientific, England) and filtered by 0.22 µm PTFE membrane before used for semi-preparative HPLC.

2.2. General Experimental Procedures

NMR spectra were measured on a JEOL-400 (400 MHz) using TMS as an internal standard. ESI-HRMS spectra were obtained from a Xevo G2-XS QTOF mass spectrometer (Waters, USA). A Shimadzu Model LC-20A Series HPLC system (Japan) with a LC-20AD detector and a Phenomenex Luna 100 RP-C18 (250 × 10 mm i.d., 5 µm) column was used for the semipreparative HPLC. Silica gel (43-63 µm, Merck) and Sephadex LH-20 (GE Healthcare, Sweden) were used for column chromatography and purified the isolated compounds. TLC was performed using normal phase silica gel Merck 60 F_{254} plates. Spots were detected by UV light (254 nm or 365 nm), 1% $FeCl_3$ solution/ethanol 95%, and 5% vanillin-sulphuric acid/absolute ethanol. NMR solvents were purchased from Sigma-Aldrich (USA).

2.3. Extraction and Isolation

The dried rhizomes of *K. parviflora* (3.0 kg) were percolated with 96% EtOH. The extract was concentrated under vacuum pressure with a rotary evaporator. The crude extract was partitioned with increasing polarity solvents to obtain *n*-hexane (H, 50.4 g), ethyl acetate (E, 80.2 g), saturated *n*-BuOH (B, 5.2 g), and water extracts (11.7 g), respectively. A precipitate (12 g) from the *n*-hexane extract was separated by open-column chromatography (CC) with silica gel using a gradient elution with EtOAc-MeOH (100:0 to 1:1, v/v) to give 18 sub-fractions (PH1-PH18), according to their TLC profiling. Sub-fraction PH2 (1.67 g) was evaporated in *vacuo* and crystalized in *n*-hexane-ethyl acetate (1:1) to yield compound **1** (1.4 g). After removal solvent, sub-fraction PH12 (458.5 mg) was crystalized in *n*-hexane-ethyl acetate (1:1) to obtain compound **2** (801 mg). Sub-fraction PH7 (105.4 mg) was loaded on a Sephadex LH-20 column, eluting with MeOH to give compounds **3** (26.8 mg) and **4** (26.5 mg), respectively. Sub-fraction PH14 (200 mg) was purified by semi-preparative HPLC (2 mL/min, water:MeOH, 3:7, v/v) to yield compounds **2** (33.1 mg) and **5** (100.5 mg), respectively. Compound **6** (17.3 mg) was obtained from sub-fraction PH17 (51.4 mg) by semi-preparative HPLC (2 mL/min, water:MeOH, 3:7, v/v).

5-Hydroxy-3,7-dimethoxyflavone (**1**): yellow amorphous powder. Molecular formula: $C_{17}H_{14}O_5$. ESI-MS: *m/z* 299.10871 $[M+H]^+$. ^{1}H- and ^{13}C-NMR data (DMSO-d_6): see Tables 1 and 2.

5,7-Dimethoxychrysin (**2**): white amorphous powder. Molecular formula: $C_{17}H_{14}O_4$. ESI-MS: *m/z* 283.11212 $[M+H]^+$. ^{1}H- and ^{13}C-NMR data (DMSO-d_6): see Tables 1 and 2.

5-Hydroxy-7-methoxyflavone (**3**): pale-yellow amorphous powder. Molecular formula: $C_{16}H_{12}O_4$. ESI-MS: *m/z* 269.0937

$[M+H]^+$. ^{1}H- and ^{13}C-NMR data (DMSO-d_6): see Tables 1 and 2.

5-Hydroxy-3,7,4'-trimethoxyflavone (**4**): yellow-brown amorphous powder. Molecular formula: $C_{18}H_{16}O_6$. ESI-MS: *m/z* 329.11578 $[M+H]^+$. ^{1}H- and ^{13}C-NMR data (DMSO-d_6): see Tables 1 and 2.

4',5,7-Trimethoxyflavone (**5**): pale-yellow amorphous powder. Molecular formula: $C_{18}H_{16}O_5$. ESI-MS: *m/z* 313.11204 $[M+H]^+$. ^{1}H- and ^{13}C-NMR data (DMSO-d_6): see Tables 1 and 2.

3,4',5,7-Tetramethoxyflavone (**6**): white amorphous powder. Molecular formula: $C_{19}H_{18}O_6$. ESI-MS: *m/z* 365.10589 $[M+Na]^+$. ^{1}H- and ^{13}C-NMR data (DMSO-d_6): see Tables 1 and 2.

3. Results and Discussion

Compound **1** was isolated as a yellow amorphous powder. The ESI-MS with an $[M+H]^+$ ion at *m/z* 299.10871, suggested a molecular formula of $C_{17}H_{14}O_5$ with 11 degrees of unsaturation. The ^{13}C NMR spectrum showed the signals of 17 carbons, including a carbonyl group at δ_C 178.9 (C-4), five oxygenated carbons at δ_C 165.9 (C-7), 161.5 (C-5), 157.1 (C-9), 156.1 (C-2), and 139.6 (C-3), two non-protonated carbons at δ_C 130.5, C-1′ and 106.0, C-10, and seven methine carbons at δ_C 131.7 (C-4′), 129.3 (C-2′/C-6′), 128.8 (C-3′/C-5′), 98.5 (C-6), and 93.0 (C-8), and two methoxy groups at δ_C 60.6, 7-OCH_3 and 56.7, 3-OCH_3 (Table 1), which suggested that compound **1** was a C_6-C_3-C_6 framework of flavonoid [16]. The ^{1}H NMR spectrum exhibited five consecutive aromatic proton signals of B-ring and *meta*-coupled proton signals of H-6 (δ_H 6.40, d, J = 2.0 Hz) and H-8 (δ_H 6.78, d, J = 2.0 Hz). In addition, the ^{1}H-NMR spectrum displayed the singlet signal of 5-OH at δ_H 12.55 and signals of two methoxy groups at δ_H 3.83 (3H, s, 7-OCH_3) and 3.87 (3H, s, 7-OCH_3) as shown in Table 2. The HMBC spectrum revealed the correlations from 5-OH to C-5/C-6/C-10, from H-6 to C-5/C-7/C-8/C-10, from H-8 to C-6/C-7/C-9/C-10, from H-2′/H-6′ to C-2/C-1′/C-3′/C-4′/C-5′, and from H-3′/H-5′ to C-1′/C-2′/C-6′ (Figure 2). Based on the above evidence and detailed analyses of the NMR spectra, compound **1** was determined as 5-hydroxy-3,7-dimethoxyflavone [17, 18].

Compound **2** was obtained as a white amorphous powder. Its molecular formula was determined as $C_{17}H_{14}O_4$ from the positive ESI-MS ion peak at *m/z* 283.11212 $[M+H]^+$, consistent with 11 degrees of unsaturation. The ^{13}C NMR spectrum showed signals of 17 carbons, including a carbonyl group at δ_C 175.7 (C-4), four oxygenated carbons at δ_C 163.8 (C-7), 160.3 (C-5), 159.6 (C-9), and 159.2 (C-2), two non-protonated carbons at δ_C 130.9, C-1′ and 108.4, C-10, and eight methine carbons at δ_C 131.4 (C-4′), 129.1 (C-2′/C-6′), 125.9 (C-3′/C-5′), 108.3 (C-3), 96.3 (C-6), and 93.4 (C-8), and two methoxy groups at δ_C 56.1, 5-OCH_3 and 56.0, 7-OCH_3 (Table 1). Six carbon signals of the B-ring of **2** were similar to those of **1**. The main difference was the decrease of 16 mass units by the loss of a hydroxy group in **2** compared to **1**. The ^{1}H NMR spectrum of **2** showed the singlet signal of H-3 at δ_H 6.79, a signal of *meta*-coupled protons between H-6 (δ_H 6.52 (1H, d, J = 2.4 Hz) and H-8 (δ_H 6.88, d, J = 2.4 Hz) and five consecutive aromatic protons at δ_H 7.57-8.05 were assigned to the structure of B-ring (Table 2). The key HMBC correlations of **2** were shown in Figure 2. In HMBC spectrum, two methoxy groups at δ_H 3.84 and 3.91 showed correlations with C-5 and C-7, respectively. Based on the comparison of the spectral data, the structure of compound **2** was determined as 5,7-dimethoxychrysin [17, 18].

Compound **3** was isolated as a pale-yellow amorphous powder. The molecular formula was $C_{21}H_{20}O_{11}$ based on the positive ESI-MS spectrum with an ion peak at *m/z* 269.0932 $[M+H]^+$. The ^{1}H and ^{13}C NMR spectra of compound **3** were closely related to those of **2**. The main difference was the change of the methoxy group in **2** at carbon C-5 by the hydroxyl group in **3**, which was demonstrated by the HMBC correlations between 5-OH (δ_H 12.82, s) and C-5 (δ_C 161.2)/C-6 (δ_C 98.2)/C-10 (δ_C 105.0) (Figure 2). The key HMBC and

COSY correlations of **3** were shown in Figure 2, from 5-OH to C-5/C-6/C-10, from H-6 to C-5/C-7/C-8/C-10, from H-8 to C-6/C-7/C-9/C-10, from H-2′/H-6′ to C-2/C-1′/C-3′/C-4′/C-5′, and from H-3′/H-5′ to C-1′/C-2′/C-6′. In comparison with the published data in the literature [18], compound **3** was identified as 5-hydroxy-7-methoxyflavone.

Compound **4** was isolated as an amorphous yellow powder. The positive ESI-MS spectrum showed a pseudomolecular ion peak at *m/z* 329.11578 $[M+H]^+$, corresponding to the molecular formula of $C_{18}H_{16}O_6$. The ^{13}C NMR spectrum showed the presence of 18 carbon signals, including a carbonyl group at δ_C 178.0 (C-4), six oxygenated carbons at δ_C 165.1 (C-7), 161.4 (C-4′), 160.8 (C-5), 156.3 (C-9), and 155.2 (C-2), two non-protonated carbons at δ_C 122.0, C-1′ and 105.2, C-10, and six methine carbons at δ_C 130.0 (C-2′/C-6′), 114.2 (C-3′/C-5′), 96.3 (C-6), and 93.4 (C-8), and three methoxy groups at δ_C 59.7 (3-OCH_3), 56.0 (4′-OCH_3), and 55.3 (7-OCH_3) (Table 1). In addition, the ^{13}C NMR spectrum exhibited two symmetric carbon signals at δ_C 130.0 (C-2′/C-6′) and 114.2 (C-3′/C-5′), which was inferred to be a flavonoid with a symmetric B-ring [16].

Table 1. ^1H NMR (400 MHz) spectroscopic data of compounds **1**-**6**

No.	**1**	**2**	**3**	**4**	**5**	**6**
	δ_H mult. (*J* in Hz)	δ_H mult. (*J* in Hz)	δ_H mult. (*J* in Hz)	δ_H mult. (*J* in Hz)	δ_H mult. (*J* in Hz)	δ_H mult. (*J* in Hz)
H-3	-	6.79 s	7.05 s	-	6.68 s	-
H-6	6.40 d (2.0)	6.52 d (2.4)	6.41 d (2.4)	6.39 d (2.4)	6.50 d (2.4)	6.49 d (2.0)
H-8	6.78 d (2.0)	6.88 d (2.4)	6.83 d (2.4)	6.78 d (2.4)	6.86 d (2.4)	6.80 d (2.0)
H-2′	8.07 m	8.05 m	8.11 d (8.4)	8.07 d (8.8)	8.00 d (8.8)	8.04 d (9.2)
H-3′	7.60 m	7.57 m	7.60 m	7.15 d (8.8)	7.10 d (8.8)	7.12 d (9.2)
H-4′	7.60 m	7.57 m	7.60 m	-	-	-
H-5′	7.60 m	7.57 m	7.60 m	7.15 d (8.8)	7.10 d (8.8)	7.12 d (9.2)
H-6′	8.07 m	8.05 m	8.11 d (8.4)	8.07 d (8.8)	8.00 d (8.8)	8.04 d (9.2)
3-OCH_3	3.83 s			3.81 s		3.74 s
5-OCH_3		3.84 s			3.83 s	3.84 s
7-OCH_3	3.87 s	3.91 s	3.88 s	3.87 s	3.90 s	3.89 s
4′-OCH_3				3.87 s	3.85 s	3.85 s
5-OH	12.55 s	-	12.82 s	12.64 s	-	-

The ^{13}C and ^1H NMR spectra of **4** were close to those of **1**, except for the increase of 31 mass units in **4** compared to **1**. In this regard, the addition of methoxy group in **4** was linked to carbon C-4′ by the correlation of 4′-OCH_3 (δ_C 3.87) observed in the HMBC spectrum (Figure 2). The ^1H NMR and ^1H-^1H COSY spectra revealed the signals of a pair of symmetric protons between H-2′/H-6′ (δ_H 8.07, d, *J* = 8.8 Hz) and H-3′/H-5′ (δ_H 7.15, d, *J* = 8.8 Hz) and a *meta*-coupled proton signal between H-6 (δ_H 6.39, d, *J* = 2.4 Hz) and H-8 (δ_H 6.71, d, *J* = 2.4 Hz) (Table 2).

In the HMBC spectrum, correlations were observed from 5-OH to C-5/C-6/C-10, from H-6 to C-5/C-7/C-8/C-10, from H-8 to C-6/C-7/C-9/C-10, from H-2′/H-6′ to C-2/C-1′/C-3′/C-4′/C-5′, and from H-3′/H-5′ to C-1′/C-2′/C-6′ (Figure 2). In comparison with the published data in the literature, compound **4**

was determined as 5-hydroxy-3,4',7-trimethoxyflavone [18].

Compound **5** was isolated as a pale-yellow amorphous powder. It has the molecular formula $C_{15}H_{10}O_7$ determined by the positive ESI-MS ion peak at *m/z* 313,11204 $[M+H]^+$. The ^{13}C NMR and HSQC data showed 18 carbon signals in the downfield region of δ_C 176-93 of a flavone aglycon [16]. The ^{13}C and ^1H-NMR spectroscopic data of **5** were close to those of **2**, except for the addition of the methoxy group linked to carbon C-4′ in **5**, which was observed by the correlation from 4′-OCH_3 to C-4′ in the HMBC spectrum (Figure 2). Similar to compound **4**, the ^1H NMR and ^1H-^1H COSY spectra revealed the signals of a pair of symmetric protons between H-2′/H-6′ (δ_H 8.00, dd, *J* = 8.8 Hz) and H-3′/H-5′ (δ_H 7.10, dd, *J* = 8.8 Hz) of the B-ring (Table 2). The key HMBC and ^1H-^1H COSY correlations of **5** were shown in Fig. 2. From the above evidence of NMR spectra and ESI-MS spectrum data, compound **5** was identified as 4',5,7-trimethoxyflavone in comparison with the spectroscopic data reported in the literature [17, 18].

Compound **6** was isolated as a white amorphous powder. The ESI-MS spectral data exhibited a negative-ion peak at *m/z* 365.10589 $[M+Na]^+$, corresponding to the molecular formula of $C_{19}H_{18}O_6$ and 11 degrees of unsaturation. The ^{13}C NMR spectrum of compound **6** showed 19 carbon signals, containing one carbonyl group at δ_C 172.7, C-4, six aromatic methine carbons at δ_C 130.1 (C-2′/C-6′), 114.7 (C-3′/C-5′), 96.4 (C-6), and 93.5 (C-8), eight non-protonated carbons at δ_C 164.2 (C-7), 161.4 (C-4′), 160.9 (C-5), 158.7 (C-9), 152.3 (C-2), 140.8 (C-3), 123.0 (C-1′), and 109.0 (C-10), and four methoxy groups at δ_C 59.8 (3-OCH_3), 56. 7 (4′-OCH_3), 56.5 (5-OCH_3), and 55.9 (7-OCH_3) (Table 1). All carbon signals appeared in the down-field region from δ_C 172-93 of a C_6-C_3-C_6 framework of polymethoxyflavone derivatives [16].

The ^1H NMR and ^1H-^1H COSY spectra showed the correlations of a pair of symmetric protons signals between protons H-2′/H-6′ and H-3′/H-5′ of the B-ring, and a *meta*-coupled proton signal between H-6 and H-8 of A-ring. The ^{13}C and ^1H NMR spectra of **6** were close to those of **4**, except for the presence of the methoxy group at C-5 in **6** instead of hydroxyl group in **4**. The key HMBC and ^1H-^1H COSY correlations were observed as shown in Fig. 2. Based on the comparison of the spectral data [19], compound **6** was identified as 3,4',5,7- etramethoxyflavone.

Table 2. ^{13}C NMR (100 MHz) spectroscopic data of compounds **1**-**6**

No	**1**	**2**	**3**	**4**	**5**	**6**
	δ_C	**δ_C**	**δ_C**	**δ_C**	**δ_C**	**δ_C**
2	156.1	159.2	163.5	155.5	160.2	152.3
3	139.6	108.3	105.4	138.1	107.3	140.8
4	178.9	175.7	182.2	178.0	176.2	172.7
5	161.5	160.3	161.2	160.8	160.8	160.9
6	98.5	96.3	98.2	97.7	96.7	96.4
7	165.9	163.8	165.4	165.1	164.2	164.2
8	93.0	93.4	92.9	92.3	93.9	93.5
9	157.1	159.6	157.4	156.3	159.7	158.7
10	106.0	108.4	105.0	105.2	108.8	109.0
1′	130.5	130.9	130.6	122.	123.6	123.0
2′	129.3	129.1	129.2	130.0	128.3	130.1

3′	128.8	125.9	126.5	114.2	115.0	114.7
4′	131.7	131.4	132.2	161.4	162.3	161.4
5′	128.8	125.9	126.5	114.2	115.0	114.7
6′	129.3	129.1	129.2	130.0	128.3	130.1
3-OCH_3	56.7			59.7		59.8
5-OCH_3		56.1			56.0	56.5
7-OCH_3	60.6	56.0	56.2	55.3	56.5	55.9
4′-OCH_3				56.0	56.6	56.7

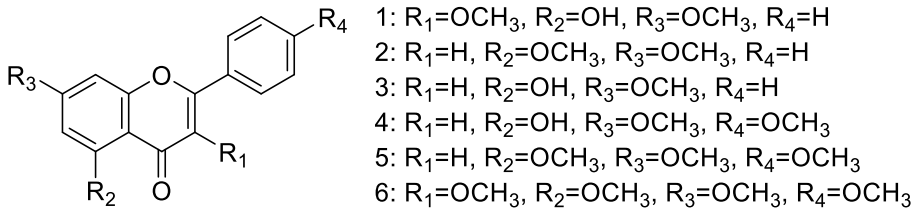

Figure 1. Chemical structures of the isolated compound **1-6**.

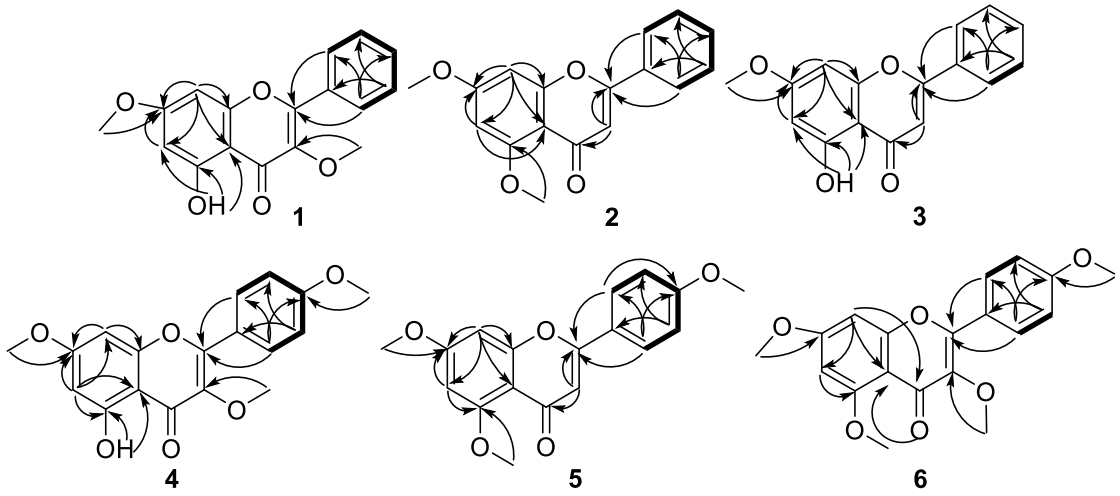

Figure 2. The key HMBC (⟶) and COSY (▬) correlations of compounds **1-6**.

The previous studies on the rhizomes of *K. parviflora* collected in Daklak province reported the isolation of eight flavonoids, including 5,7-dimethoxy flavanone, 5-hydroxy-3,7-dimethoxyflavone, 3,7,4'-trimethyl kaempferol, apigenin trimethyl ether, tectochrysin, apigenin 7,4′-dimethyl ether, 3,5,7-trimethoxyflavone, 5,7-dimethoxyflavone and two sterols (*β*-sitosterol and daucosterol) [17, 20]. Five compounds, including 5,7-dimethoxyflavone, 3,5,7-trimethoxyflavone, di-*O*-methylpinocembrin, bisdemethoxycurcumin, aloe-emodin were isolated from the rhizomes of *K. parviflora* collected in An Giang province [4]. Recently, twenty-three compounds including six phenolic glycosides (*kaempanoside* A-C, kaempferiaoside E, rel-(5a*S*,10b*S*)-5a,10b-dihydro-1,3,5a,9-tetrahydroxy-8-methoxy-6*H*-benz[b]indeno[1,2-d]furan-6-one 5a-*O*-[α-*L*-rhamnopyranosyl-(1→6)-β-*D*-glucopyranoside],

rel-(5a*S*,10b*R*)-5a,10b-dihydro-1,3,5a,9-tetrahydroxy-8-methoxy-6*H*-benz[b]indeno[1,2-d]furan-6-one 5a-O-[α-*L*-rhamnopyranosyl-(1→6)-β-*D*-glucopyranoside], thirteen flavones (3,7-dimethoxyflavanone, *trans*-3-hydroxy-5,7-dimethoxyflavanone, taxifolin, 5-hydroxy-3,7-dimethoxyflavone, 5-hydroxy-3,7,3′,4′-tetramethoxyflavone, kaempferol, 5-hydroxy-7-methoxyflavone, 5-hydroxy-3,7,4′-trimethoxyflavone, trimethoxyflavone, 5-hydroxy-7,3′,4′-trimethoxyflavone, 3,5,7,3′,4′-pentamethoxyflavone, 5,7,4′-trimethoxyflavone, 5-hydroxy-7,4′-dimethoxyflavone), and four phenolic compounds (methyl 3,5-dimethoxybenzoate, vanillic acid, methyl vanillate, and methyl syringate) were isolated from the rhizomes of *K. parviflora* collected in Thai Nguyen province [18]. Regarding this study, compound **6** was presented as the newly-isolated constituent of *K. parviflora* collected in An Giang province. As reported in literature, 5-hydroxy-3,7-dimethoxyflavone (**1**) moderatly inhibited NO production on RAW 264.7 with IC_{50} value of 41.6 μM [8]. 5,7-Dimethoxyflavone (**2**), a chrysin derivative significantly suppressed the development of preneoplastic lesions induced by N-nitrosodiethylamine (DEN) in Wistar rats [11]. 5-Hydroxy-7-methoxyflavone (**3**) showed the ability to prevent cancer cell proliferation at a concentration of 25 μM and triggered cell death via reactive oxygen species (ROS) signaling in colorectal carcinoma cells (HCT-116 cell line) [12]. 5-Hydroxy-3,7,4'-trimethoxyflavone (**4**) exhibited weak anti-allergy activity against the release of enzyme *β*-hexosaminidase from RBL-2H3 cells with IC_{50} value of 49.9 μM [6]. 5,7,4'-Trimethoxyflavone (**5**) showed a potential antiplasmodial activity against *Plasmodium falciparum* with IC_{50} value of 3.70 μg/ml [10] and potential inhibitory effects on *β*-secrectase (BACE1) [21], and 3,5,7,4′-Tetramethoxyflavone (**6**) showed antifungal activity against *Candida albicans* with respective IC_{50} value of 39.71 μg/ml and moderate antimycobacterial activity with a minimum inhibitory concentrations (MIC) of 200 μg/ml [10].

4. Conclusion

In conclusion, six flavonoids were isolated and purified from the precipitate of the *n*-hexane extract of *Kaempferia parviflora* rhizomes using various chromatographic methods. Their structures, namely 5-hydroxy-3,7-dimethoxyflavone (**1**), 5,7-dimethoxychrysin (**2**), 5-hydroxy-7-methoxyflavone (**3**), 5-hydroxy-3,4',7-trimethoxyflavone (**4**), 4',5,7-trimethoxyflavone (**5**), and 3,4',5,7-tetramethoxyflavone (**6**) were determined through detailed analysis of 1D and 2D NMR spectral data and comparison with published literature. Among them, compound **6** was first reported from the rhizomes of *K. parviflora* collected in Vietnam.

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