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Divergent synthesis of cytotoxic styryl lactones isolated from *Polyalthia crassa*. The first total synthesis of crassalactone B

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ABSTRACT

The first total synthesis of (+)-crassalactone B (**2**) and a new syntheses of (+)-crassalactone C (**3**) has been achieved starting from D-glucose. The natural products **2** and **3** can be selectively accessed by changing the conditions for TBDPS cleavage in the final intermediate **16**. The synthesized natural products were evaluated for their cytotoxic activity against a panel of human tumour cell lines.

Ethno-botanical uses of several species of the genus *Polyalthia* are well known in South East Asia. The extracts from these plants are traditionally used as a galactagogue, as a remedy for the treatment of abdominal pain, and as an aphrodisiac.[1] Chemical constituents of *Polyalthia* species have been extensively investigated by different groups; many of these constituents exhibit antimicrobial,[2] cytotoxic,[3] antimalarial,[4] and anti-HIV[5] activity. A number of styryl lactones have been recently isolated from the tropical plant *Polyalthia crassa*.[6] The bioassay-directed separation of the ethyl acetate soluble extract of the leaves and twigs led to the isolation of the known antitumor agent (+)-goniofufurone (**1**, Fig. 1), along with several new cytotoxic lactones including the cinnamate derivatives of **1**: (+)-crassalactones B (**2**) and C (**3**). Their structures were determined on the basis of spectroscopic methods. The absolute configurations of **2** and **3** were established by chemical semisynthesis, starting from a natural sample of goniofufurone.

We recently disclosed the first total synthesis of (+)-**3** by chirality transfer from D-xylose.[7] In continuation of this work, we report herein the first total synthesis of (+)-crassalactone B (**2**) and an alternative synthesis of (+)-crassalactone C (**3**) starting from D-glucose, along with their effects on the proliferation of some human malignant cell lines.

A retrosynthetic pathway outlining our strategy for the synthesis of (+)-crassalactone B (**2**) is shown in Scheme 1. The strategy is based on the utility of protected furanose **4**, comprising the requisite 3-*O*-cinnamoyl and 5-*C*-phenyl functionalities, as well as four contiguous stereocentres of definite configuration. It was assumed that the required [3.3.0] bicyclic lactone core of **2** could be formed from **4** through a two-step sequence that involved hydrolytic removal of the 1,2-*O*-isopropylidene protecting group, followed by hydroxy directed condensation of a protected lactol derivative with Meldrum's acid.[8] The key intermediate **4** could be prepared from the protected dialdose **7** or **10** by means of stereoselective Grignard addition of phenylmagnesium bromide. Synthesis of aldehydes of type **7** or **10** is visualised from D-glucose by well established chemical reactions.[9] In such a way, the chirality at C-4, C-5 and C-6 in the target **2** would be directly translated from D-glucose.

At the outset, we focused on the synthesis of the key building block **4** starting from commercially available diacetone-D-glucose (**5**, Scheme 2).

Treatment of **5** with cinnamic acid under the conditions shown in Scheme 2, provided an almost quantitative yield of the expected cinnamic ester **6**. The terminal isopropylidene functionality in **6** was directly converted into an aldehyde group, in a single step, by treatment with periodic acid in dry ethyl acetate,[10] whereupon the corresponding aldehyde **7** was obtained in 93% yield. Addition of phenylmagnesium bromide in THF, to a solution of **7** in toluene, led to a mixture of two epimeric alcohols. The mixture was not separated but was oxidised with PCC in dichloromethane to give the corresponding ketone **8** in 34% yield. Stereocontrolled reduction of the ketone functionality was achieved using the method

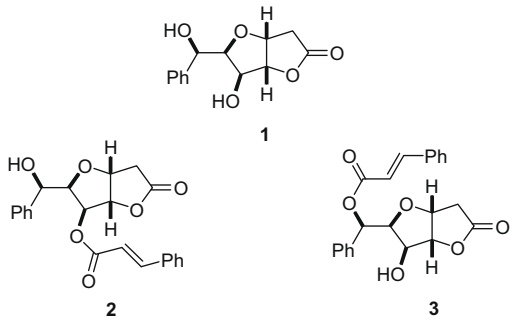

Figure 1. Structures of (+)-goniofufurone (**1**), (+)-crassalactone B (**2**) and (+)-crassalactone C (**3**).

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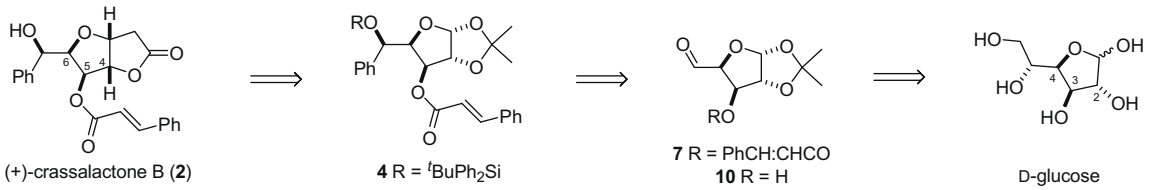
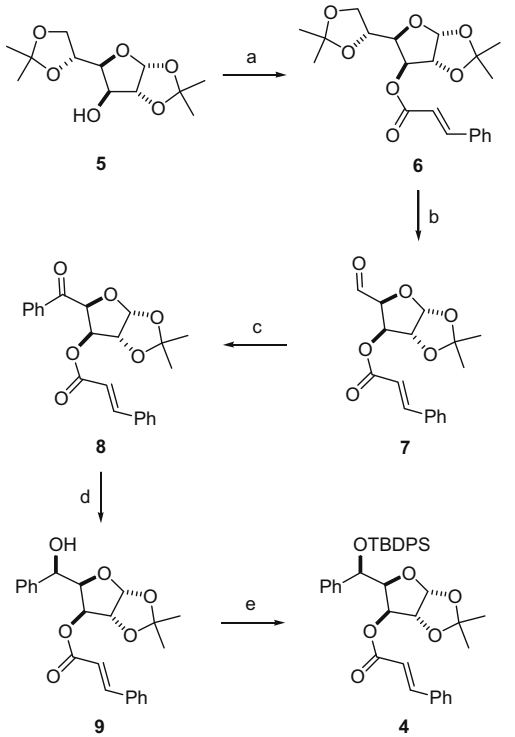


Scheme 1. Retrosynthetic analysis of (+)-crassalactone B.

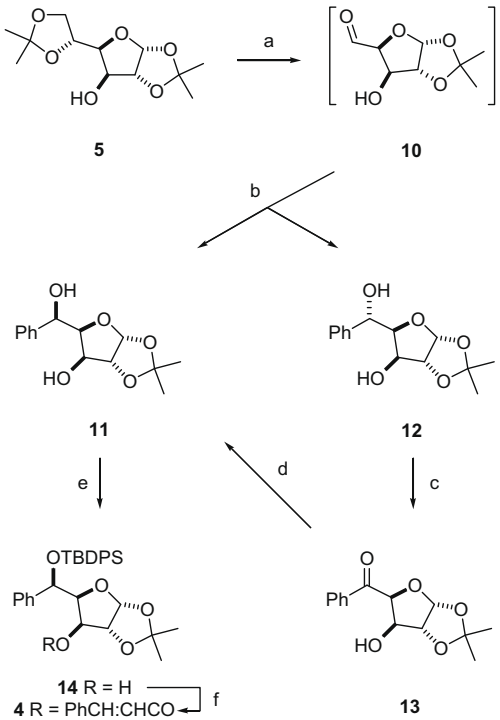
of Yatagai and Ohnuki for prochiral ketones.[11] The reduction of **8** with $NaBH_4$ in the presence of L-tartaric acid gave the required alcohol **9** as the only stereoisomer in 92% yield. The intermediate **9** was thus obtained in 28% overall yield with respect to the starting compound **5**. However, when this four-step sequence was carried out without purification of the intermediates **7** and **8** the desired product **9** was obtained in a somewhat higher overall yield (34% from **5**). To prevent the benzylic hydroxy group in **9** from taking part in ensuing reactions, it was blocked as a silyl ether by treatment of **9** with *tert*-butyldiphenylsilyl chloride to give the key intermediate **4**[12] in 90% yield (31% overall yield from **5**).

An alternative and more efficient route to the intermediate **4** is outlined in Scheme 3. This sequence started with treatment of **5** with periodic acid in dry ethyl acetate. The resulting crude aldehyde **10** was not purified, but was immediately treated with PhMgBr to give a 75% combined yield of known[9,13] alcohols **11** and **12** in the ratio 3:2. Efforts to improve the epimeric ratio in favour of **11** possessing the correct stereochemistry for **2** were unsuccessful. However access to **11** was possible by selective DDQ-oxidation of the benzylic hydroxy function in **12**[14] followed by reduction of the resulting prochiral ketone **13** with $NaBH_4$/L-tartaric acid. This procedure provided the requisite intermediate **11** in 64% overall yield (four steps from **5**). The benzylic hydroxy group in **11** was silylated selectively to give the corresponding silyl ether **14** in 97% yield. Further esterification of **14** with cinnamic acid (DCC, DMAP) gave the key chiral intermediate **4** in 96% yield. Thus, this six-step sequence (Scheme 3) represents a more convenient route toward the key intermediate **4**, since it provided a considerably higher overall yield (59% from **5**) compared to the six-step sequence presented in Scheme 2 (31% from **5**).

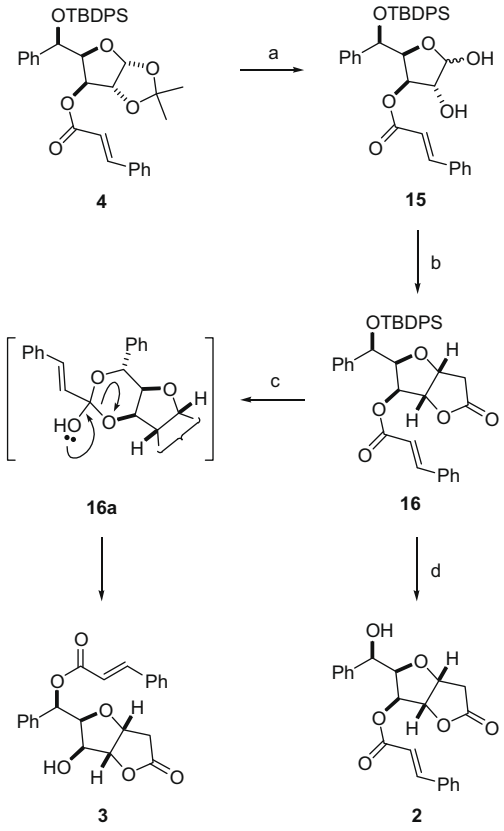
Conversion of **4** into the targets **2** and **3** is outlined in Scheme 4. Hydrolytic removal of the isopropylidene protecting group in **4** gave the expected lactol **15** (92%), which upon treatment with Meldrum's acid in the presence of triethylamine gave the protected furano-lactone **16** in 65% yield. We initially carried out the TBDPS cleavage in **16** using TBAF,[15] which surprisingly gave (+)-crassalactone C (**3**) as the major product (43%), accompanied by a minor amount of **2** (14%). Both the ^1H and ^{13}C NMR data of compound **3** were consistent with naturally occurring (+)-crassalactone C and its physical properties were in good agreement with those reported in the literature.[6,7] Product **3** was presumably formed by a competitive intramolecular cinnamate migration process in **2**, via the cyclic orthoester intermediate **16a**. A similar intramolecular acyl migration during silyl deprotection with TBAF has been described recently in the literature.[16] Moreover, treatment of **16** with $SOCl_2$ in MeOH gave a 56% yield of (+)-crassalactone B (**2**).



Scheme 2. Reagents and conditions: (a) $PhCH{=}CHCO_2H$, DCC, DMAP, CH_2Cl_2, rt, 20 h, 98%; (b) H_5IO_6, EtOAc, rt, 1.5 h, 93%; (c) (i) PhMgBr, THF, PhMe, 0 °C, 3 h; (ii) PCC, CH_2Cl_2, reflux, 4 h, or Ac_2O, DMSO, rt, 24 h, 32% over two steps; (d) $NaBH_4$, THF, L-tartaric acid, −7 °C→rt, 4 h, 92%; (e) $^tBuPh_2SiCl$, imidazole, CH_2Cl_2, rt, 47 h, 90%.



Scheme 3. Reagents and conditions: (a) H_5IO_6, EtOAc, rt, 4 h; (b) PhMgBr, THF, PhMe, 0 °C, 5 h, 46% of **11**, 29% of **12**; (c) DDQ, CH_2Cl_2, H_2O, rt, 72 h, 79%; (d) $NaBH_4$, THF, L-tartaric acid, −7 °C→rt, 24 h, 78% of **11**, 7% of **12**; (e) $^tBuPh_2SiCl$, imidazole, CH_2Cl_2, rt, 48 h, 97%; (f) $PhCH{=}CHCO_2H$, DCC, DMAP, CH_2Cl_2, rt, 50 h, 96%.



Scheme 4. Reagents and conditions: (a) TFA/H_2O (9:1), CH_2Cl_2, 0 °C, 0.5 h, 92%; (b) Meldrum's acid, Et_3N, DMF, 47 °C, 63 h, 65%; (c) TBAF, AcOH, THF, rt, 144 h, 43% of **3**, 14% of **2**; (d) $SOCl_2$, MeOH, rt, 6 h, 56%.

Although our optical rotation value is greater than the reported value for (+)-crassalactone B {lit.[6] $[\alpha]_D^{20}$ +8.0 (*c* 0.5, EtOH); this work: $[\alpha]_D^{20}$ +45.7 (*c* 0.5, EtOH)}, the melting point and NMR data[17] of synthetic sample **2** were in full agreement with those reported in the literature.[6] Thus, the first synthesis of natural product **2** was successfully accomplished in nine steps with an overall yield of 19.8% starting from commercially available D-glucose derivative **5**. The new synthesis of crassalactone C (**3**) proceeded in nine linear steps, with 15.2% overall yield from the same starting compound **5**. The preceding preparation of **3** was accomplished starting from D-xylose in 7.8% overall yield over ten linear steps.[7]

Both (+)-crassalactones B (**2**) and C (**3**) were evaluated for their antitumor activity against human myelogenous leukaemia (K562), promyelocytic leukaemia (HL-60), T cell leukaemia (Jurkat), breast adenocarcinoma (MCF-7), cervix carcinoma (HeLa), as well as a normal human cell line (foetal lung fibroblasts, MRC-5). In vitro cytotoxicity was evaluated after 48 h cell treatment by the MTT assay.[18] The results, including the data for the reference compound (+)-goniofufurone (**1**), are given in Table 1.

Table 1
In vitro cytotoxicity of **1**, **2** and **3**

Compound	IC_{50}[a] (μM)					
	K562	HL-60	Jurkat	MCF-7	HeLa	MRC-5
1	7.29	>100	65.87	25.31	10.36	>100
2	1.57	5.46	0.45	1.22	2.12	>100
3	13.78	>100	61.24	15.26	27.36	>100

[a] IC_{50} is the concentration of compound required to inhibit cell growth by 50% compared to an untreated control. The values are means of three independent experiments performed in quadruplicate. Coefficients of variation were <10%.

All three naturally occurring styryl lactones **1**–**3** retained the selectivity between the normal human cells (MRC-5) and tumour cell lines. These results are consistent with previous findings that the cytotoxicity of styryl lactones is specific to neoplastic cells since only negligible effects of these natural products on normal cells were observed.[19] Interestingly, only (+)-crassalactone C (**3**) has the same growth inhibition pattern as the parent compound **1**. As with (+)-goniofufurone (**1**), compound **3** was inactive against promyelocytic leukaemia (HL-60), but showed moderate to weak antiproliferative activity towards K562, MCF-7, HeLa and Jurkat malignant cells. In contrast to **1** and **3**, (+)-crassalactone B (**2**) exhibited significant antiproliferative effects towards all the tested tumour cell lines, including notable activity against HL-60 cells. The most potent antiproliferative activity of **2** was recorded in the Jurkat cell line, whereupon this molecule exhibited 146- and 136-fold stronger activities when compared to **1** and **3**, respectively. Molecule **2** also showed remarkable antiproliferative activities toward K562, MCF-7 and HeLa cells, being 5- to 17-fold, more potent than (+)-goniofufurone (**1**), and 9- to 13-fold more potent than (+)-crassalactone C (**3**).

In summary, we have developed the first total synthesis of (+)-crassalactone B (**2**) and an alternative synthesis of (+)-crassalactone C (**3**) by chirality transfer from D-glucose, whereby the C-2, C-3 and C-4 stereocenters of D-glucose have been translated into the C-4, C-5 and C-6 positions of the targets. Selective access to either **2** or **3** was accomplished by simply changing the conditions for TBDPS cleavage in the final intermediate **16**. The main characteristic of this approach is its generality and flexibility. It enables the preparation of a variety of (+)-crassalactone B and C analogues by changing the *O*-acyl functionality at the C-3 position of D-glucose. In vitro antiproliferative activities of **2** and **3** against a number of human tumour cell lines were recorded and compared with those observed for (+)-goniofufurone. The obtained biological data revealed that (+)-crassalactone B showed superior activity against all the tested malignant cell lines, and was devoid of any significant cytotoxicity against normal MRC-5 cells. Based upon these results, we believe that this compound may serve as a convenient lead in the synthesis of more potent and selective antitumor agents.

Acknowledgement

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References and notes

1. (a) Kanokmedhakul, S.; Kanokmedhakul, K.; Ohtani, I. I.; Isobe, M. *Phytochemistry* **1998**, *47*, 131–133; (b) Connolly, J. D.; Haque, E.; Kadir, A. A. *Phytochemistry* **1996**, *43*, 295–297.
2. (a) Faizi, S.; Khan, R. A.; Azher, S.; Khan, S. A.; Tauseef, S.; Ahmad, A. *Planta Med.* **2003**, *69*, 350–355; (b) Murthy, M. M.; Subramanyam, M.; Bindu, M. H.; Annapurna, J. *Fitoterapia* **2005**, *76*, 336–339.
3. (a) Wu, Y. C.; Duh, C. Y.; Wang, S. K.; Chen, K. S.; Yang, T. H. *J. Nat. Prod.* **1990**, *53*, 1327–1331; (b) Zhao, G.; Jung, J. H.; Smith, D. L.; Wood, K. V.; McLaughlin, J. L. *Planta Med.* **1991**, *57*, 380–383; (c) Ma, X.; Lee, I. S.; Chai, H. B.; Zaw, K.; Farnsworth, N. R.; Soejarto, D. D.; Cordell, G. A.; Pezzuto, J. M.; Kinghorn, A. D. *Phytochemistry* **1994**, *37*, 1659–1662; (d) Chen, C. Y.; Chang, F. R.; Shih, Y. C.; Hsieh, T. J.; Chia, Y.-C.; Tseng, H. Y.; Chen, H. C.; Chen, S. J.; Hsu, M. C.; Wu, Y. C. *J. Nat. Prod.* **2000**, *63*, 1475–1478.
4. Kanokmedhakul, S.; Kanokmedhakul, K.; Yodbuddee, D.; Phonkerd, N. *J. Nat. Prod.* **2003**, *66*, 616–619.
5. (a) Li, H. Y.; Sun, N. J.; Kashiwada, Y.; Sun, L.; Snider, J. V.; Cosentino, L. M.; Lee, K. H. *J. Nat. Prod.* **1993**, *56*, 1130–1133; (b) Tuchinda, P.; Pohmakotr, M.; Reutrakul, V.; Thanyachareon, W.; Sophasan, S.; Yoosook, C.; Santisuk, T.; Pezzuto, J. M. *Planta Med.* **2001**, *67*, 572–575.
6. Tuchinda, P.; Munyoo, B.; Pohmakotr, M.; Thinapong, P.; Sophasan, S.; Santisuk, T.; Reutrakul, V. *J. Nat. Prod.* **2006**, *69*, 1728–1733.
7. (a) Popsavin, V.; Benedeković, G.; Srećo, B.; Popsavin, M.; Francuz, J.; Kojić, V.; Bogdanović, G. *Org. Lett.* **2007**, *9*, 4235–4238; (b) Popsavin, V.; Benedeković, G.; Srećo, B.; Francuz, J.; Popsavin, M.; Kojić, V.; Bogdanović, G.; Divjaković, V. *Tetrahedron* **2009**, *65*, 10596–10607.

8. For a recent review on applications of Meldrum's acid in the synthesis of natural products, see: Ivanov, A. S. *Chem. Soc. Rev.* **2008**, *37*, 789–811.
9. Gracza, T.; Szolcsányi, P. *Molecules* **2000**, *5*, 1386–1398.
10. Reddy, L. V. R.; Swamy, G. N.; Shaw, A. K. *Tetrahedron: Asymmetry* **2008**, *19*, 1372–1375.
11. Yatagai, M.; Ohnuki, T. *J. Chem. Soc., Perkin Trans. 1* **1990**, 1826–1828.
12. *Selected data for* **4** (syrup): $[\alpha]_D^{20}$ +18.6 (*c* 2.0, $CHCl_3$). ^1H NMR (250 MHz, $CDCl_3$): δ 0.93 (s, 9 H, Me_3CPh_2Si), 1.30 and 1.58 (2 × s, 3H each, CMe_2), 4.57 (d, 1H, $J_{1,2}$ = 3.7 Hz, H-2), 4.70 (dd, 1H, $J_{3,4}$ = 2.6, $J_{4,5}$ = 8.7 Hz, H-4), 4.95 (d, 1H, $J_{4,5}$ = 8.7 Hz, H-5), 5.47 (d, 1H, $J_{3,4}$ = 2.6 Hz, H-3), 5.77 (d, 1H, $J_{1,2}$ = 3.7 Hz, H-1), 6.15 (d, 1H, $J_{2',3'}$ = 16.0 Hz, H-2′), 7.08–8.65 (m, 21H, 4 × Ph and H-3′). ^{13}C NMR (62.9 MHz, $CDCl_3$): δ 19.2 (Me_3CPh_2Si), 26.2 and 26.7 (CMe_2 and Me_3CPh_2Si), 72.6 (C-5), 76.4 (C-3), 82.1 and 82.9 (C-2 and C-4), 104.4 (C-1), 111.9 (CMe_2), 117.3 (C-2′), 127.0, 127.5, 127.8, 128.0, 128.06, 128.1, 128.9, 129.2, 129.4, 130.5, 132.9, 133.1, 134.0, 135.8, 136.0 and 140.6 (4 × Ph), 145.2 (C-3′), 165.7 (C-1′). HRMS (ESI): *m*/*z* 657.2651 (M^++Na), calcd for $C_{39}H_{42}NaO_6Si$: 657.2643.
13. (a) Inch, T. D. *Carbohydr. Res.* **1967**, *5*, 45–52; (b) Bruns, R.; Wernicke, A.; Koll, P. *Tetrahedron* **1999**, *55*, 9793–9800.
14. Stereoisomers **11** and **12** were separated by flash column chromatography before oxidation.
15. Mori, M.; Somada, A.; Oida, S. *Chem. Pharm. Bull.* **2000**, *48*, 716–728.
16. (a) Nishiyama, T.; Kusumoto, Y.; Okumura, K.; Hara, K.; Kusaba, S.; Hirata, K.; Kamiya, Y.; Isobe, M.; Nakano, K.; Kotsuki, H.; Ichikawa, Y. *Chem. Eur. J.* **2010**, *16*, 600–610; (b) Chevallier, O. P.; Migaud, M. E. *Beilstein J. Org. Chem.* **2006**, 2. article no. 14.
17. *Selected data for* **2**: mp 172–174 °C (Et_2O), $[\alpha]_D^{20}$ +45.7 (*c* 0.5, EtOH); lit.[6] mp 171–173 °C (EtOH), $[\alpha]_D^{20}$ +8.0 (*c* 0.5, EtOH). ^1H NMR (250 MHz, $CDCl_3$): δ 2.59 (d, 1H, $J_{2a,2b}$ = 18.8 Hz, H-2a), 2.74 (dd, 1H, $J_{2a,2b}$ = 18.8, $J_{2b,3}$ = 5.8 Hz, H-2b), 4.27 (dd, 1H, $J_{5,6}$ = 2.5, $J_{6,7}$ = 8.6 Hz, H-6), 4.69 (d, 1H, $J_{6,7}$ = 8.6 Hz, H-7), 5.00–5.09 (m, 2H, H-3 and H-4), 5.76 (d, 1H, $J_{5,6}$ = 2.5 Hz, H-5), 6.55 (d, 1H, $J_{2',3'}$ = 15.9 Hz, H-2′), 7.30–7.65 (m, 10 H, 2 × Ph), 7.85 (d, 1H, $J_{2',3'}$ = 15.9 Hz, H-3′). ^{13}C NMR (62.9 MHz, $CDCl_3$): δ 35.6 (C-2), 70.8 (C-7), 75.0 (C-5), 77.1 (C-3), 83.1 (C-6), 85.3 (C-4), 116.0 (C-2′), 126.8, 128.4, 128.6, 129.1, 130.8, 131.1, 133.7 and 140.3 (2 × Ph), 147.6 (C-3′), 166.5 (C-1′), 174.8 (C-1). HRMS (ESI): *m*/*z* 403.1143 (M^++Na), calcd for $C_{22}H_{20}NaO_6$: 403.1152.
18. Scudiero, D. A.; Shoemaker, R. H.; Paull, K. D.; Monks, A.; Tierney, S.; Nofziger, T. H.; Currens, M. J.; Seniff, D.; Boyd, M. R. *Cancer Res.* **1988**, *48*, 4827–4833.
19. For a recent review on biological activities of styryl lactones and analogues, see: de Fatima, A.; Modolo, L. V.; Conegero, L. S.; Pilli, R. A.; Ferreira, C. V.; Kohn, L. K.; de Carvalho, J. E. *Curr. Med. Chem.* **2006**, *13*, 3371–3384.