Pertanika J. Sci. & Technol. 22 (1): 163 - 174 (2014)

SCIENCE & TECHNOLOGY

Journal homepage: http://www.pertanika.upm.edu.my/

Chemical Constituents of *Aglaia lanuginose*

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ABSTRACT

Our continuing research on the *Aglaia* genus (family Meliaceace) has led us to this first study on the chemical constituents of *Aglaia lanuginose* (bark). The dichloromethane extract from the bark of *Aglaia lanuginose* showed cytotoxicity against HL-60 leukaemia cell line (45% inhibition) at 20 µg/ml and was prioritised for further investigation. Repeated chromatography of the dichloromethane extract yielded the known dammarane triterpenes which were identified as cabralealactone (1), methyl eichlerianate (2), cabraleone (3), ocotillone (4), eichleriatone (5), eichlerianic acid (6) and shoreic acid (7) together with the known sterols, sitosterol (9) and stigmasterol (10). Another isolated compound was the aromatic 4-hydroxycinnamyl-acetate (8), which has not been reported to be present in a plant from the Meliaceae family. The structures of all the compounds were elucidated on the basis of spectroscopic methods (IR, MS and NMR). Cytotoxicity testing of 1-10 showed activity only for mixtures of (3, 4), and (5, 6).

Keywords: Aglaia lanuginose, Aglaia, dammaranes, Meliaceae

INTRODUCTION

Aglaia Lour, the largest genus of the Meliaceae family, consists of approximately 130 species that can be found in the Indo-Malaysian region in South China and on the Pacific Island. It occurs in a variety of habitats ranging from rainforests and mangrove swamps to semi deserts (Muellner *et al.,* 2003). Most of *Aglaia spp*. are trees that can reach up to 40 meters in heights. *Aglaia* spp. are important to the people in the South East Asia region because the trees are useful as sources for timber and their fruit are consumed, sometimes for their medicinal values (Simantujal *et al.,* 1999). In Vietnam, the crude extracts from the leaves and flowers of *Aglaia spp.* are used to treat inflammatory skin disease and allergic disorders such as asthma (Proksh *et al.,* 2005).

Article history:
Received: 15 August 2011
Accepted: 10 September 2012

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ISSN: 0128-7680

Aglaia plants are used to treat fever and as preparations in leprosy treatment by using the latex from the plant to smear on the affected parts of the skin (Christensen *et al.,* 2002). In India, these *Aglaia* plants are used to treat yellow fever, common fever and as an antidote (Kritikar *et al.,* 1975). In addition, the flowers of *Aglaia odorata* are used as perfume in China and Indonesia.

During the past few years, the genus *Aglaia* has been receiving increased focus in scientific research due to its potential in producing biologically active compounds. The most common compounds from various *Aglaia* species that have been reported so far are triterpenes and benzofurans, and many of these compounds possess biological activities. Dammaranes, cycloartanes, tirucallanes, apotirucallanes, glabretals, bacharanes and lupanes represent the most widespread triterpenes within the genus of *Aglaia* (Silvia *et al.,* 2008). As for the benzofuran compounds, studies have shown that those with the cyclopentabenzofuran core are phytochemically confined to the genus *Aglaia* (Brader *et al.,* 1998). These cyclopentabenzofurans are also commonly known as recoglate or flavagline derivatives and have been observed to be biologically active including cytotoxic activity against human cancer cells. An example of a cyclopentabenzofuran is silvestrol; isolated from *Aglaia leptantha* Miq. and collected in Borneo Island, it is currently being developed as an anti-cancer drug for chronic lymphocytic leukaemia (Lucas *et al.,* 2009).

Previous studies have indicated that many *Aglaia* plants contain toxic chemical compounds that can be used as pesticides or anticancer drugs (Simantujak *et al.,* 1999). As part of our continuing research on the *Aglaia* genus, we observed that the dichloromethane extract from the bark of *Aglaia lanuginose* showed moderate in vitro cytotoxic activity against HL60 cell line. As *Aglaia lanuginose* has not been studied for its phytochemical constituents and some of the constituents may exhibit cytotoxicity, here we describe our findings on the chemical constituents from the bark extract of *A. lanuginose* and the cytotoxicity of these compounds against HL60 cells.

MATERIALS AND METHODS

Plant Material

The bark of *Aglaia lanuginose* King was collected in Peninsular Malaysia near the town of Jeli, Kelantan, which is located 127 km along the Jeli-Gerik route from Kota Bharu to Ipoh. A voucher specimen (KL 4232) was deposited at the Herbarium of Department of Chemistry, University Malaya, Kuala Lumpur, Malaysia.

Experimental Design

Fractionation and purification. The dichloromethane crude extract (10 g) which was cytotoxic against HL60 leukaemia cells (45% inhibition) at 20 μg/ml, was subjected to column chromatography (CC) over silica gel and eluted with dichloromethane/ methanol with 1% increments of methanol to give a total of 11 fractions.

Fraction 3 (175.9 mg), which was eluted from the previous CC (SiO_2) with 100% dichloromethane, was further purified by CC (SiO_2) using increasing amounts of ethyl acetate in hexane to give compounds 1, 2, 8, 9 and 10 (all eluted with 5% ethyl acetate in hexane). Fraction

4 (543 mg), which was eluted from the first column with 2% methanol in dichloromethane, was further purified by CC (SiO_2) before it was eluted using increasing amounts of ethyl acetate in hexane to obtain compounds 3 and 4. Both compounds 3 and 4 were eluted with 10% ethyl acetate in hexane. Fraction 5 (800 mg), which was eluted from the first column with 2% methanol in dichloromethane, was further purified by CC (SiO_2) and then eluted using increasing amounts of ethyl acetate in hexane to obtain compound 5, which was eluted at 15% ethyl acetate in hexane. Fraction 6 (3.7 g), which was eluted from the first column with 2% methanol in dichloromethane, was further purified by CC (SiO_2) before it was eluted using increasing amounts of acetone in hexane to obtain compounds 6 and 7.

Compounds 1, 2, 5, 8 were further purified with prep-thin layer chromatography (TLC) using a mixture of hexane and ethyl acetate or a mixture of hexane and acetone solvent systems depending on the polarity of the compounds, while compounds 6 and 7 were obtained in pure form by recrystalization from methanol. Compounds 9 and 10 were identified as the common sterols and were not purified further. Structure elucidation was carried out by ^{1}H-NMR, ^{13}C-NMR, COSY, HMQC, HMBC, DEPT, IR, UV and Mass-spectroscopy.

Cytotoxicity Assay. HL60 cells were acquired from ATCC (American Type Cell Culture) and were cultured in RPMI 1640 (Gibco) media added with 10% of fetal bovine serum (FBS, Gibco). Briefly, test compounds or extracts were prepared as stock solutions of 20 mg/ml in dimethyl sulfoxide (DMSO) and diluted accordingly in phenol red free culture medium supplemented with 5% FBS before use. In a 96-well plate, samples in 4 replicates (n = 4) at a concentration of 5 µg/ml were tested against 15,000 of HL60 cells per well for 2 hours at 37°C in 5% CO_2. The viability of the cells was indirectly determined by MTT dye reduction, where 15 µl of MTT (5 mg/ml in PBS) was added into each well which contained the cells. After incubating for 4 hours, 70 µl of the supernatant was pipetted out from each well. The formazan crystals formed were dissolved by the addition of 100 µl of DMSO. The absorbance was recorded at 570 nm and the viability of the cells was calculated.

RESULTS AND DISCUSSION

Dried ground barks (2.5 kg) were extracted sequentially with hexane, dichloromethane and methanol at room temperature. The hexane, dichloromethane and MeOH extracts were evaporated in vacuum to yield 58.57 g of hexane extract, 20.20 g of dichloromethane extract and 47.05 g of methanol extract separately. Cytotoxicity assay on HL60 using 20 µg/mL of the three extracts showed that the dichloromethane extract had moderate activity (45% inhibition) while the other two extracts were inactive. The dichloromethane extract was selected for further fractionation as part of our focus on isolating chemical constituents that may have anti-cancer properties.

Isolation and Structural Elucidation

Cabralealactone (1) was isolated as a white amorphous powder. The MS spectrum of compound 1 showed a $[M]^+$ peak at *m/z* 414 which corresponded to the molecular formula $C_{27}H_{42}O_3$. The triterpenoidal nature of compound 1 was revealed by ^{1}H-NMR (Table 1) which showed six methyl groups and some complex multiplets belonging to the alicyclic protons. The ^{13}C-NMR

showed 27 peaks assignable to six methyl (CH_3), ten methylene (CH_2), four methine (CH) groups and seven quaternary peaks which were distinguishable from the DEPT spectra. The ^{13}C-NMR spectrum showed additional features of tertiary carbon resonances at δ_C 89.9 and δ_C 176.9, assignable to the lactone system. The ^{1}H-NMR and MS data of compound 1 were similar to those of cabralealactone in the literature (Cascon *et al.,* 1972). On the basis of the evidence presented, compound 1 was identified as cabralealactone.

Methyl isoeiclerianate (2) was isolated as a white amorphous powder. The MS spectrum of compound **2** showed a $[M+H]^+$ peak at *m/z* 489 which corresponded to the molecular formula $C_{31}H_{52}O_4$. The ^{1}H-NMR spectrum (Table 1) showed seven tertiary methyls, an oxomethine at δ_H 3.75 (*t*) and multiplet signals belonging to the alicylic protons. The open seco skeleton was confirmed by the presence of two broad singlets at δ_H 4.66 and 4.84 (2 *brs*, 1H each), which corresponded to the two terminal protons (H-28α and H-28β). Furthermore, a methoxy group located at C-3 proved that compound 2 is a methyl ester. The EIMS of 2 showed a characteristic fragment ion at *m/z* 143 (100%), consistent with the presence of a hydroxy-methyl tetrahydrofuran side chain $[C_8H_{15}O_2]^+$ in the molecule. The configuration of compound 2 was determined to be 24*S* based on the resonance of C-24 at δ_C 86.4 in ^{13}C NMR. The assignments of carbons in the ^{13}C-NMR of compound 2 were similar to those of methyl isoeichlerianate in the literature (Seger *et al.,* 2008). From these observations, compound 2 was identified as methyl isoeiclerianate.

Cabraleone (3) was isolated as a white amorphous powder. The EIMS spectrum of compound 3 showed a $[M + H]^+$ peak at *m/z* 459 which corresponded to the molecular formula $C_{30}H_{50}O_3$. The triterpenoidal nature of compound 3 was revealed in the ^{1}H-NMR spectrum (Table 1) with eight tertiary methyl groups as sharp singlets, multiplet signals of the alicyclic protons and an oxomethine signal at δ_H 3.64 (*t*). The EIMS of 3 showed a characteristic fragment ion at *m/z* 143 (100%) which was similar to compound 2 and consistent with the presence of a hydroxy-methyl tetrahydrofuran side chain $[C_8H_{15}O_2]^+$ in the molecule. The configuration of compound **3** was determined to be 24*S* based on the resonance of C-24 at δ_C 86.4 in ^{13}C NMR. The NMR and MS data of compound 3 were similar to those of cabraleone in the literature (Albersberg *et al.,* 1991). On the basis of the evidence above, the structure of compound 3 was assigned as cabraleone.

Ocotillone (4) was isolated as a white amorphous powder. the MS spectrum of compound 4 showed a $[M + H]^+$ peak at *m/z* 459 - similar to compound 3 - which corresponded to the molecular formula $C_{30}H_{50}O_3$. Signals of ^{1}H- (Table 1) and ^{13}C-NMR spectra of compound 4 were similar to those of its isomer 3, and both compounds can be distinguished by the chemical shifts of C-24. The configuration of compound 4 was determined to be 24*R* due to the resonance of C-24 at δ_C 84.5 in ^{13}C NMR. The other proton assignments in the ^{1}H-NMR of compound 4 were similar to those of ocotillone in the literature (Nuanyai *et al.* 2011). From these observations, compound 4 was identified as ocotillone.

Eichlerialactone (5) was isolated as a white amorphous powder. The MS spectrum of compound 5 showed a $[M + H]^+$ peak at *m/z* 431 which corresponded to the molecular formula $C_{27}H_{42}O_4$. The ^{1}H-NMR spectrum (Table 1) showed five tertiary methyls and multiplet signals belonging to the alicylic protons. In addition, the ^{1}H-NMR spectrum revealed signals of two terminal protons (H-25α and H-25β) as broad singlets at δ_H 4.67 and 4.88 (2 *brs*, each 1 H)

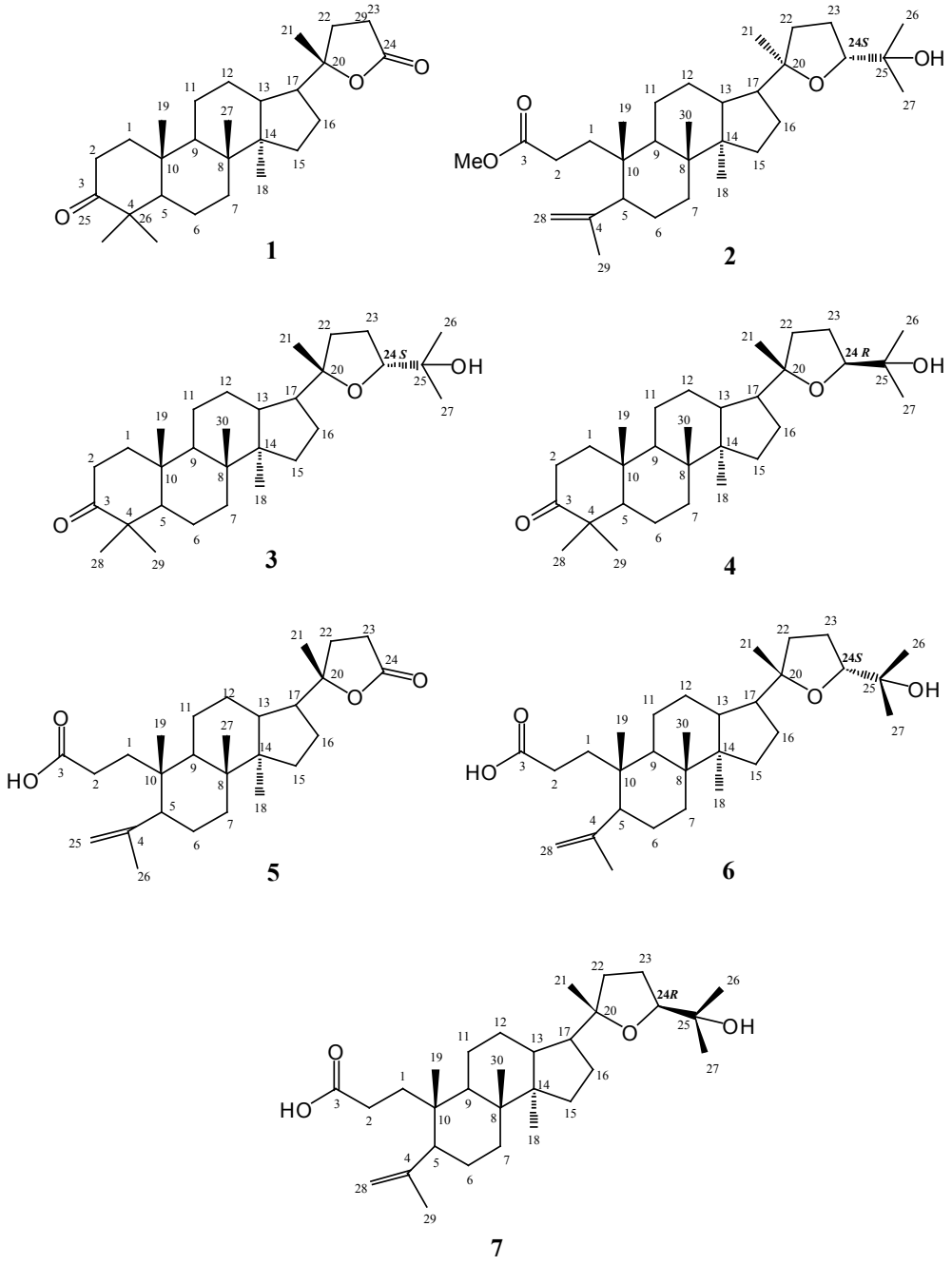

Fig.1: Structures of dammarane triterpenes isolated from *Aglaia lanuginose* King (bark)

which are typical of a seco-dammarane triterpenoid. The ^{13}C-NMR spectrum analyzed by the aid of DEPT spectrum indicated five tertiary methyls, seven quaternary carbons and three oxygen bearing carbons at δ_C 176.9, 89.9, 177.0 for C-3, C-20 and C-24, respectively. The NMR and MS data of compound 5 were similar to those of eiclerialactone in the literature (Singh *et al.,* 1991). On the basis of the evidence above, the structure of 5 was identified as eichlerialactone.

Eichlerianic acid (6) was isolated as a white powder. The MS spectrum of compound 6 showed a molecular ion peak $[M]^+$ at *m/z* 474 corresponding to the molecular formula of $C_{30}H_{50}O_4$. The triterpenoid nature of compound 6 was revealed in the ^{1}H-NMR spectrum (Table 1) by having seven tertiary methyl groups as sharp singlets and some complex multiplet signals belonging to the methylene and methine protons. Additionally, there was a signal assignable to a methine proton H-24 at δ_H 3.57 (*dd*, *J* = 10.3 and 5.0 Hz, 1H) and broad singlets at δ_H 4.60 and 4.78 (2 *brs*, each 1H) which were assignable to the terminal protons H-28α and H-28β. The configuration of compound 6 was determined to be 24*S* based on the resonance of C-24 at δ_C 86.3 in ^{13}C NMR. The other proton assignments in the ^{1}H-NMR and carbon in the ^{13}C-NMR of compound 6 were similar to those of eiclerianic acid in the literature (Roux *et al.,* 1998). On the basis of the evidence above the structure was assigned to be eichlerianic acid.

Shoreic acid (7**)** was isolated as a white amorphous powder. The MS spectrum of compound (7) showed a molecular ion peak $[M]^+$ at *m/z* 474 corresponding to the molecular formula of $C_{30}H_{50}O_4$. The triterpenoid nature of compound 7 was revealed in the ^{1}H-NMR spectrum (Table 1) by having seven tertiary methyl groups as sharp singlets, and some complex multiplet signals belonging to the alicyclic protons. Similarly, with respect to compounds 5 and 6, two doublets appeared as two broad singlets in the ^{1}H-NMR spectrum at δ_H 4.60 and 4.78 (2 *brs*, each 1H), which corresponded to the two terminal protons H-28α and H-28β. The configuration of compound 7 was determined to be 24*S* based on the resonance of C-24 at δ_C 84.4 in ^{13}C NMR. The other assignments in the ^{1}H-NMR and the ^{13}C-NMR of compound **7** were similar to those of shoreic acid in the literature (Roux *et al.* 1998). On the basis of the evidence presented, compound 7 was identified as shoreic acid.

4-hydroxycinnamyl acetate (8**)** was isolated as yellow oil. The MS spectrum of compound (8) showed a molecular ion $[M]^+$ peak at *m/z* 192 which was in agreement with the molecular formula $C_{11}H_{12}O_3$. The ^{1}H-NMR spectrum (Table 2) showed the presence of a singlet peak at δ_H 2.28 assignable to C(11) methyl. The ^{1}H-NMR spectra of compound **8** showed signals of H-9 methylene at δ_H 4.30 (*d*, *J* = 6.1 Hz, 2H), a H-8 *trans* olefin pair at δ_H 6.29 (*dt*, *J* = 6.1, 16.1 Hz, 1H) for H-8 and a H-7 proton at δ_H 6.57 (*d*, *J* = 16.1 Hz, 1H). There were also proton signals corresponding to a *para*-substituted aromatic ring with characteristic chemical shifts for [H-3, H-5] and [H-2, H-6] at δ_H 7.02 and 7.36 respectively (*d*, *J* = 8.6 Hz, 2H each). The assignments of protons in the ^{1}H-NMR of compound **8** were similar to those of 4- hydroxycinnamic acetate in the literature (Kuichi *et al.,* 2002). On the basis of the data presented above, structure **8** was identified to be 4-hydroxycinnamyl acetate.

The MS spectrum of mixtures of sterols (9, 10) showed a molecular ion $[M]^+$ peaks at *m/z* 414 and 412, respectively. The identity of compounds 9 and 10 was established by direct comparison of ^{13}C-NMR and ^{1}H-NMR to the data published in literature (Eknmakul *et al.,* 2003).

TABLE 1a
^{1}H-NMR data of triterpene compounds 1, 2, 3, 4, 5, 6 and 7 in $CDCl_3$

Position	1	2	3	4	5	6	7
1	1.84 (m)	1.75 (m), 1.53 (m)	1.45 α (m), 1.72 β (m)	1.46 α (m), 1.94 β (m)	1.24 (m), 1.6 (m)	1.75 (m), 1.53 (m)	1.75 (m), 1.53 (m)
2	2.5 (m)	2.14 (m) 2.32 (m)	2.46 α (m) 2.54 β (m)	2.42 α (m) 2.50 β (m)	1.50 β (m), 1.18 α (m)	2.14 (m), 2.32 (m)	2.14 (m), 2.32 (m)
3	-	-	-	-	-	-	-
4	-	-	-	-	-	-	-
5	-	-	1.38 (m)	1.38 (m)	1.98 (m)	-	-
6	-	-	1.45 α (m) 1.55 β (m)	1.40 α (m) 1.55 β (m)	1.38	-	-
7	1.49 (m)	-	1.3 α (m) 1.58 β (m)	1.66 α (m) 1.70 β (m)	1.54β (m), 1.24α (m)	1.15 (m)	-
8	-	-	-	-	-	-	-
9	1.31	-	1.88 (m)	1.80 (m)	1.50 (m)	1.43 (m)	1.15 (m)
10		-	-	-		-	
11	-	-	1.28 α (m) 1.50 β (m)	1.25 α (m) 1.50 β (m)	1.30 β (m), 1.44 α (m)	1.35 (m)	1.43 (m)
12	1.56 (m), 1.21 (m)	-	1.25 (m)	2.00 (m)	1.90 β (m), 1.28 α (m)	-	-
13	1.52 (m)	-	1.58 (m)	1.70 (m)	1.68 (m)	1.60 (m)	1.35 (m)
14	1.31 (m)	-	-	-	-	-	-
15	-	-	1.1α (m) 1.48 β (m)	1.1α (m) 1.48β (m)	1.50 β (m), 1.24 α (m)	1.40 (m)	1.60 (m)
16	-	-	1.32 (m)	1.32	1.82 β (m), 1.32 α (m)	1.75 (m)	1.40 (m)
17	1.85 (m)	-	1.44 (m)	1.44 (m)	1.94 (m)	1.80 (m)	1.75 (m)
18	0.83 (s)	0.88 (s)	0.89 (m)	0.88 (s)	0.86 (s)	0.82 (s)	0.79 (s)
19	0.92 (s)	0.84 (s)	0.95 (m)	0.94 (s)	0.90 (s)	0.79 (s)	0.82 (s)
20	-	-	-	-	-	-	-
21	1.32 (s)	1.14 (s)	1.11(m)	1.11(s)	1.34(s)	1.08 (s)	1.81 (s)
22	2.00 (m)	-	1.7 α (m) 1.85 β (m)	1.70 α (m) 1.85 β (m)	2.16 β (m), 1.94 α (m)	1.57 (m)	1.57 (m)
23	1.85 (m)	-	1.3 α (m) 1.75 β (m)	1.80 (m)	2.56 (m)	1.80 (m)	1.80 (m)
24	-	3.75 (t)	3.64 (t)	3.76 (t)		3.57 (dd, J = 10.3, 5.0 Hz))	3.73 (t)
25	1.06 (s)	-	-	-	4.67 (brs), 4.88 (brs)	-	-
26	1.01 (s)	1.18	1.19 (s)	1.15 (s)	1.76 (s)	1.13 (s)	1.13 (s)
27	0.98 (s)	1.11	1.20 (s)	1.20 (s)	1.0 (s)	1.05 (s)	1.05 (s)

TABLE 1a (*continue*)

Position	1	2	3	4	5	6	7
28	-	4.66 (brs), 4.84 (brs)	1.08 (s)	1.08 (s)	-	4.60 (brs), 4.78 (brs)	4.60 (brs), 4.78 (brs)
29	-	1.76 (s)	1.04 (s)	1.04 (s)	-	1.67 (s)	1.67 (s)
30	-	0.99 (s)	1.01(s)	1.00 (s)	-	0.99 (s)	0.99 (s)
31	-	3.66 (s)	-	-	-	-	-

The protons at H-24 of compounds 2, 3, 4 and 7 are pseudo-*dd* which appear as *t*, with approximate $J = 5$ Hz.

TABLE 1b

^{13}C-NMR data of triterpene compounds **1**, **2**, **3**, **4**, **5**, **6** and **7 in $CDCl_3$**

Position	1	2	3	4	5	6	7
1	39.8	34.4	39.9	39.9	34.4	34.3	34.3
2	34.5	28.3	34.1	34.1	28.4	28.5	28.5
3	218.0	174.6	218.1	218.1	176.9	179.5	179.4
4	47.4	147.5	47.4	47.4	147.4	147.5	147.4
5	55.3	50.7	55.4	55.3	50.8	50.8	55.3
6	19.6	24.6	19.7	19.7	24.6	24.6	19.7
7	34.1	33.9	34.8	37.6	33.9	33.9	37.6
8	42.7	40.0	40.3	40.3	40.1	40.0	40.3
9	50.0	41.0	49.8	50.1	41.1	41.2	50.1
10	36.8	37.5	36.9	36.9	39.1	39.0	39.9
11	21.8	21.7	22.3	22.0	21.8	22.3	22.0
12	25.0	26.9	27.0	27.0	26.4	26.9	27.0
13	43.0	42.9	43.0	43.0	42.7	42.9	43.0
14	49.4	50.6	50.0	49.9	50.4	50.3	49.9
15	31.0	31.2	31.4	31.2	31.1	31.4	31.2
16	28.6	25.8	25.8	25.9	25.0	25.8	25.9
17	49.4	50.0	50.2	50.1	49.5	49.7	49.7
18	16.1	16.0	16.3	16.1	16.1	16.3	16.0
19	15.2	20.1	16.1	16.1	20.1	20.2	20.1
20	89.9	86.0	86.5	86.5	89.9	86.6	86.5
21	22.1	21.9	27.2	26.8	22.5	27.1	24.2
22	33.2	37.5	34.6	34.6	33.0	34.7	34.7
23	26.4	29.6	26.4	25.8	28.7	26.3	26.3
24	176.9	86.4	86.4	84.5	177.0	86.3	84.4
25	26.7	71.1	70.3	71.1	147.4	70.3	76.7
26	21.0	27.6	24.1	21.7	23.2	27.8	27.5
27	16.0	24.3	27.8	27.7	15.3	23.2	21.7
28		113.3	26.4	24.3		113.4	113.4
29		23.2	21.0	21.0		24.0	23.2
30		15.3	15.2	15.2		15.3	15.3
31		51.6					

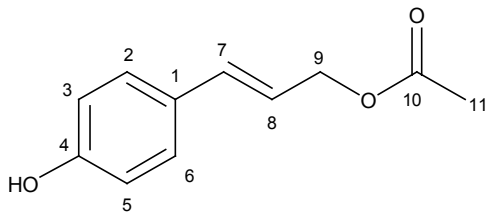

Fig.2: Structure of compound 8

TABLE 2
1D and 2D NMR of compound 8 in $CDCl_3$

No	δ_C (ppm)	δ_H (ppm)	HMBC (H-C)
1	134.5	-	-
2	121.7	7.36 (*d*, 8.6 *Hz*)	1
3	127.4	7.02 (*d*, 8.6 *Hz*)	1,4,5
4	150.6	-	-
5	127.4	7.02 (*d*, 8.6 *Hz*)	1,3,4
6	121.7	7.36 (*d*, 8.6 *Hz*)	2,4,8
7	130.1	6.57 (*d*, 16.1 *Hz*)	2,6
8	128.7	6.29 (*dt*, 6.1, 16.1 *Hz*)	1
9	63.6	4.30 (*d*, 6.1 *Hz*)	7,8
10	169.6	-	-
11	21.1	2.28 (*s*)	10

In this study, the compounds isolated from the bark of *Aglaia lanuginose* were the dammarane triterpenoids identified as cabralealactone 1, methyl isoeichlerianate 2, cabraleone 3, ocotillone 4, eihlerialactone 5, eichlerianic acid 6, shoreic acid 7 and two sterols identified as sitosterol 9 and sigmasterol 10. Compounds 2, 6 and 7 may be further classified as 3,4-seco-dammarane triterpene. In addition, we isolated one aromatic compound, 4-hydroxycinnamyl-acetate 8, which previously has only been isolated from *Alpinia galaga* (Zingeberaceae) (Eknmakul *et al.,* 2003). To the best of our knowledge 4-hydroxycinnamyl-acetate has never been isolated from any *Aglaia* species.

All the damarrane triterpenes isolated in this study (1-7) have similar structures, by having the 20, 24-epoxy ring. All of the compounds with *20R* configuration and isolated from *Aglaia lanuginose*, were first reported as isolates from *Cabralea eichleriana (C*ascon *et al*., 1972)*, Cabralea polytricha* (Rao *et al*., 1975) and *Dysoxylum richii* (Aalbersberg *et al*., 1991) plants from the Meliaceae family, as well as from many other plants in other families in subsequent studies. For example, compound 1 was also reported in *Betula platyphylla* (Betulaceae) (Byung *et al.,* 1977) and *Cleome africana* (Cleomaceae) (Tsichritzis *et al.,* 1993). In the case of compound 2 which has *20S* configuration, it has only been reported in recent years as a natural product from *Aglaia silvestris* (Pointinger *et al.,* 2008).

Compounds 3 and 4, as well as compounds 6 and 7 were pairs of stereoisomer where they were different in their stereochemistries at position C-24. The stereochemistries of compound (3, 4) and (6, 7) were determined by comparing the ^{13}C chemical shift of C-24 with those in the literature (Hisham *et al.,* 1996), whereby the *R* and *S* stereo-centres at C-24 has distinguishable C-13 chemical shifts of approximately 84.3 and 86.3 ppm respectively. In addition, compound 2 which previously has only been isolated from *Aglaia silvestris* was different from the rest of the dammarane compounds isolated so far due to its uncommon 20*R* stereochemical configuration, instead of the usual 20*S* configuration (Pointinger *et al.,* 2008). The 20*R* stereochemistry of compound 2 was determined by comparing the ^{13}C-NMR chemical shifts of the neighboring C-21 and C-22 with those in the literature (Seger *et al.,* 2008).

We observed that a mixture of compounds 3 and 4, and a mixture of compounds 6 and 7 to have cytotoxicity against HL60 cells at 5 µg/ ml, where the mixture of 3 and 4 (1:1 ratio) showed 62.0 ± 2.7% inhibition while the mixture of 6 and 7 (1:1 ratio) showed 75.0 ± 3.0% inhibition. All the other fractions, including those that contained mixtures of compounds 1, 2, 5 and 8 until 10 were not active at 5 µg/ ml. From the literature, damarrane type triterpenoids such as compounds 1, 3, 4, 5, 6, 7 have been reported to be cytotoxic in various cell lines. Compound 2 has not been reported for its cytotoxic activity while compound 8 was only reported to have trypanocidal activity (Kuichi *et al.,* 2002). A study on phytochemicals from *Aglaia leucophylla*, Abdelilah *et al.* (1994) showed that compound 3 was cytotoxic against KB cell line. In addition, another study by Charles *et al.* (2010) which showed that the mixtures of cabraleone and ocotillone (3, 4), and eichlerianic acid and shoreic acid (6, 7) were cytotoxic towards HeLa cells. With respect to compounds 1 and 5, the disagreement between our results and the published cytotoxicity data in the literature may be due to differences in the cell lines used. Compound **1** was tested to be potent with micromolar IC_{50} value in P388 leukemia cell line (Hidekazu *et al.,* 1997) and a breast cancer cell line (Jarinporn *et al.,* 2008). In the case of compound **5**, it was reported to be moderately active against NCI-H187 small-cell lung cancer cell line and weakly active towards a breast cancer cell line (Jarinporn *et al.,* 2008).

CONCLUSION

10 compounds were obtained from the cytotoxic dichloromethane fraction from the bark of *Aglaia lanuginose*. Out of these, the aromatic compound 8, 4-hydroxycinnamyl-acetate has never been isolated from any *Aglaia* species. From the cytotoxicity, we found that some of these compounds are cytotoxic and may have potential as cytotoxic agents for cancer.

ACKNOWLEDGEMENTS

The authors are grateful to the Malaysian IRPA grant for its financial support.

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