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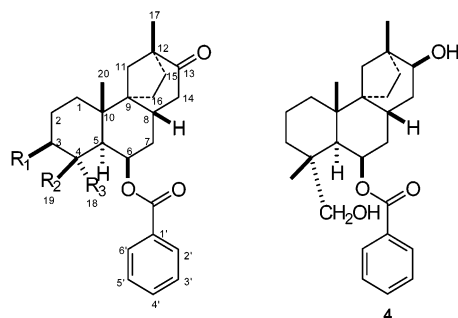
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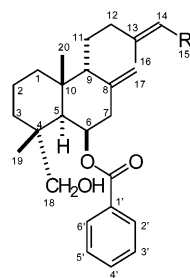
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Four new labdane-derived diterpenes, *iso*-dulcinol (**1**), 4-*epi*-scopadulcic acid B (**2**), dulcidiol (**4**), and scopanolal (**5**), together with two known diterpenes, dulcinol/scopadulciol (**3**) and scopadiol (**6**), were isolated from the aerial parts of *Scoparia dulcis*. The structures were determined by extensive NMR studies. The crude extracts as well as the pure diterpenes showed cytotoxicity against a panel of six human stomach cancer cell lines.

*Scoparia dulcis* L. (Scrophulariaceae) is a perennial herb, which grows in tropical and subtropical regions. The roots and leaves of the plant are used as a cure for toothache, blennorrhagia, and stomach troubles.<sup>1</sup> The plant is also used to treat diabetes.<sup>2</sup> Several groups of investigators studied the plant and reported the isolation of diterpenes,<sup>3–6</sup> triterpenes,<sup>7,8</sup> and flavonoids.<sup>9</sup> We have examined the petroleum ether extract of *S. dulcis* and isolated six benzoylated diterpenes (**1**–**6**), four of which were new. The crude extracts and the pure compounds were tested on a panel of six human cancer cell lines. A spectrum of cytotoxicity was indicated by the crude extracts as well as the pure compounds tested.



- 1** R<sub>1</sub> = OH, R<sub>2</sub> = R<sub>3</sub> = CH<sub>3</sub>  
**2** R<sub>1</sub> = H, R<sub>2</sub> = COOH, R<sub>3</sub> = CH<sub>3</sub>  
**3** R<sub>1</sub> = H, R<sub>2</sub> = CH<sub>3</sub>, R<sub>3</sub> = CH<sub>2</sub>OH



- 5** R = CHO  
**6** R = CH<sub>2</sub>OH

**Table 1.** <sup>1</sup>H NMR Spectral Data of Compounds **1**, **2**, **4**, and **5**<sup>a</sup>

proton	<b>1</b>	<b>2</b>	<b>4</b>	<b>5</b>
H-3	3.20 dd (8.9, 6.2)			
H-5	1.20 d (2.2)	1.37 d (1.9)	1.65 d (2.1)	2.06 d (2.3)
H-6	5.74 q (2.6)	5.56 br d (1.9)	5.59 br d (2.2)	5.64 br d (2.3)
H-8	2.47 m	2.43 m	2.33 m	
H-13			3.45 br d (3.4)	
H-14	2.27 dd (16, 6.6) 1.98 dd (16, 12)	2.20 dd (16, 6.0) 1.98 dd (16, 12)	1.25–1.35 m, 2H	5.92 d (8)
H-15				10.01 d (8)
H-16	2.10 br t (9.0) 1.72 m		1.86 m 1.55 m	2.18 s, 3H
H-17	1.10 s, 3H	1.08 s, 3H	1.04 s, 3H	4.77 br s 4.76 br s
H-18	1.11 s, 3H	1.00 s, 3H	3.57 d (10.9) 3.12 d (10.9)	3.60 d (10.9) 3.16 d (10.9)
H-19	0.97 s, 3H		0.93 s, 3H	0.92 s, 3H
H-20	1.51 s, 3H	1.49 s, 3H	1.56 s, 3H	1.48 s, 3H
H-2', 6'	8.05 d (7.6)	8.00 d (7.6)	8.06 d (7.6)	8.03 d (7.6)
H-3', 5'	7.47 t (7.6)	7.41 t (7.6)	7.46 t (7.6)	7.45 t (7.6)
H-4'	7.59 t (7.6)	7.53 t (7.6)	7.57 t (7.6)	7.57 t (7.6)

<sup>a</sup> Spectra recorded at 400 MHz in CDCl<sub>3</sub>. The values are in ppm. Assignments were made on the basis of COSY, NOESY, HCCOBI, and HMBC experiments. *J* values (Hz) in parentheses.

## Results and Discussion

The petroleum ether extract of aerial parts of *S. dulcis* afforded compounds **1**–**6**, 6-methoxybenzoxazolinone,<sup>7</sup> and glutinol.<sup>8</sup> Compounds **3** and **6** were identified as dulcinol (or scopadulciol)<sup>5,6,10</sup> and scopadiol<sup>5</sup> respectively, on the basis of 2D NMR studies and also by comparison of their NMR data with those published in the literature. The other four compounds (**1**, **2**, **4**, **5**) were all new labdane-derived diterpenes and named as *iso*-dulcinol, 4-*epi*-scopadulcic acid B, dulcidiol, and scopanolal, respectively.

The FABMS of compound **1** showed a peak at *m/z* 425, C<sub>27</sub>H<sub>37</sub>O<sub>4</sub>, [M + H]<sup>+</sup>. The <sup>1</sup>H NMR spectrum indicated the presence of four tertiary methyl groups ( $\delta$  0.97, 1.10, 1.11, 1.51), two of which are geminal, two oxymethine protons ( $\delta$  3.20 dd, *J* = 8.9, 6.2 Hz, 5.74 q, *J* = 2.6 Hz), and a monosubstituted benzene ring ( $\delta$  7.47, 2H, 7.59, 1H, 8.05, 2H). The <sup>1</sup>H NMR spectrum (Table 1) of this compound was quite similar to that of **3**. The main difference was that compound **1** has four tertiary methyl groups while **3** has three tertiary methyls and one oxymethylene group. These findings indicated that the hydroxymethylene group at C-4 of compound **3** is absent in compound **1**. The *J*-modulated <sup>13</sup>C NMR spectrum (Table 2) showed 27 carbons including two carbonyl carbons at  $\delta$  213.5 and 166.3, and it is also

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**Table 2.**  $^{13}\text{C}$  NMR Spectral Data of **1**, **2**, **4**, and **5**<sup>a</sup>

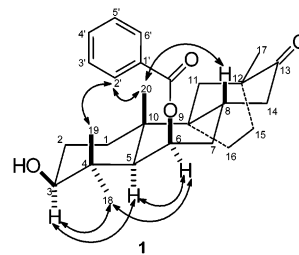
carbon	<b>1</b>	<b>2</b>	<b>4</b>	<b>5</b>
C-1	32.4	34.5	34.4	38.6
C-2	27.5	19.4	18.4	18.7
C-3	79.2	38.9	38.0	38.3
C-4	39.8	42.5	38.7	38.7
C-5	50.4	52.4	43.5	41.5
C-6	70.3	69.1	70.8	71.6
C-7	35.9	34.9	35.6	37.1
C-8	35.8	35.9	29.6	144.0
C-9	53.1	52.9	53.5	57.3
C-10	39.2	39.5	39.0	38.9
C-11	45.6	45.5	38.1	23.9
C-12	52.5	52.5	43.8	39.4
C-13	213.5	213.4	75.1	164.6
C-14	42.7	42.6	36.6	127.5
C-15	36.8	36.8	36.5	191.5
C-16	23.9	23.6	22.6	18.1
C-17	19.9	19.9	24.2	113.9
C-18	28.5	29.6	71.7	71.4
C-19	16.8	181.8	20.7	20.5
C-20	21.4	20.5	22.5	26.1
C=O	166.3	166.0	166.7	166.4
C-1'	130.8	130.6	131.1	131.0
C-2',6'	129.8	129.8	129.9	129.9
C-3',5'	128.8	128.7	128.6	128.7
C-4'	133.3	133.1	133.0	133.1

<sup>a</sup> Spectra recorded at 100 MHz in  $\text{CDCl}_3$ . The values are in ppm. Assignments were made on the basis of COSY, NOESY, HCCOBI, and HMBC experiments.

**Table 3.**  $^1\text{J}$ ,  $^2\text{J}$ , and  $^3\text{J}$  Heteronuclear Interactions through HCCOBI and HMBC Experiments of Compound **1**

proton	$^1\text{J}$ interaction	$^{2/3}\text{J}$ interactions
H-3	79.2 (C-3)	16.8 (C-19), 28.5 (C-18),
H-5	50.4 (C-5)	16.8 (C-19), 21.4 (C-20),
		53.1 (C-9)
H-6	70.3 (C-6)	35.8 (C-8), 39.2 (C-10)
H-8	35.8 (C-8)	23.9 (C-16), 35.9 (C-7),
		42.7 (C-14), 53.1 (C-9)
H-14	42.7 (C-14)	35.8 (C-8), 53.1 (C-9),
		213.5 (C-13)
H-16	23.9 (C-16)	35.8 (C-8), 39.2 (C-10),
		53.1 (C-9)
H-17	19.9 (C-19)	36.8 (C-15), 45.6 (C-11),
		52.5 (C-12), 213.5 (C-13)
H-18	28.5 (C-18)	16.8 (C-19), 39.8 (C-4),
		50.4 (C-5), 79.2 (C-3)
H-19	16.8 (C-19)	28.5 (C-18), 39.8 (C-4),
		50.4 (C-5), 79.2 (C-3)
H-20	21.4 (C-20)	32.4 (C-1), 39.2 (C-10),
		50.4 (C-5), 53.1 (C-9)
H-2',6'	129.8 (C-2',6')	133.3 (C-4'),
		166.3 (C=O)
H-3',5'	128.8 (C-3',5')	130.8 (C-1')
H-4'	133.3 (C-4')	129.8 (C-2',6')

similar to that of **3** except for the presence of an oxymethine carbon at  $\delta$  79.2 (C-3, compound **1**) instead of an oxymethylene carbon at  $\delta$  71.6 (C-18, compound **3**). The position of the OH group was found to be at C-3 by an HMBC experiment (Table 3). The geminal methyls, H-18 and -19, both correlated with C-3, as well as C-4 and C-5. The relative stereochemistry was determined by a NOESY experiment and was found to be similar to that of **3**. In the NOESY experiment (Figure 1), the H-5 protons showed strong interactions with H-3 and H-6, and the latter two protons revealed interaction with the 18-Me, indicating that all these groups were on the same side of the molecule. The  $\beta$ -methyl group at C-20 showed strong interactions with H-8, and H-2' and H-6' of the benzoyl group. A strong interaction was also observed between 19-Me, H-2', and H-6'. Thus the structure of compound **1** was determined to be 3 $\beta$ -hydroxy-6 $\beta$ -benzoyl-12-methyl-9(12)<sub>a</sub>,9(12)<sub>b</sub>-di-

**Figure 1.** Structure of **1** showing NOESY interactions.

homopodocarpane-13-one, and it has been given the trivial name *iso*-dulcinol.

The molecular formula of 4-*epi*-scopadulcic acid **2** was determined as  $\text{C}_{27}\text{H}_{34}\text{O}_5$  from combined analysis of FABMS and  $^{13}\text{C}$  NMR data. The  $^1\text{H}$  and  $^{13}\text{C}$  NMR data of **2** (Tables 1 and 2) were similar to those of **1** except that compound **2** lacked one oxymethine and showed three tertiary methyls ( $\delta$  1.00, 1.08, 1.49) instead of four (none of which were geminal) and a carboxyl carbon at  $\delta$  181.8. The HMBC experiment revealed the COOH group to be at C-4, as the methyl at  $\delta$  1.00 (H-18) and H-5 exhibited  $^3\text{J}$  correlation to the carboxyl carbon at  $\delta$  181.8. The relative stereochemistry was determined by a NOESY experiment and the COOH group was determined to be  $\beta$  (C-19), as the equatorial methyl group at  $\delta$  1.00 (H-18) showed a strong interaction with the  $\alpha$  proton at position 6. Compound **2** is therefore the 4-*epimer* of scopadulcic acid **B**<sup>4,10</sup> and was named 4-*epi*-scopadulcic acid **B**.

Compound **4** was shown by FABMS to have the molecular formula  $\text{C}_{27}\text{H}_{38}\text{O}_4$ , its structure being quite similar to that of **3**, but with two hydrogens more. The  $^1\text{H}$  and  $^{13}\text{C}$  NMR data of **4** (Tables 1 and 2) indicated replacement of the carbonyl group at C-13 by an oxymethine group ( $\delta$  3.45 br d,  $J = 3.4$  Hz). The location of the latter proton was determined as C-13 by an HMBC experiment. The small coupling constant of H-13 determined its orientation to be equatorial, which was further confirmed by a NOESY experiment. Thus the structure of compound **4** was determined, and it was given the name dulcidol.

The HREIMS of compound **5**,  $\text{C}_{27}\text{H}_{36}\text{O}_4$  exhibited peaks at  $m/z$  395  $[\text{M} - \text{CHO}]^+$  and 319  $[\text{M} - \text{COC}_6\text{H}_5]^+$ . The IR spectrum showed two peaks at 1710 and 1668  $\text{cm}^{-1}$ , the latter indicative of an  $\alpha,\beta$ -unsaturated aldehydic group.<sup>11</sup> A  $J$ -modulated  $^{13}\text{C}$  NMR spectrum revealed signals for 27 carbons. The  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR spectra of **5** were very similar to that of scopadiol (**6**), except that it showed an aldehydic group ( $\delta_{\text{H}}$  10.01 d,  $J = 8.0$  Hz;  $\delta_{\text{C}}$  191.5) instead of an oxymethylene group at C-15 ( $\delta_{\text{H}}$  4.17 d,  $J = 7.0$  Hz;  $\delta_{\text{C}}$  59.6). The COSY spectrum revealed coupling between H-14 and the aldehydic proton at C-15. The relative stereochemistry was determined on the basis of NOESY experiments. Thus, **5** was determined to be 6 $\beta$ -benzoyl-9 $\beta$ H-15- $\alpha$ -labda-8(17),13-dien-18-ol, and it was assigned the trivial name scopanolal.

Compounds **1**–**4** and **6** were found to be cytotoxic against one or more of the cancer cell lines tested (Table 4). Compounds **3** and **6** showed cytotoxicity ( $\text{ED}_{50}$  from 8.9 to 37.7  $\mu\text{M}$ ) that was comparable to the positive control against some of the cell lines noted in Table 4. Compounds **1** and **2** showed activity ( $\text{ED}_{50}$  from 19.5 to 59.3  $\mu\text{M}$ ) against the SCL, SCL-37/6, and SCL-9 cell lines. Compound **4** also showed cytotoxicity ( $\text{ED}_{50}$  from 29.0 to 46.0  $\mu\text{M}$ ) except against the NUGC-4 line. Compound **5** was not tested due to its insufficient amount. Glutininol and 6-methoxybenzoxazolinone were noncytotoxic against all of the six cell lines tested.

**Table 4.** Cytotoxicities of Compounds **1–6**, Vinblastine Sulfate, and Mitomycin C (ED<sub>50</sub> in  $\mu\text{M}^a$ )

compound	cancer cell line					
	SCL	SCL-6	SCL-37'6	SCL-9	Kato-3	NUGC-4
<b>1</b>	19.5 $\pm$ 2.4 <sup>mb</sup>	62.9 $\pm$ 2.9	45.8 $\pm$ 3.3	57.9 $\pm$ 6.0	71.7 $\pm$ 7.8	122.9 $\pm$ 12.5
<b>2</b>	37.0 $\pm$ 4.7	136.9 $\pm$ 12.7	59.3 $\pm$ 8.2	48.3 $\pm$ 5.7	124.3 $\pm$ 6.3	109.9 $\pm$ 4.5
<b>3</b>	22.0 $\pm$ 2.1 <sup>mc</sup>	32.8 $\pm$ 3.5	24.4 $\pm$ 3.1 <sup>md</sup>	37.7 $\pm$ 4.8	35.5 $\pm$ 4.0	33.3 $\pm$ 3.6
<b>4</b>	45.3 $\pm$ 3.3	46.0 $\pm$ 2.5	42.6 $\pm$ 5.7	41.6 $\pm$ 1.7	29.0 $\pm$ 2.5	105.1 $\pm$ 8.4
<b>6</b>	22.8 $\pm$ 3.0 <sup>me</sup>	12.2 $\pm$ 2.0	8.9 $\pm$ 1.7	12.2 $\pm$ 2.0 <sup>mf</sup>	9.7 $\pm$ 1.8 <sup>v</sup>	16.6 $\pm$ 2.6 <sup>mg</sup>
vinblastine sulfate	5.9 $\pm$ 0.6	6.1 $\pm$ 0.8	5.3 $\pm$ 0.6	5.3 $\pm$ 0.6	6.1 $\pm$ 0.7	5.3 $\pm$ 0.5
mitomycin C	20.2 $\pm$ 2.0	20.7 $\pm$ 2.2	19.0 $\pm$ 2.1	17.4 $\pm$ 2.0	49.0 $\pm$ 6.0	19.1 $\pm$ 2.6

<sup>a</sup> The ED<sub>50</sub> values of the compounds were significantly higher ( $p < 0.05$ ) relative to those of vinblastine sulfate and mitomycin C, except the <sup>mv</sup> marked values. <sup>mv</sup>ED<sub>50</sub> values for compounds comparable to those for mitomycin C and vinblastine sulfate, respectively, differences of which were statistically insignificant ( $p > 0.05$ ). <sup>mb</sup> $p = 0.847$ , <sup>mc</sup> $p = 0.562$ , <sup>md</sup> $p = 0.257$ , <sup>me</sup> $p = 0.507$ , <sup>mf</sup> $p = 0.137$ , <sup>mg</sup> $p = 0.537$ , <sup>v</sup> $p = 0.137$ .

## Experimental Section

**General Experimental Procedures.** Optical rotations were measured on a Perkin-Elmer 341 polarimeter. IR spectra were recorded on a Mattson Galaxy 5000 FTIR spectrometer. <sup>1</sup>H NMR (400 MHz) and <sup>13</sup>C NMR (100 MHz), <sup>1</sup>H–<sup>1</sup>H COSY, NOESY, HCCOBI, and HMBC spectra were measured on Bruker AMX-400 and DPX-400 instruments. HREIMS were recorded on a JEOL JMS-AX505HA mass spectrometer at 70 eV using the direct insertion probe. FABMS were measured using a matrix of glycerol/nitrobenzoyl alcohol on the same mass spectrometer with xenon as the atom source.

**Plant Material.** The aerial parts of *Scoparia dulcis* were collected in Dhaka in August 1999. A voucher specimen (DACB 28069) has been deposited in the National Herbarium, Dhaka, Bangladesh.

**Extraction and Isolation.** The dried aerial parts of the plant (650 g) were extracted successively in a Soxhlet with petroleum ether (60–80 °C), EtOAc, and MeOH. The extracts were concentrated under vacuum to yield 15, 12, and 37 g of crude residues, respectively. The petroleum ether extract was fractionated by vacuum liquid chromatography using petrol, EtOAc, and MeOH with increasing polarity. A total of 25 fractions (250 mL each) were collected. Fraction 12 (40% EtOAc in petrol) was further fractionated by Sephadex LH-20 column chromatography to give 30 fractions, using CHCl<sub>3</sub> as the eluting solvent. Sephadex fractions 8–10 showed a bright pinkish purple spot with vanillin/H<sub>2</sub>SO<sub>4</sub>; these were combined and then subjected to preparative TLC (silica gel, EtOAc/toluene, 15:85, multiple development) to obtain **2** (12 mg,  $R_f$  0.51, EtOAc/toluene, 40:60). Similarly, VLC fraction 14 (50% EtOAc in petrol) was fractionated on a Sephadex column into 30 fractions. Final purifications were achieved by preparative TLC (EtOAc/toluene, 20:80, multiple development) and yielded **1** (9 mg), **3** (100 mg), **4** (15 mg), and 6-methoxybenzoxazolinone (150 mg) (detection: vanillin/H<sub>2</sub>SO<sub>4</sub> reagent,  $R_f$  **1**, 0.37; **3**, 0.33; **4**, 0.38 in EtOAc/toluene, 40:60). Similarly VLC fraction 17 (80% EtOAc in petrol) afforded **5** (5 mg) and **6** (120 mg) (PTLC EtOAc/toluene, 35:65, multiple development,  $R_f$  **5**, 0.20; **6**, 0.14 in EtOAc/toluene, 40:60). Glutanol (12 mg) was crystallized out when VLC fraction 5 (15% EtOAc in petrol) was fractionated on a Sephadex column using CHCl<sub>3</sub> as the eluting solvent.

**iso-Dulcinol (1):** gum; [ $\alpha$ ]<sub>D</sub><sup>25</sup> –21.4° ( $c$  0.45, CHCl<sub>3</sub>); IR (film)  $\nu_{\text{max}}$  3479, 2945, 2866, 1710, 1699, 1450, 1279, 1111, 1026, 712 cm<sup>–1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Tables 1 and 2; FABMS  $m/z$  425 [M + H]<sup>+</sup> (78), 336 (31), 304 (28), 303 (73), 301 (31), 285 (67), 242 (100); HRFABMS  $m/z$  425.26731 (calcd for C<sub>27</sub>H<sub>37</sub>O<sub>4</sub>, 425.26918).

**4-epi-Scopadulcic acid B (2):** gum; [ $\alpha$ ]<sub>D</sub><sup>25</sup> +3.0° ( $c$  0.50, CHCl<sub>3</sub>); IR (film)  $\nu_{\text{max}}$  3465, 2931, 2868, 1710, 1450, 1279, 1109, 1026, 756, 710 cm<sup>–1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Tables 1 and 2; FABMS  $m/z$  439 [M + H]<sup>+</sup> (17), 318 (20), 317 (100), 301 (12), 299 (15), 272 (13), 271 (53), 269 (13), 253 (13), 243 (12), 213 (12), 212 (12); HRFABMS  $m/z$  439.24757 (calcd for C<sub>27</sub>H<sub>35</sub>O<sub>5</sub>, 439.24845).

**Dulcidiol (4):** gum; [ $\alpha$ ]<sub>D</sub><sup>25</sup> –33.5° ( $c$  0.50, CHCl<sub>3</sub>); IR (film)  $\nu_{\text{max}}$  3415, 2931, 2866, 1710, 1450, 1388, 1284, 1115, 1065,

1027, 758, 710 cm<sup>–1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Tables 1 and 2; FABMS  $m/z$  427 [M + H]<sup>+</sup> (52), 398 (53), 392 (64), 379 (62), 287 (90), 283 (70), 274 (59), 267 (72), 265 (74), 243 (51), 242 (86), 240 (52), 236 (67), 228 (63), 227 (71), 219 (100); HRFABMS  $m/z$  427.28162 (calcd for C<sub>27</sub>H<sub>39</sub>O<sub>4</sub>, 427.28484).

**Scopanlol (5):** gum; [ $\alpha$ ]<sub>D</sub><sup>25</sup> –7.1° ( $c$  0.48, CHCl<sub>3</sub>); IR (film)  $\nu_{\text{max}}$  3454, 2931, 2870, 1710, 1668, 1450, 1277, 1111, 1026, 756, 712 cm<sup>–1</sup>; <sup>1</sup>H and <sup>13</sup>C NMR data, see Tables 1 and 2; EIMS  $m/z$  395 [M – CHO]<sup>+</sup> (8), 319 [M – COC<sub>6</sub>H<sub>5</sub>]<sup>+</sup> (9), 302 (21), 287 (16), 273 (21), 272 (25), 271 (28), 246 (33), 227 (21), 218 (56), 187 (100), 161 (37), 159 (63), 151 (68); HREIMS  $m/z$  395.25927 (calcd for C<sub>26</sub>H<sub>35</sub>O<sub>3</sub>, 395.25862).

**Cytotoxicity Assay.** A panel of six human stomach cancer cell lines SCL, SCL-6, SCL-37'6, SCL-9, Kato-3, and NUGC-4<sup>12–14</sup> were used to test the cytotoxicity of the pure compounds. The MTT assay as described by Mosmann<sup>15</sup> was employed to estimate the cell mortality. A series of serial dilutions (250, 125, 62.5, 31.25, and 15.63  $\mu\text{g/mL}$ ) of the pure compounds and controls were tested against each of the cell lines. For every concentration, three replicate analyses were performed. Percent cell mortality for each of the concentrations was estimated. ED<sub>50</sub> values in  $\mu\text{M}$  unit were then determined. Vinblastine sulfate and mitomycin C (Sigma Chemicals Co.) were used as positive controls. RPMI<sup>C</sup> (RPMI-1640 complete medium, which was used to culture the cancer cells for their confluent growth) and RPMI<sup>C</sup>-DMSO (RPMI<sup>C</sup> containing 0.25% DMSO, which was used to prepare the test materials and to culture the cells in the presence of the test materials) were used as negative controls. Cells grown in the RPMI<sup>C</sup> and RPMI<sup>C</sup>-DMSO were found to be the same and were considered 100% cell survival (that is, cell mortality was nil) to estimate cell mortality and to determine the ED<sub>50</sub> for the compounds tested. The SPSS software package (version 9.0) was used to analyze the data. Descriptive statistics were performed, and values were expressed as mean  $\pm$  standard error. Comparison of ED<sub>50</sub> values between the pure compounds and positive controls was performed by independent sample  $t$ -test.

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