Cytotoxic Geranylated Xanthones and *O*-Alkylated Derivatives of α-Mangostin

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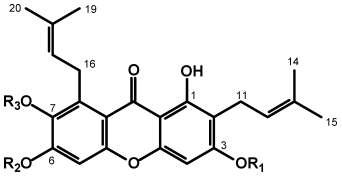
Two new geranylated xanthones, 6-*O*-methylcowanin (4) and oliverixanthone (5), along with five known compounds, cowanin, rubraxanthone, cowaxanthone, cowanol, and β-mangostin, have been isolated from the bark of *Garcinia oliveri*. For comparison of their biological activities, one mono- and seven di-*O*-alkylated α-mangostin derivatives were synthesized from α-mangostin. The structures of all compounds were assigned by spectroscopic methods (1D and 2D NMR and MS). Cytotoxicity of selected xanthones against MCF-7 and DLD-1 cell lines was examined. Evaluation of the structure–activity relationship showed that α-mangostin had the strongest activity, and all the *O*-alkylated α-mangostin derivatives showed reduced activity compared to the naturally occurring α-mangostin.

Key words *Garcinia oliveri*; xanthone; cytotoxicity; 6-*O*-methylcowanin; oliverixanthone; *O*-alkylated α-mangostin

Xanthones are widely distributed in the genus *Garcinia* (Guttiferae), which provides compounds with a wide range of pharmacological effects such as cytotoxic, anti-bacterial, antioxidant and anti-human immunodeficiency virus (HIV) activities.[1—3] The diverse substitution patterns of xanthones provide a good basis for different biological activity and for structure–activity relationship studies.[4]

α-Mangostin (**1**), the dominating constituent of the fruit hulls of *Garcinia mangostana*, together with β-mangostin (**2**) and γ-mangostin (**3**) (Fig. 1), have exhibited various biological activities, such as cytotoxicity in different cell lines together with anti-oxidative, anti-inflammatory, and anti-bacterial activities.[5] As examples, inhibition of the cell proliferation of DLD-1 cells by **1** was observed at concentrations above 2 μM with an IC_{50} value at 7.5 μM.[6] This value corresponds to the activity observed in HL-60 cells.[7] **3** seems to have a stronger effect compared with **1** and **2**[8] but this is not confirmed by all reports.[7—9] Besides their slightly different potencies, three mangostins (**1**—**3**) differ in their mechanism of action. **3** caused accumulation of the cells in S-phase, whereas the cells treated with **1** and **2** showed G_0/G_1 accumulation. Furthermore, the three mangostins (**1**—**3**) showed different expression profiles for proteins involved in the cell cycle regulation.[8] The cytotoxic activity of **1** is only partly explained by apoptosis, as higher concentrations are needed for apoptosis than necessary for cytotoxicity.[6] Apoptosis seems to be caused by **1**-induced dysfunction of mitochondria.[10]



R_1=R_2=H; R_3=CH_3 (1)
R_1=R_3=CH_3; R_2=H (2)
R_1=R_2=R_3=H (3)
R_1=R_2=-CH_2CH=$C(CH_3)_2$; R_3=CH_3 (10)
R_1=R_2=-$CH_2C_6H_4OCH_3$; R_3=CH_3 (11)
R_1=R_2=-$CH_2C_6H_5$; R_3=CH_3 (12)
R_1=R_2=-$CH_2C_6H_4CH_3$, R_3=CH_3 (13)
R_1=R_2=-$(CH_2)_4CH_3$; R_3=CH_3 (14)
R_1=R_2=-$(CH_2)_3CH_3$; R_3=CH_3 (15)
R_1=R_2=R_3=CH_3 (16)
R_1=H; R_2=-CH_2CH=CH_2; R_3=CH_3 (17)

Fig. 1. Structures of Mangostins (**1**—**3**) and *O*-Alkylated α-Mangostin (**10**—**17**)

Beside **1**—**3**, several other xanthones have been tested for their cytotoxic activity and a structure–activity relationship can partly be elucidated. In all such experiments, the mangostins showed the strongest effect.[7—9] Analysis of the cytotoxic effect of 6-methoxy-β-mangostin[8] indicated that the free hydroxyl group at C-6 was necessary for the cytotoxic activity. Furthermore, the apoptosis activity also depends on the number of free hydroxyl groups.[7,9,10]

Pyranoxanthones, produced naturally or synthetically from **1** by acid-catalysed cyclisation of the prenyl group, have been found to have reduced cytotoxic potency compared with **1**.[9] For 3,6-di-*O*-alkylated α-mangostin, the inhibitory effects on antifungal activity were reduced dramatically as compared with α-mangostin.[11] However, the cytotoxic effects of these derivatives on MCF-7 cells (human breast cancer cells) or DLD-1 (human colon cancer cells) have not been investigated yet.

In this paper, we report the isolation of two new geranylated xanthones (**4**, **5**) and five known compounds (**2**, **6**—**9**) from the bark of *Garcinia oliveri* PIERRE, a Vietnamese tall tree whose young buds and sour fruit are used for soup cooking. However, no use in folk medicine nor identification of specific compounds has been previously reported.[12] Furthermore, conversion of α-mangostin into *O*-alkylated α-mangostin derivatives (**10**—**17**) is described. Cytotoxic activity for compounds **1**—**3** and **6**—**17** was analysed in two human tumor cell lines, MCF-7 and DLD-1, to investigate the structure–activity relationship.

Results and Discussion

A petroleum ether extract of the bark of *G. oliveri* was chromatographed using column chromatography (CC) and

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preparative TLC to give 6-*O*-methylcowanin (**4**), oliverixanthone (**5**), cowanin (**6**),[13] cowaxanthone (**7**),[14] rubraxanthone (**8**),[15,16] cowanol (**9**),[13] and β-mangostin (**2**).[15]

6-*O*-Methylcowanin (**4**) was obtained as a yellow gum, which reacted positively with $FeCl_3$ reagent. The molecular formula was found to be $C_{30}H_{36}O_6$ (*m/z* 491.2423 $[M-H]^-$). The UV [λ_{max} 245, 268, 315 and 368 nm] and IR spectra [ν_{max} 3431 (O–H), 1639 (chelated C=O), 1639 (aromatic ring) cm^{-1}] indicated the presence of a xanthone skeleton in the molecule.

The ^{1}H-NMR spectrum (Table 1) exhibited signals for a chelated hydroxyl group [δ 13.87 (1H, s, 1-OH)], two isolated aromatic protons [δ 6.76 and 6.29 (1H each, s, H-5 and H-4)], a 3-methylbut-2-enyl group [δ_H 5.27 (1H, m, H-12), 3.46 (2H, d, J=7.2 Hz, H_2-11), 1.85 and 1.78 (3H each, s, H_3-14 and H_3-15)], a geranyl group [δ 5.27 (1H, m, H-17), 5.03 (1H, m, H-21), 4.14 (2H, d, J=6.0 Hz, H_2-16), 2.00 (4H, m, H_2-19 and H_2-20)], 1.85, 1.61, and 1.56 (3H each, s, H_3-24, H_3-25, and H_3-23)], and two methoxy [δ 3.97 and 3.79 (3H each, s, 6-OCH_3 and 7-OCH_3)]. The ^{13}C-NMR spectrum (Table 1) revealed resonances for 30 carbons, comprising a conjugated carbonyl carbon (δ 182.1, C-9), twelve aromatic carbons, a geranyl, a prenyl, and two methoxyl groups [δ 61.0 and 56.1, 7-OCH_3 and 6-OCH_3).

The spectra are quite similar to those of cowanin (**6**) previously isolated from *G. cowa*.[13] The only difference is the appearance of one additional methoxyl group in **4**. In the heteronuclear multiple bond correlation (HMBC) plot (Fig. 2), the chelated hydroxyl proton (δ_H 13.87) showed cross-peaks with an oxygenated aromatic carbon (δ_C 160.7, C-1), and two substituted aromatic carbons [δ_C 108.4 (C-2) and 103.8 (C-9a)]. Meanwhile, the less deshielded singlet proton (δ_H 6.29) showed correlations to C-2, C-9a, and two oxygenated aromatic carbons [(δ_C 161.6 (C-3) and 155.5 (C-4a)], indicating that C-4 carried the proton and C-3 was oxygenated. A cross-peak observed between the free hydroxyl proton (δ_H 6.15) and C-3 showed that the hydroxyl group was bonded to C-3. The benzylic methylene protons of the isoprenyl side chain (δ_H 3.46, H_2-11) correlated to C-2, revealing the linkage of this group to the carbon.

The downfield shift of the benzylic methylene protons of the geranyl group (δ_H 4.16, H_2-16) revealed that the group was *peri* to the carbonyl carbon. The protons gave cross-peaks with an oxygenated aromatic carbon (δ_C 144.0) and two substituted aromatic carbons [δ_C 137.5 (C-8) and 112.0 (C-8a)], indicating that C-7 was oxygenated. The less deshielded methoxyl protons (δ_H 3.79) correlated to C-7, showing the attachment of the methoxyl group to the carbon. Cross-peaks between the remaining isolated aromatic proton (δ_H 6.76, H-5) with C-7, C-8a and two oxygenated aromatic carbons (δ_C 158.1, 155.5) indicated that C-5 was protonated and C-6 carried the methoxyl group. This was confirmed by the correlation observed between the methoxyl protons with the oxygenated aromatic carbon at δ_C 158.1. The structure of **4** was therefore determined to be (*E*)-1,3-dihydroxy-6,7-dimethoxy-2-(3-methylbut-2-enyl)-8-(3,7-dimethyl-2,6-octadienyl)xanthone or 6-*O*-methylcowanin, which is a new natural product.

Oliverixanthone (**5**) was isolated as a yellow gum, $C_{30}H_{36}O_7$ (*m/z* 507.2394 $[M-H]^-$). The UV, IR (Experimen-

Table 1. NMR Data of Compounds **4** and **5** (δ in $CDCl_3$)[a,b]

#	**4**		**5**	
	δ_H	δ_C	δ_H	δ_C
1		160.7		158.2
2		108.4		107.6
3		161.6		166.2
4	6.29 s	93.2	6.29 s	88.1
4a		155.5		157.1
5	6.75 s	98.3	6.77 s	98.3
6		158.1		158.1
7		144.0		144.2
8		137.5		137.3
8a		112.0		112.0
9		182.1		182.1
9a		103.8		104.3
10a		155.1		155.5
11	3.46 d (6.9)	21.5	3.18 dd, 3.14 dd (15.6, 8.7)	26.9
12	5.27 m	121.5	4.78 t (8.7)	91.8
13		135.3		72.0
14	1.85 s	17.7[c]	1.36 s	25.9[c]
15	1.78 s	25.9[d]	1.24 s	23.8[d]
16	4.14 d (6.6)	26.1	4.14 d (6.6)	26.1
17	5.27 m	124.5	5.29 m	123.5
18		135.9		135.4
19	2.00 m	39.7	2.02 m	39.9
20	2.00 m	26.9	1.99 m	26.7
21	5.03 m	123.2	5.04 m	123.2
22		131.2		131.2
23	1.56 s	17.9[c]	1.55 s	17.7[c]
24	1.85 s	16.5	1.85 s	16.5
25	1.61 s	25.6[d]	1.60 s	25.6[d]
1-OH	13.87 s		13.67 s	
3-OH	6.13 s			
6-OMe	3.97 s	56.1	3.97 s	56.1
7-OMe	3.79 s	61.0	3.79 s	61.0

a) Chemical shift values were in ppm and *J* values (in Hz) were presented in parentheses. *b*) The assignments were based on the ^{1}H-NMR, ^{13}C-NMR, HMQC and HMBC experiments. *c*, *d*) Interchangeable.

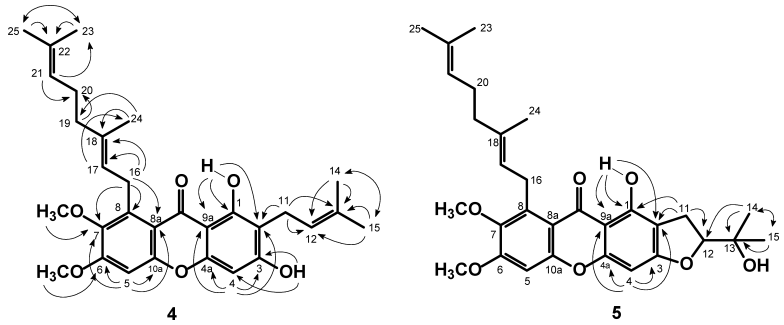

Fig. 2. HMBC Correlations for Compounds **4** and **5**

tal), ^{1}H- and ^{13}C-NMR spectra (Table 1) have close similarities to **4**. However, the signals for a 3-methylbut-2-enyl group are replaced by resonances for a dihydrofuran ring [δ_H 3.18 and 3.14 (1H each, dd, J=15.6, 8.7 Hz, H_2-11), 4.78 (1H, t, J=8.7 Hz, H-12), 1.36 and 1.24 (3H each, s, H_3-14 and H_3-15); δ_C 91.8 (C-12), 72.0 (C-13), 26.9 (C-11), 25.9 (C-14) and 23.8 (C-15)].

Comparison of the ^{1}H- and ^{13}C-NMR spectra as well as the HMBC plot of **5** (Fig. 2) with those of **4** confirmed that both compounds possessed the same xanthone B ring. In the xanthone A ring, correlations of the chelated hydroxyl proton helped to identify the shifts for C-1, C-2 and C-9a (δ_C 158.2, 107.6 and 104.3, respectively). The isolated aromatic proton (δ_H 6.29) gave cross-peaks to C-2, C-9a and two oxygenated aromatic carbons (δ_C 166.2 and 157.1), indicating that C-4 was protonated and C-3 carried an oxygen. The benzylic methylene protons of the dihydrofuran ring (H_2-11) showed correlations to C-1 and C-2, revealing the fusion of the dihydrofuran ring at C-2 and C-3 with an ether linkage at C-3. The structure of oliverixanthone (**5**) was thus established to be (*E*)-2,3-dihydro-4-hydroxy-2-(1-hydroxy-1-methylethyl)-7,8-dimethoxy-6-(3,7-dimethyl-2,6-octadienyl)-5*H*-furo[3,2-*b*]xanthen-5-one, which has hitherto not been reported.

Treatment of α-mangostin (**1**) with alkyl halides and K_2CO_3 in acetone afforded 3,6-di-*O*-alkylated products (**10**—**16**), except in the case where the alkyl halide was propenyl chloride. In this case only the 6-mono-*O*-alkylated derivative (**17**) was obtained (Fig. 1). NMR assignments for **1** (Experimental) were established by comparison of its spectral data with those of literature values.[15] The *O*-alkylation at C-3 and C-6 led to a decrease of approximately 4 ppm in the chemical shifts of C-4 and C-5, respectively. For **17**, the chemical shift of C-5 (δ 99.3 ppm) reduced 4 ppm relative to **1**, indicating that the *O*-alkylation occurred at C-6.

Four of the xanthones isolated from *G. oliveri* (**6**—**9**) (Fig. 3) and α-mangostin (**1**) were tested for their effect on cell number using the SRB assay. The results are expressed as IC_{50} (μM) values after 48 h (or 72 h) of exposure of the compounds to human cell lines, derived from breast (MCF-7) and colon (DLD-1). Cowanin (**6**) and cowanol (**9**) were the most active in reducing the cell number tested on MCF-7 cell lines (IC_{50} values around 15 μM) whilst **6**, **9** and **1** showed the highest cytotocixity on DLD-1 (IC_{50} values around 18 μM) (Table 2). Our IC_{50} values in DLD-1 cells were about 2.5 times higher than previously found by Nakagawa *et al.*[6] Furthermore, it was recently shown that rubraxanthone (**8**) had some cytotoxicity against HL-60 cells with IC_{50} around 18 μM,[16] but in our hand, **8** have an IC_{50} values 3 times higher than reported by Ee and Cheow.[16]

Cowanin (**6**) and cowanol (**9**) had the same cytotoxic activity although they have different side chains at C-2. This showed that the replacement of a methyl group at C-13 by a hydroxymethyl group does not affect the cytotoxicity. The removal of the prenyl group at C-2 in **6** as found in **8** or the switching of the geranyl group from C-8 as in **8** to C-2 as in **7** resulted in a decrease of activity (IC_{50}=34 μM for **8** and 24 μM for **7**). Furthermore, as both **6** and **9** are less active than **1**, the length of the alkyl side chains at C-2 or C-8 is obviously also important.

To identify the most active mangostin derivative, in total eight derivatives were prepared (**10**—**17**) and they were tested for their cytotoxic effect as MTT assay in DLD-1 cells at a single concentration (Table 3). The parental mangostins (**1**—**3**) were the most active also in the MTT assay, whereas the substituted mangostins only showed marginal effects. This observation fits with the apoptosis-inducing activity[8] and cell proliferation inhibition[10] activities reported by Ma-

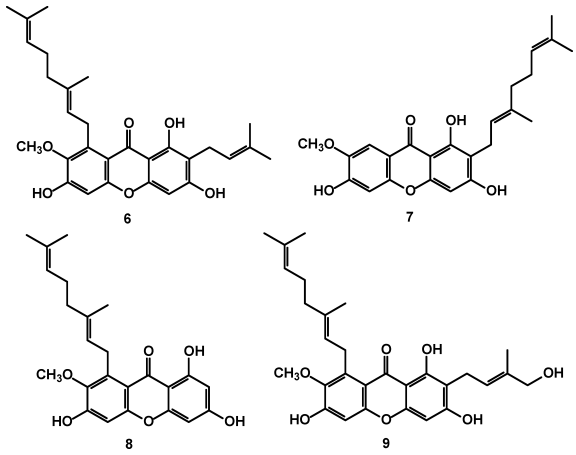

Fig. 3. Structures of Compounds **6**—**9**

Table 2. Cell Number Inhibitory Effects *in Vitro* of Compounds **6**—**9** Isolated from *G. oliveri* Compared with α-Mangostin (**1**)

Compound	IC_{50} (μM)		
	MCF-7	DLD-1	
	48 h	48 h	72 h
6	14.8±0.2[a]	18.2±0.2	13.2±0.2
7	24.5±1.0	43.9±2.2	24.4±0.7
8	59.0±2.5	50.7±1.8	33.9±2.2
9	14.9±0.5	18.2±0.1	14.8±2.1
1	ND[b]	19.0±0.3	12.2±0.4
Positive control[c]	40.5±1.2	30.9±2.1	62.1±1.9

The cell number was estimated using the SRB assay. *a*) The numbers are means of three experiments±S.D. *b*) ND: not determined. *c*) % growth inhibition of 0.01% campothecin and 60 μM resveratrol in MCF-7 cells and DLD-1 cells, respectively.

Table 3. Cell Proliferation Inhibitory Effects of Mangostin Derivatives (**10**—**17**) Compared with Compounds **1**—**3**

Compound[b]	Inhibition of cell proliferation[a] (%)	
	48 h	72 h
10	22.2±2.0[c]	8.9±0.7
11	18.7±3.5	20.1±2.3
12	27.4±6.1	14.5±1.4
13	23.6±3.5	44.5±4.5
14	12.3±1.5	9.0±2.2
15	4.4±2.8	3.8±1.7
16	9.0±2.5	24.7±6.2
17	37.1±2.8	66.3±3.2
1	87.1±1.0	96.7±0.4
2	79.3±1.8	93.9±0.3
3	95.4±0.7	100.0±0.1
60 μM resveratrol	37.2±2.7	75.7±3.1

a) The inhibition of cell proliferation was estimated in DLD-1 cells using the MTT assay. *b*) The cells were exposed to the compounds (110 μM of **10**—**17** or 22 μM of **1**—**3**) for 48 or 72 h. *c*) The data presented are means of three experiments±S.D.

tsumoto *et al.* The results showed that γ-mangostin (**3**) having three free hydroxyl groups demonstrated the strongest cytotoxic activity, the mono-*O*-alkylated derivative **17** leads to less inhibition than di-*O*-alkylated compounds. The least active compound is the di-*O*-butyl derivative (**15**) (Table 3). The suggestion by Matsumoto *et al.*[10] about the importance of the number of free hydroxyl groups is now confirmed using DLD-1 cells.

In summary, the inhibition of cell number or cytotoxicity in DLD-1 cells showed that xanthones with free 3,6-dihydroxyl groups and prenyl side chains at C-2 and C-8 in their structures gave strong activity. The removal or replacement of a prenyl group at C-2 or C-8 by a geranyl group reduced the cytotoxicity, and the activities decreased dramatically to be close to zero when the 3,6-dihydroxyl groups were alkylated.

Experimental

General Methods UV spectra were obtained using a Shimadzu-MPS 2000 spectrophotometer or a UV Synergy HT-Bio-Tek microplate ELISA plate reader (for SRB and MTT assay). IR spectra were measured in KBr using a Perkin-Elmer FT-2000 spectrophotometer. NMR spectra were recorded using a Varian Mercury 300 or a Varian-Unity Inova 600 MHz NMR instrument with TMS as an internal standard and $CDCl_3$ or acetone-d_6 as solvents. HR-MS were performed on a Waters Acquity TQD mass spectrometer. Melting points were determined on an Electrothermal 9100 melting point apparatus.

For CC, silica gel Sigma (230—400 mesh) was used and TLC was performed on Merck Si_{60}-GF_{254} and RP_{18}-GF_{254} TLC plates. Cytotoxic activity was set up in Nunclon™ surface 96 wells (Nunc™). 3,3-Dimethylallyl bromide, 4-methoxybenzyl bromide, 4-methylbenzyl bromide, benzyl bromide, *n*-butyl bromide, *n*-pentyl bromide, pentyl cloride, and iodomethane were purchased from Aldrich. SRB and 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) were purchased from Sigma. McCoy's 5A medium, EMEM medium, PBS, and versene were bought from GIBCO.

Plant Material The bark of *G. oliveri* Pierre was collected in Phu Yen Province, central Vietnam. The fruits of *G. mangostana* L. were collected in Binh Duong Province, south Vietnam. The plant material was identified by Mr. Nguyen Huu Tuan, Department of National Resources, Institute of Tropical Biology, HCM City. Voucher specimens (GO-Buanui-09 and GM-Mgcut-03) are deposited at Natural Products and Medicinal Chemistry Research Lab, HCM City University of Natural Sciences.

Extraction and Isolation The dried and ground bark of *G. oliveri* (4.5 kg) was extracted by continuous percolation with hot petroleum ether. Evaporation of the solvent gave a petroleum ether extract (36 g). The petroleum ether extract was separated using CC (10×30 cm, silica gel, with gradient mixture of petroleum ether–acetone 0→100%) to give seven frs. (GOH1-7). Fr. GOH6 (4.5 g) was fractionated by CC (5.0×40 cm, silica gel, petroleum ether–acetone 0→100%) to give seven frs. (GOH6.1—7). Fr. GOH62 (0.54 g) was subjected to CC (silica gel, *n*-hexane–EtOAc 5→50%) followed by CC (1.5×30 cm, silica gel, $CHCl_3$–MeOH 98 : 2) to give 3-*O*-methylcowanin (**4**) (5.5 mg). Fr. GOH624 (21 mg) was refractionated using CC (1.5×30 cm, silica gel, $CHCl_3$–MeOH 98 : 2) and purified by preparative TLC ($CHCl_3$–MeOH 95 : 5) to yield oliverixanthone (**5**) (4 mg). Fr. GOH63 (2.05 g) was fractionated using CC (2.5×40 cm, silica gel, petroleum ether–acetone 5→100%) to give six frs. (GOH63.1—6). Fr. GOH632 (0.72 g) was subjected to CC (2.5×40 cm, silica gel, *n*-hexane–acetone 5→50%) to yield cowanin (**6**) (20 mg).

Fr. GOH7 (5.1 g) was fractionated using CC (5.0×40 cm, silica gel, petroleum ether–acetone 5→100%) to give seven frs. (GOH7.1—7). Fr. GOH72 (0.64 g) was separated by repeated CC on silica gel (2.5×40 cm, *n*-hexane–acetone 5→30% and 1.5×30 cm, $CHCl_3$–MeOH 98 : 2, respectively) followed by recrystallization in $CHCl_3$–*n*-hexane to yield cowaxanthone (**7**) (21 mg) and rubraxanthone (**8**) (4.5 mg). Fr. GOH74 (0.31 g) was subjected to CC (2.5×40 cm, silica gel, *n*-hexane–acetone 5→30%) and purified by CC (1.5×30 cm, silica gel, *n*-hexane–acetone 10→25%) to afford cowanol (**9**) (35 mg). Fr. GOH75 (110 mg) was chromatographed on silica gel (5.0×40 cm, *n*-hexane–acetone 8 : 2) to give β-mangostin (**2**) (25 mg).

The dried and ground fruit hulls of *G. mangostana* (2.4 kg) were successively extracted with petroleum ether and EtOAc respectively using Soxhlet extractor to give a petroleum ether extract (GMH, 36.2 g) and an EtOAc extract (GME, 223 g). 10 g of GME was recrystallized from toluene and then from H_2O–EtOH (1 : 1) at 10 °C to give α-mangostin (**1**)[17] (5 g). 1 g of GME was separated using CC (silica gel, acetone–*n*-hexane 10→50%) to furnish α-mangostin (**1**) (246 mg) and γ-mangostin (**3**)[15] (25 mg).

***O*-Alkylation of α-Mangostin** α-Mangostin (**1**) (102.5 mg, 0.25 mM) was dissolved in acetone (20 ml). Alkyl halides (0.5 mM) and anhydrous K_2CO_3 (50 mg) were added and the solution was refluxed at 40 °C for 4 h. The suspension was filtered, and the filtrate after evaporation was purified by CC (silica gel, EtOAc–*n*-hexane 2→5%). Recrystallization in petroleum ether–acetone gave *O*-alkylated α-mangostins (**10**—**17**).

6-*O*-Methylcowanin (**4**): Yellow gum; UV λ_{max} (EtOH) nm: 245, 268, 315 and 368; IR (KBr) cm^{-1}: 3431, 2927, 2876, 1639, 1578, 825; ^{1}H- and ^{13}C-NMR: see Table 1; HMBC: see Fig. 2; HR-MS *m/z*: 491.2423 $[M-H]^-$ (Calcd for $C_{30}H_{36}O_6$, 491.2434).

Oliverixanthone (**5**): Yellow gum; $[\alpha]_D^{25}$ +71.5° (*c*=0.4, MeOH), UV λ_{max} (EtOH) nm: 251, 268, 315, 343 and 368; IR (KBr) cm^{-1}: 3435, 2923, 2850, 1732, 1653, 1579, 823; ^{1}H- and ^{13}C-NMR: see Table 1; HMBC: see Fig. 2; HR-MS: *m/z* 507.2394 $[M-H]^-$ (Calcd for $C_{30}H_{36}O_7$, 507.2387).

α-Mangostin (**1**): Yellow needles; mp 182—184 °C; EI-MS $C_{24}H_{26}O_6$, *m/z*: 409.1 $[M-H]^-$; ^{1}H-NMR (300 MHz, acetone-d_6) δ: 13.78 (1H, s, 1-OH), 6.80 (1H, s, H-5), 6.38 (1H, s, H-4), 5.27 (2H, t, *J*=6.6 Hz, H-12 and H-17), 4.11 (2H, d, *J*=6.6 Hz, H_2-16), 3.79 (3H, s, 7-OMe), 3.34 (2H, d, *J*=6.6 Hz, H_2-11), 1.85, 1.78 (each 3H, s) and 1.64 (6H, s), H_3-14, H_3-15, H_3-19 and H_3-20; ^{13}C-NMR (75 MHz, acetone-d_6) δ: 182.9 (C, C-9), 163.1 (C, C-3), 161.7 (C, C-1), 157.6 (C, C-6), 156.3 (C, C-10a), 155.8 (C, C-4a), 144.6 (C, C-7), 138.2 (C, C-8), 131.4 (C, C-18), 131.2 (C, C-13), 124.5 (CH, C-17), 123.6 (CH, C-12), 111.9 (C, C-8a), 111.1 (C, C-2), 103.7 (C, C-9a), 102.8 (CH, C-5), 93.2 (CH, C-4), 61.4 (CH_3, 7-OMe), 25.9 and 25.8 (each CH_3, C-15 and C-20), 26.1 (CH_2, C-16), 22.1 (CH_2, C-11), 18.4 and 18.0 (each CH_3, C-14 and C-19).

3,6-Di-*O*-3,3-dimethylallyl α-Mangostin (**10**): Yellow gum; EI-MS $C_{34}H_{42}O_6$, *m/z*: 545.1 $(M-H)^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.52 (1H, s, 1-OH), 6.71 (1H, s, H-5), 6.30 (1H, s, H-4), 5.52 and 5.50 (each 2H, m, H_2-21 and H_2-26), 5.25 (2H, t, *J*=6.6 Hz, H-12 and H-17), 4.13 (2H, d, *J*=6.6 Hz, H_2-16), 3.80 (3H, s, 7-OMe), 3.35 (2H, d, *J*=6.6 Hz, H_2-11), 1.85, 1.84, 1.81×2, 1.79, 1.76, 1.68, and 1.67 (each 3H, s, H_3-14, H_3-15, H_3-19, H_3-20, H_3-24, H_3-25, H_3-29 and H_3-30); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 181.9 (C, C-9), 162.8 (C, C-3), 159.8 (C, C-1), 157.2 (C, C-6), 155.3 (C, C-10a), 155.1 (C, C-4a), 144.2 (C, C-7), 138.9 and 138.2 (C, C-23 and C-28), 137.1 (C, C-8), 131.6 and 131.4 (C, C-13 and C-18), 123.4 and 122.5 (CH, C-12 and C-17), 119.3 and 118.7 (CH, C-22 and C-27), 111.9 (C, C-8a), 111.7 (C, C-2), 103.9 (C, C-9a), 99.1 (CH, C-5), 89.6 (CH, C-4), 65.7 and 65.4 (CH_2, C-21 and C-26), 60.8 (CH_3, 7-OMe), 26.2 (CH_2, C-16), 25.9 and 25.8×3 (CH_3, C-15, C-20, C-25 and C-30), 21.5 (CH_2, C-11), 18.2, 18.4, 18.3 and 17.9 (CH_3, C-14, C-19, C-24 and C-29).

3,6-Di-*O*-4-methoxybenzyl α-Mangostin (**11**): Yellow gum; EI-MS $C_{40}H_{42}O_6$, *m/z*: 649.1 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.50 (1H, s, 1-OH), 7.36 and 6.92 (8H, m, Ar-H), 6.75 (1H, s, H-5), 6.33 (1H, s, H-4), 5.24 (2H, t, *J*=6.6 Hz, H-12 and H-17), 5.09 and 5.06 (each 2H, s, H_2-21 and H_2-28), 3.82 (6H, s, 25-OMe and 32-OMe), 3.80 (3H, s, 7-OMe), 4.13 (2H, d, *J*=6.9 Hz, H_2-16), 3.37 (2H, d, *J*=6.6 Hz, H_2-11), 1.84, 1.73, 1.69 and 1.66 (each 3H, s, H_3-14, H_3-15, H_3-19 and H_3-20); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 182.0 (C, C-9), 162.5 (C, C-3), 159.9 (C, C-1), 157.0 (C, C-6), 159.7 and 159.5 (C, C-25 and C-32), 155.2 (C, C-10a), 155.1 (C, C-4a), 144.2 (C, C-7), 137.2 (C, C-8), 131.7 and 131.5 (C-13 and C-18), 129.3, 129.2, 128.9 and 127.7 (each CH×2, C-23, C-24, C-26, C-27, C-30, C-31, C-33, C-34), 123.3 and 122.4 (CH, C-12 and C-17), 112.2 (C, C-8a), 114.1 and 113.9 (C, C-22 and C-29), 111.8 (C, C-2), 104.1 (C, C-9a), 99.4 (CH, C-5), 89.8 (CH, C-4), 26.2 (CH_2, C-16), 70.5 and 70.1 (CH_2, C-21 and C-28), 60.9 (CH_3, 7-OMe), 25.9 and 25.8 (CH_3, C-15 and C-20), 22.7 (CH_2, C-11), 18.2 and 17.9 (CH_3, C-14 and C-19).

3,6-Di-*O*-benzyl α-Mangostin (**12**): Yellow fine needles, mp. 109—111 °C; EI-MS $C_{38}H_{38}O_6$, *m/z*: 589.1 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.50 (1H, s, 1-OH), 7.40 (10H, m, Ar-H), 6.72 (1H, s, H-5), 6.31 (1H, s, H-4), 5.26 (2H, t, *J*=6.6 Hz, H-12 and H-17), 5.15 and 5.12 (each 2H, s, H_2-21 and H_2-28), 4.13 (2H, d, *J*=6.9 Hz, H_2-16), 3.80 (3H, s, 7-OMe), 3.34 (2H, d, *J*=6.6 Hz, H_2-11), 1.85, 1.70, 1.68 and 1.67 (each 3H, s, H_3-14, H_3-15, H_3-19 and H_3-20); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 182.0 (C, C-9), 162.4 (C, C-3), 159.9 (C, C-1), 157.0 (C, C-6), 155.1 (C, C-10a), 155.0 (C, C-4a), 143.3 (C, C-7), 137.3 (C, C-8), 136.3 and 135.8 (C, C-22 and C-29), 131.5 and 131.7 (C, C-13 and C-18), 128.7, 128.6, 128.3, 127.4 and 127.2 (each CH_2, C-23, C-24, C-25, C-26, C-27, C-30, C-31, C-32, C-33 and C-34), 123.3 and 122.4 (CH, C-12 and C-17), 112.2 (C, C-8a), 111.8 (C, C-2), 104.1 (C, C-9a), 99.4 (CH, C-5), 89.8 (CH, C-4), 70.6 and 70.2 (CH_2, C-21

and C-28), 60.9 (CH_3, 7-OMe), 26.2 (CH_2, C-16), 21.6 (CH_2, C-11), 25.9 and 25.8 (CH_3, C-15 and C-20), 18.2 and 17.9 (CH_3, C-14 and C-19).

3,6-Di-*O*-4-methylbenzyl α-Mangostin (**13**): Yellow fine needles, mp 98—100 °C; EI-MS $C_{40}H_{42}O_6$, *m/z*: 617.1 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.50 (1H, s, 1-OH), 7.32 and 7.22 (8H, m, Ar-H), 6.74 (1H, s, H-5), 6.32 (1H, s, H-4), 5.26 (2H, t, J=6.6 Hz, H-12 and H-17), 5.13 and 5.09 (each 2H, s, H_2-21 and 22), 4.13 (2H, d, J=6.9 Hz, H_2-16), 3.81 (3H, s, 7-OMe), 3.39 (2H, d, J=6.6 Hz, H_2-11), 2.37 and 2.36 (each 3H, s, H_3-25 and H_3-32), 1.85, 1.71, 1.68 and 1.67 (each 3H, s, H_3-14, H_3-15, H_3-19 and H_3-20); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 182.0 (C, C-9), 162.4 (C, C-3), 159.9 (C, C-1), 157.0 (C, C-6), 155.1 (C, C-10a), 155.0 (C, C-4a), 144.2 (C, C-7), 138.2 and 137.8 (C, C-22 and C-29), 137.3 (C, C-8), 131.5 and 131.7 (C, C-18 and C-13), 133.3, 132.7 (C, C-25 and C-32), 129.4, 129.3, 127.4 and 127.3 (each CH×2, C-23, C-24, C-26, C-27, C-30, C-31, C-33, C-34), 123.3 and 122.4 (CH, C-12 and C-17), 112.2 (C, C-8a), 111.8 (C, C-2), 104.1 (C, C-9a), 99.4 (CH, C-5), 89.8 (CH, C-4), 70.6 and 70.2 (CH_2, C-21 and C-28), 60.9 (CH_3, 7-OMe), 26.2 (CH_2, C-16), 25.9 and 25.8 (CH_3, C-15 and C-20), 22.7 (CH_2, C-11), 21.5 and 21.2 (CH_3, 25-CH_3 and 32-CH_3), 18.2 and 17.9 (CH_3, C-14 and C-19).

3,6-Di-*O*-*n*-pentyl α-Mangostin (**14**): Yellow fine needles, mp 135—138 °C; EI-MS $C_{34}H_{46}O_6$, *m/z*: 549.3 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.49 (1H, s, 1-OH), 6.54 (1H, s, H-5), 6.24 (1H, s, H-4), 5.25 (2H, t, J=6.6 Hz, H-12 and H-17), 4.12 (2H, d, J=6.6 Hz, H_2-16), 4.02 (4H, m, H_2-21and H_2-26), 3.80 (3H, s, 7-OMe), 3.35 (2H, d, J=6.6 Hz, H_2-11), 1.92 (4H, m, H_2-22 and H_2-27), 1.85, 1.80 (each 3H, s) and 1.68 (6H, s), H_3-14, H_3-15, H_3-19 and H_3-20, 1.50 and 1.41 (each 4H, m, H_2-23, H_2-24, H_2-28 and H_2-29), and 0.97 (6H, m, H_3-25 and H_3-30); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 182.0 (C, C-9), 162.9 (C, C-3), 159.8 (C, C-1), 157.5 (C, C-6), 155.3 (C, C-10a), 155.1 (C, C-4a), 144.0 (C, C-7), 137.0 (C, C-8), 131.5 and 131.2 (C, C-13 and C-18), 123.4 and 122.6 (CH, C-12 and C-17), 111.8 (C, C-8a), 111.4 (C, C-2), 103.8 (C, C-9a), 98.7 (C, C-5), 89.2 (C, C-4), 60.8 (CH_3, 7-OMe), 68.8 and 68.4 (CH_2, C-21 and C-26), 26.2 (CH_2, C-16), 25.9 and 25.8 (CH_3, C-15 and C-20), 28.9, 28.7, 28.3, 28.2, 22.5 and 22.4 (CH_2, C-22, C-23, C-24, C-27, C-28 and C-29), 22.7 (CH_2, C-11), 18.2 and 17.8 (CH_3, C-14 and C-19), 14.1 and 14.0 (CH_3, C-25 and C-30).

3,6-Di-*O*-*n*-butyl α-Mangostin (**15**): Yellow fine needles; mp 129—132 °C; EI-MS $C_{32}H_{42}O_6$, *m/z*: 521.3 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.49 (1H, s, 1-OH), 6.68 (1H, s, H-5), 6.26 (1H, s, H-4), 5.25 (2H, t, J=6.6 Hz, H-12 and H-17), 4.12 (2H, d, J=6.6 Hz, H_2-16), 4.04 (4H, m, H_2-21 and H_2-25), 3.80 (3H, s, 7-OMe), 3.35 (2H, d, J=6.6 Hz, H_2-11), 1.85, 1.79 (each 3H, s) and 1.68 (6H, s, CH_3), H_3-14, H_3-15, H_3-19 and H_3-20, 1.90 (4H, m, H_2-22 and H_2-26), 1.54 (4H, m, H_2-23 and H_2-27), 1.01 (6H, m, H_3-24 and H_3-28); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 182.0 (C, C-9), 162.9 (C, C-3), 159.8 (C, C-1), 157.5 (C, C-6), 155.3 (C, C-10a), 155.1 (C, C-4a), 144.0 (C, C-7), 137.0 (C, C-8), 131.6 and 131.3 (C, C-13 and C-18), 123.4 and 122.6 (CH, C-12 and C-17), 111.8 (C, C-8a), 111.4 (C, C-2), 103.8 (C, C-9a), 98.6 (C, C-5), 89.2 (C, C-4), 60.8 (CH_3, 7-OMe), 68.5 and 68.1 (CH_2, C-21 and C-25), 26.2 (CH_2, C-16), 22.5 (CH_2, C-11), 25.9 and 25.8 (CH_3, C-14 and C-19), 18.2 and 17.8 (CH_3, C-15 and C-20), 31.2, 31.0, 19.3×2 (CH_2, C-22, C-26, C-23, C-27), 14.1 and 14.0 (C_3, C-24 and C-28).

3,6-Di-*O*-methyl α-Mangostin (**16**): Yellow fine needles, mp 119—122 °C; EI-MS $C_{26}H_{30}O_6$, *m/z*: 437.1 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.47 (1H, s, 1-OH), 6.71 (1H, s, H-5), 6.29 (1H, s, H-4), 5.23 (2H, t, J=6.6 Hz, H-12 and H-17), 4.13 (2H, d, J=6.6 Hz, H_2-16), 3.95, 3.90 and 3.80 (each 3H, s, 3-OMe, 6-OMe and 7-OMe), 3.34 (2H, d, J=6.6 Hz, H_2-11), 1.85, 1.84, 1.80 and 1.68 (each 3H, s, H_3-14, H_3-15, H_3-19 and H_3-20); ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 181.2 (C, C-9), 162.1 (C, C-3), 158.6 (C, C-1), 157.0 (C, C-6), 156.6 (C, C-10a), 154.3 (C, C-4a), 142.9 (C, C-7), 137.2 (C, C-8), 131.4 and 131.2 (C, C-13 and C-18), 123.8 and 122.8 (CH, C-12 and C-17), 120.5 (C, C-8a), 114.6 (C, C-2), 110.9 (C, C-9a), 97.8 (CH, C-5), 94.2 (CH, C-4), 60.9 (CH_3, 7-OMe), 56.0 and 55.8 (CH_3, 3-OMe and 6-OMe), 26.2 (CH_2, C-16), 22.4 (CH_2, C-11), 25.9 and 25.8 (CH_3, C-15 and C-20), 18.2 and 17.8 (CH_3, C-14 and C-19).

6-*O*-2-Propenyl α-Mangostin (**11**): Yellow gum; EI-MS $C_{27}H_{29}O_6$, *m/z*: 449.1 $[M-H]^-$; ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 13.84 (1H, s, 1-OH), 6.74 (1H, s, H-5), 6.28 (1H, s, H-4), 5.25 (2H, t, J=7.1 Hz, H-12 and H-17), 5.50 (1H, dd, J=17.4, 1.3 Hz, H_E-23), 5.36 (1H, dd, J=10.5, 1.3 Hz, H_Z-23), 5.10 (1H, m, H-22), 4.68 (2H, d, J=5.1 Hz, H_2-21), 4.13 (2H, d, J=7.1 Hz, H_2-16), 3.80 (3H, s, 7-OMe), 3.35 (2H, d, J=6.6 Hz, H_2-11), 1.85 (6H, s), 1.78 (3H, s), and 1.68 (3H, s), H_3-14, H_3-15, H_3-19 and H_3-20; ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 182.0 (C, C-9), 161.5 (C, C-3), 160.6 (C, C-1), 156.0 (C, C-6), 155.3 (C, C-10a), 155.0 (C, C-4a), 144.2 (C, C-7), 137.1 (C, C-8), 135.9 (CH, C-22), 131.6 and 131.4 (C-13 and C-18), 123.4 and 122.5 (C-12 and C-17), 118.5 (CH_2, C-23), 111.8 (C, C-8a), 108.3 (C, C-2), 103.8 (C, C-9a), 99.3 (C, C-5), 93.2 (C, C-4), 69.5 (CH_2, C-21), 60.9 (CH_3, 7-OMe), 26.2 (CH_2, C-16), 21.5 (CH_2, C-11), 25.9 and 25.8 (CH_3, C-15 and C-20), 18.2 and 17.9 (CH_3, C-14 and C-19).

Cytotoxicity Assay The cells for cytotoxicity assays, MCF-7 (human mammary cancer cells) was obtained from Division of Cancer Treatment and Diagnosis (NCI, Maryland, U.S.A.) and DLD-1 (human colonic cells) from ATCC (American Type Culture Collections). The *in vitro* cytotoxicity assay was carried out according to the procedures described previously using SRB[18] with 0.01% campothecin or 60 μM resveratrol as positive control. The absorbance at 492 and 620 nm (reference wavelength) was recorded using Synergy HT microplate reader (Bio-Tek). All mangostin derivatives (**10**—**17**) were screened at 110 μM concentration by comparison with 22 μM of **1**, **2** and **3** on MTT assay[19] using 60 μM resveratrol as positive control. For the MTT assay, the absorbance was recorded at 570 and 620 nm (reference wavelength) using Synergy HT microplate reader (Bio-Tek).

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