Short Note

A New Diketopiperazine, Cyclo-(4-S-hydroxy-R-proline-R-isoleucine), from an Australian Specimen of the Sponge *Stelletta* sp. †

Simon P. B. Ovenden 1,2, Jonathan L. Nielson 1,3, Catherine H. Liptrot 1,4, Richard H. Willis 1, Dianne M. Tapiolas 1, Anthony D. Wright 1,5 and Cherie A. Motti 1,*

1 Australian Institute of Marine Science, PMB no. 3, Townsville MC, Townsville 4810, Australia; E-Mails: simon.ovenden@dsto.defence.gov.au (S.P.B.O.); jonathon.nielson@acdlabs.com (J.L.N.); catherine.liptrot@jcu.edu.au (C.H.L.); r.willis@aims.gov.au (R.H.W.); d.tapiolas@aims.gov.au (D.M.T.); adwright@hawaii.edu (A.D.W.)

2 Defence Science & Technology Organisation, 506 Lorimer St. Fishermans Bend, Victoria 3207, Australia

3 ACD Labs UK, Building A, Trinity Court, Wokingham Road, Bracknell, Berkshire RG42 1PL, UK

4 Advanced Analytical Centre, James Cook University, Townsville, QLD 4811, Australia

5 College of Pharmacy, University of Hawaii, 34 Rainbow Drive, Hilo, HI 96720, USA

† Dedication: We dedicate this paper to the memory of Dr. Peter Murphy (Townsville, Australia), a former AIMS colleague and dear friend, for his passionate contributions to the field of natural products chemistry and marine biodiversity, in particular recognising the need for access and benefit sharing arrangements between scientific organisations and local communities.

* Author to whom correspondence should be addressed; E-Mail: c.motti@aims.gov.au; Tel.: +61-7-4753-4143; Fax: +61-7-4772-5852.

Received: 29 August 2011; in revised form: 11 November 2011 / Accepted: 16 November 2011 / Published: 22 November 2011

**Abstract:** While investigating the cytotoxic activity of the methanol extract of an Australian marine sponge *Stelletta* sp. (Demospongiae), a new diketopiperazine, cyclo-(4-S-hydroxy-R-proline-R-isoleucine) (1), was isolated together with the known bengamides; A (2), F (3), N (4), Y (5), and bengazoles; Z (6), C4 (7) and C6 (8). The isolation and structure elucidation of the diketopiperazine (1), together with the activity of 1–8 against a panel of human and mammalian cell lines are discussed.
Keywords: Stelletta; diketopiperazine (DKP); cyclo-(4-S-hydroxy-R-proline-R-isoleucine); bengamide; bengazole; anti-cancer activity

1. Introduction

Since the first reported isolation of a diketopiperazine (DKP) from the sponge Dysidea herbacea [1], there have been several reports describing the isolation of this class of compound from other marine sponges [2–4]. DKPs are also reported from marine microbial sources [5–8], including the proteobacteria Alcaligenes faecalis, isolated from the sponge Stelletta tenuis [9]. The metabolites reported in these investigations are mostly the products of 4-hydroxy-proline [2,5,6,8] or proline [7] reacting with phenylalanine [2,5], arginine [4], leucine [5–7], isoleucine [7], norvaline [3] or alanine [8].

Sponges from the genus Stelletta are known to produce a number of other bioactive classes of compounds, including but not limited to steroids [10], alkaloids [11,12], isomalabaricane triterpenes [13], acetylenic acids [14] and lysophosphatidylcholines [15]. Initial interest in the methanol (MeOH) extract of the sponge Stelletta sp. was motivated by its potent activity in the NCI 60 cell line screen and a unique COMPARE analysis profile (average GI50 0.5 µg/mL) [16]. This profile was different to that of the standard chemotherapeutic agents paclitaxel, cisplatin, gemcitabine, bryostatin 1, didemnin B, tamoxifen and vinblastine (data provided by NCI). Subsequent bioassay-guided investigations of this extract led to the isolation of a new DKP cyclo-(4-S-hydroxy-R-proline-R-isoleucine) (1), the previously reported bengamides; A (2) [17], F (3) [18], N (4) [19], Y (5) [20], and the previously reported bengazoles; Z (6) [20], C4 (7) [21] and C6 (8) [21]. Described in this publication is the isolation and structure elucidation of 1, together with the activity of compounds 1–8.

2. Results and Discussion

The DKP cyclo-(4-S-hydroxy-R-proline-R-isoleucine) 1 was isolated and the molecular formula C11H18N2O3, corresponding to four double-bond equivalents, was determined by (+)-ESI-FTMS accurate mass measurement. The 13C NMR data of 1 contained resonances consistent with the presence of two amide carbonyl groups (δC 170.5 (C-7), 165.4 (C-1)) as the only multiple bonds within the molecule, and a hydroxy methine (δC 66.8 (C-4); δH 4.28, 1H, dd, J = 4.6, 4.6 Hz) (Table 1). These functionalities accounted for all of the oxygen and nitrogen atoms and all of the multiple bonds in 1, indicating the molecule to be bicyclic. Analysis of the COSY NMR data of 1 showed an extended 1H–1H spin system from H-9 to H3-12 via H-10 and H-11, as well as a vicinal COSY NMR correlation from H-10 to H3-13. Observed gHMBC NMR correlations from δH 7.97 to the 13C NMR resonances at δC 59.1 (C-9), δC 56.7 (C-6), δC 43.8 (C-10) and C-1 positioned this hydrogen at N-8. Further gHMBC NMR correlations from δH 4.00 (H-9) to δC 23.9 (C-11) and δC 14.9 (C-13), as well as to C-1 and C-10, clearly positioned H-9 adjacent to the C-1 carbonyl and N-8, giving rise to an isoleucine moiety (1A). Additional gHMBC NMR correlations from 8-NH and H-9 to C-7 revealed it was attached to N-8. A further contiguous 1H–1H spin system from H-6 to H2-3, in addition to gHMBC NMR correlations from the 8-NH and Hb-3 to C-6, and from Hb-3 to C-1 established the remaining nitrogen
(N-2) to be attached to C-1, C-6 and C-3, giving rise to the two rings within 1. The planar structure of 1 is as shown (Scheme 1).

**Table 1. NMR data for 1** (600 MHz, $d_6$-DMSO), cyclo-[S-proline-$S$-isoleucine)] (300 MHz, CDCl$_3$) and $^1$H NMR data for cyclo[L-(4-hydroxyprolinyl)-L-leucine)] (300 MHz, CD$_3$OD).

<table>
<thead>
<tr>
<th>No.</th>
<th>$^1$C $\delta$ (m)</th>
<th>$^1$H $\delta$ (m, J Hz)</th>
<th>COSY</th>
<th>gHMBC</th>
<th>$^1$H $\delta$ (m, J Hz) of cyclo-[S-proline-$S$-isoleucine)] [3]</th>
<th>$^1$H $\delta$ (m, J Hz) of cyclo[L-(4-hydroxyprolinyl)-L-leucine)] [6]</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>165.4 (s)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>53.8 (t)</td>
<td>3.51 (1H, dd, 12.5, 4.6)</td>
<td>H$_2$-3, H-4</td>
<td>C-1, C-4, C-5, C-6</td>
<td>3.6–3.5 (2H, m)</td>
<td>3.65 (1H, dd, 12.5, 4.3)</td>
</tr>
<tr>
<td></td>
<td>3.20 (1H, d, 12.5)</td>
<td>H$_2$-3</td>
<td>C-1, C-4, C-5, C-6</td>
<td>3.42 (1H, d, 12.5)</td>
<td>3.65 (1H, dd, 12.5, 4.3)</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>66.8 (d)</td>
<td>4.28 (1H, br dd, 12.5, 4.6)</td>
<td>H$_2$-3, 4-OH, H$_3$-5</td>
<td>C-3, C-6</td>
<td>2.0–1.9 (1H, m)</td>
<td>1.9–1.8 (1H, m)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4.28 (1H, br dd, 12.5, 4.6)</td>
<td>H$_2$-3, 4-OH, H$_3$-5</td>
<td>C-3, C-6</td>
<td>4.28 (1H, t, 4.3)</td>
<td></td>
</tr>
<tr>
<td>4-OH</td>
<td>5.10 (OH, br s)</td>
<td>H-4</td>
<td>-</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>37.2 (t)</td>
<td>2.03 (1H, dd, 12.9, 6.1)</td>
<td>H$_2$-5, H-6</td>
<td>C-3, C-4</td>
<td>2.3–2.2 (1H, m)</td>
<td>2.27 (1H, dd, 13.3, 6.5)</td>
</tr>
<tr>
<td></td>
<td>1.88 (1H, dd, 12.9, 6.1)</td>
<td>H-4, H$_2$-5, H-6</td>
<td>C-4, C-6, C-7</td>
<td>2.1–2.0 (1H, m)</td>
<td>2.08 (1H, dd, 13.3, 11.1, 4.3)</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>56.7 (d)</td>
<td>4.31 (1H, dd, 11.0, 6.1)</td>
<td>H$_2$-5</td>
<td>C-5, C-7</td>
<td>4.07 (1H, t, 7.5)</td>
<td>4.51 (1H, dd, 11.1, 6.5)</td>
</tr>
<tr>
<td>7</td>
<td>170.5 (s)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8-NH</td>
<td>7.97 (1H, s)</td>
<td>H-9</td>
<td>C-1, C-6, C-7, C-9, C-10</td>
<td>5.99 (1H, br s)</td>
<td>exchangeable</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>59.1 (d)</td>
<td>4.00 (1H, br s)</td>
<td>8-NH (w), H-10</td>
<td>C-1, C-7, C-10, C-11, C-13</td>
<td>3.96 (1H, br s)</td>
<td>4.15 (1H, m)</td>
</tr>
<tr>
<td>10</td>
<td>34.8 (d)</td>
<td>2.01 (1H, m)</td>
<td>H-9, H$_2$-11, H$_1$-13</td>
<td>C-1, C-13, C-11</td>
<td>2.4–2.3 (1H, m)</td>
<td>1.90 (1H, m)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>H-9, H$_2$-11, H$_1$-13</td>
<td>C-1, C-13, C-11</td>
<td>1.50 (1H, dd, 8.0)</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>23.9 (t)</td>
<td>1.32 (1H, qdd, 11.8, 7.4, 4.5)</td>
<td>H$_2$-11, H$_2$-12, H$_1$-12</td>
<td>C-9, C-10, C-12, C-13</td>
<td>1.5–1.4 (1H, m)</td>
<td>1.88 (1H, m)</td>
</tr>
<tr>
<td></td>
<td>1.26 (1H, qqdd, 11.8, 9.2, 7.2)</td>
<td>H$_2$-11, H$_2$-12, H$_1$-12</td>
<td>C-9, C-10, C-12, C-13</td>
<td>1.3–1.1 (1H, m)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>12.3 (q)</td>
<td>0.82 (3H, t, 7.4)</td>
<td>H$_2$-11</td>
<td>C-10, C-11</td>
<td>0.92 (3H, t, 7.4)</td>
<td>0.95 (3H, d, 6.4)</td>
</tr>
<tr>
<td>13</td>
<td>14.9 (q)</td>
<td>0.97 (3H, d, 7.0)</td>
<td>H-10</td>
<td>C-9, C-10, C-11</td>
<td>1.05 (3H, d, 7.2)</td>
<td>0.96 (3H, d, 6.4)</td>
</tr>
</tbody>
</table>

The configuration at C-4, C-6 and C-9 of 1 was established through analysis of $^1$H–$^1$H coupling constants, optical rotation measurement, molecular minimisations and comparison with literature compounds [3,6,7]. The magnitude of the coupling constants associated with H-6 (dd, $J = 11.0$, 6.1 Hz) and the observed COSY NMR correlations between H-6 and H$_{a,b}$-5 established it to have a pseudo-axial orientation, similar to that of cyclo[L-(4-hydroxyprolinyl)-L-leucine)] (H-6, dd, $J = 11.1$, 6.1 Hz) [6]. An apparent zero coupling between H-4 ($J = 4.6$, 4.6 Hz) and H$_{a}$-3 or H$_{b}$-5 as evident by lack of observed COSY NMR correlations, and observed couplings to H$_{a}$-3 ($J = 12.5$, 4.6 Hz) and H$_{b}$-5 ($J = 12.9$, 11.0, 4.6 Hz), was indicative of H-4 being orientated at approximately 90° to both H$_{a}$-3 and
Ha-5. The observed weak COSY NMR correlation between 8-NH and H-9, and the broad singlet for H-9 (similar to that observed in cyclo-[S-proline-S-isoleucine]) [3], revealed H-9 to be axial. Molecular modelling studies showed that the observed coupling constants were in agreement with either R,R (Figure 1) or S,S configuration at C-6/C-9 but definitely not R,S or S,R (Supplementary Data S6-S13 and Table S1). Based on optical rotation trends of DKPs from the literature [3,7], the overall positive $[\alpha]^{21}_{D} = +12^\circ$ indicated the absolute configuration at C-6 should be $R$, therefore supporting the R,R configuration. The magnitude of the optical rotation is also in agreement with other C-4 hydroxylated DKPs [5,7]. The molecular model shown in Figure 1, with calculated dihedral angles for $H_a$-5–C-5–C-6–H-6 ($\Phi = 41^\circ$), $H_b$-5–C-5–C-6–H-6 ($\Phi = 163^\circ$), H-4–C-4–C-5–$H_a$-5 ($\Phi = 79^\circ$), H-4–C-4–C-5–$H_b$-5 ($\Phi = -42^\circ$), $H_a$-3–C-3–C-4–H-4 ($\Phi = 29^\circ$), $H_b$-3–C-3–C-4–H-4 ($\Phi = -93^\circ$) and 8-NH–N-8–C-9–H-9 ($\Phi = 91^\circ$), best explained the observed COSY NMR correlations, $^1$H–$^1$H coupling constants and the positive sign of $[\alpha]^{21}_{D}$ confirmed the absolute configuration at C-3, C-6 and C-8 to be as shown. It is likely that this DKP was produced by an enzymatically controlled condensation reaction between D-isoleucine and 4-S-hydroxy-D-proline (Scheme 1) [22].

**Scheme 1.** Structures of the bengazoles, bengamides and 1 isolated from *Stelletta* sp. and the proposed enzymatically controlled condensation reaction between D-isoleucine and 4-S-hydroxy-D-proline to yield 1.
Figure 1. Minimum energy conformation of 1 obtained from MM2 calculations without applying any dihedral angle constraints [23]. The calculated dihedral angles for Hb-3–C-3–C-4–H-4 (−93°), H-4–C-4–C-4–Ha-5 (79°) and for 8-NH–N–8–C-9–H-9 (91°), all which approximate 90° as observed experimentally from the 1H–1H coupling constants, are indicative of the absolute configurations at C-4 as being S and at both C-6 and C-9 being R.

The cytotoxicity of 1–8 was investigated against the human tumour cell lines H460, SF-268, MCF-7, HT-29 and a normal mammalian cell line CHO-K1. The DKP 1 exhibited minimal activity towards MCF-7, H460 and HT-29 cells and no activity towards SF-268 or CHO-K1 cells at the highest dose (Table 2). In contrast, the GI50 values (µM) for bengamides A (2), F (3), N (4), Y(5), and bengazoles Z (6), C4 (7) and C6 (8) were comparable to those reported in previous studies [19,20], and accounted for the activity observed in the original MeOH extract.

Table 2. GI50 (µM) data for compounds 1–8 against SF-268, MCF-7, H460, HT-29 and CHO-K1.

<table>
<thead>
<tr>
<th>No.</th>
<th>SF-268 a</th>
<th>MCF-7 b</th>
<th>H460 c</th>
<th>HT-29 d</th>
<th>CHO-K1 e</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>&gt;295</td>
<td>204</td>
<td>234</td>
<td>270</td>
<td>&gt;295</td>
</tr>
<tr>
<td>2</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>0.1</td>
</tr>
<tr>
<td>3</td>
<td>1.8</td>
<td>0.7</td>
<td>0.6</td>
<td>1.5</td>
<td>32</td>
</tr>
<tr>
<td>4</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>&lt;0.02</td>
<td>0.2</td>
</tr>
<tr>
<td>5</td>
<td>72</td>
<td>52</td>
<td>25</td>
<td>48</td>
<td>&gt;184</td>
</tr>
<tr>
<td>6</td>
<td>22</td>
<td>18</td>
<td>8</td>
<td>13</td>
<td>94</td>
</tr>
<tr>
<td>7</td>
<td>0.3</td>
<td>0.8</td>
<td>0.1</td>
<td>0.6</td>
<td>1.2</td>
</tr>
<tr>
<td>8</td>
<td>0.02</td>
<td>0.06</td>
<td>&lt;0.02</td>
<td>0.1</td>
<td>0.8</td>
</tr>
</tbody>
</table>

a SF-268 Central nervous system-glioblastoma cells; b MCF-7 Breast-pleural effusion adenocarcinoma cells; c H460 Lung-large cell carcinoma cells; d HT-29 Colon-recto-sigmoid colon adenocarcinoma cells; e CHO-K1 Sub-clone of Chinese hamster ovary cells.
3. Experimental

3.1. General Experimental Procedures

General experimental details have been described previously [29].

3.2. Animal Material

This specimen of the sponge *Stelletta* sp., (Family Ancorinidae) was collected from the west side of Jamieson Reef, Bonaparte Archipelago, North West Western Australia, at depths ranging from 16 m to 20 m, in August 1991. A voucher specimen (Accession number QMG312281) has been lodged with the Queensland Museum.

3.3. Bioassay

Cellular bioassays were undertaken as previously described [19].

3.4. Extraction and Isolation

Freeze dried sponge material (125 g dry weight) was extracted sequentially with dichloromethane (CH$_2$Cl$_2$), MeOH and H$_2$O; activity was confined to the CH$_2$Cl$_2$ and MeOH fractions. The MeOH fraction was subjected to reversed phase C18 flash vacuum chromatography (RP-C18, 40%, 60%, 80%, 100% MeOH in H$_2$O, and 100% CH$_2$Cl$_2$) with activity located in the 40% and 100% MeOH fractions. The 100% MeOH fraction was further separated using RP HPLC (4 mL/min, gradient elution from 60% acetonitrile (CH$_3$CN):H$_2$O (+0.1% formic acid [HCO$_2$H]) to 100% CH$_3$CN (+0.1% HCO$_2$H) over 10 min, then isocratic 100% CH$_3$CN (+0.1% HCO$_2$H) for 15 min through a 150 mm × 10 mm 5 μ Phenomenex Luna C18 column), to give thirteen fractions. The first active fraction, fraction 1, was subjected to RP HPLC (4 mL/min, gradient elution from 20% CH$_3$CN:H$_2$O (+0.1% HCO$_2$H) to 100% CH$_3$CN (+0.1% HCO$_2$H) over 20 min through a 150 × 10 mm 5 μ Phenomenex Luna Phenyl-Hexyl column) to yield bengamide Y (5 mg, 0.0006%). The additional active fractions 3 and 4 were both partitioned with n-hexane and MeOH (1:1) to yield bengamides N (4 mg, 0.001%) and A (2 mg, 0.003%), respectively.

The 40% MeOH fraction was subjected to further RP-C18 (10%, 20%, 30%, 40%, 50% and 100% MeOH in H$_2$O) and the active fractions (30% and 40% MeOH) fractionated on RP HPLC (4 mL/min, gradient elution from 10% CH$_3$CN:H$_2$O (+0.1% HCO$_2$H) to 64% CH$_3$CN:H$_2$O (+0.1% HCO$_2$H) over 12 min, then isocratic 100% CH$_3$CN (+0.1% formic acid) for an additional 5 min through a 150 mm × 10 mm 5 μ Phenomenex Luna C18 column) to yield bengamide F (3 mg, 0.002%), bengazoles Z (6 mg, 0.004%), C$_4$ (7 mg, 0.011%) and C$_6$ (8 mg, 0.012%) and the new DKP cyclo-(4-S-hydroxy-R-proline-R-isoleucine) (1 mg, 0.001%).

3.4.1. Cyclo-(4-S-hydroxy-R-proline-R-isoleucine) (1)

Isolated as a colourless oil. [α]$_{D}^{21}$ +12° (c 0.025, CHCl$_3$); IR (film) $\nu_{max}$ 3391, 1649 cm$^{-1}$; UV (PDA, CH$_3$CN/H$_2$O) $\lambda_{max}$ 220 nm; $^1$H (600 MHz, $d_6$-DMSO) and $^{13}$C (150 MHz, $d_6$-DMSO) NMR data see Table 1; ESI-FTMS [M + Na]$^+$ 249.1203 (calcd. for C$_{11}$H$_{18}$N$_2$O$_3$Na 249.1215).
3.4.2. Bengamide A (2)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [17].

3.4.3. Bengamide F (3)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [18].

3.4.4. Bengamide N (4)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [19].

3.4.5. Bengamide Y (5)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [20].

3.4.6. Bengazole Z (6)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [20].

3.4.7. Bengazole C$_4$ (7)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [21].

3.4.8. Bengazole C$_6$ (8)

Isolated as a colourless oil. $^1$H NMR and $^{13}$C NMR spectral data were consistent with published values [21].

4. Conclusion

The DKP cyclo-(4-$S$-hydroxy-$R$-proline-$R$-isoleucine) (1), together with the known bengamides; A (2), F (3), N (4), Y (5), and bengazoles; Z (6), C$_4$ (7) and C$_6$ (8), was isolated from the Australian marine sponge *Stelletta* sp. Interestingly, this is the first report of bengamides or bengazoles from the genus *Stelletta*, however, it should be noted that they have previously been reported from species of *Dorypleres splendens* [24], which has since been reclassified as *Jaspis splendens*, and from *Jaspis* sp. [24], both of which belong to the Ancorinidae family of sponges. The cyclo-(4-$S$-hydroxy-$R$-proline-$R$-isoleucine) (1) was not cytotoxic against the cell lines MCF-7, H460, HT-29, SF-268 or CHO-K1. The DKP class of compounds has recently gained interest in drug discovery [25] due to their chiral, rigid and functionalised structures. These features enable them to bind to a large variety of
receptors with high affinity giving rise to a broad range of biological activities, including cytotoxicity, quorum sensing, antibacterial, antifungal, antiviral, antiprion, antitumor, and immunosuppressive functions, even plant-growth regulators [7,26–28]. Our report adds to the vast knowledge of these potentially therapeutic molecules.

Acknowledgements

Collection of this sponge was made possible by the access and benefit sharing arrangements between AIMS and the Australian Commonwealth Government. The authors are grateful to those AIMS staff, both past and present, involved in the collection of the sponge. We thank A. Carroll, Eskitis Institute, Griffith University for facilitating measurement of optical rotations, and B. Bowden, Department of Pharmacy and Molecular Sciences, James Cook University for use of the Departments’ FTIR instrument. We also thank A-M. Babey, School of Veterinary and Biomedical Sciences, James Cook University for initial cytotoxicity screening data and for the SF268 cell line, and C. Hooi, R. Anderson and C. Cullinane, of the Peter MacCallum Cancer Centre, Melbourne, Australia, for the HT-29, H460, MCF-7 and CHO-K1 cell lines.

References


Samples Availability: Available from the authors.

© 2011 by the authors; licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution license (http://creativecommons.org/licenses/by/3.0/).