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Wittig reaction with partially protected sugar lactol derivatives. Preparation of highly cytotoxic goniofufurone analogues

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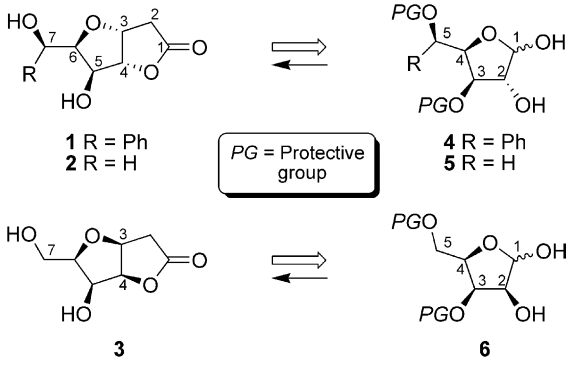
Abstract—A new approach to the dephenyl goniofufurone analogue **2** and the corresponding (3*S*,4*R*)-stereoisomer **3** is reported. The resulting furanolactones **2** and **3** have shown a potent and selective in vitro cytotoxicity against certain human tumour cell lines.

Natural styryl lactones have received a great deal of interest due to their widespread occurrence[1] and their varied antitumour activities.[2] Notable among them is (+)-goniofufurone (**1**, Scheme 1), a cytotoxic styryl lactone that was isolated from the stem bark of *Goniothalamus giganteus* (Annonaceae).[3] Due to the unique structural features and significant biological activity, (+)-**1** and its stereoisomers have been synthesized by many groups from different starting materials.[4] Owing to their potential as antitumour agents a number of goniofufurone analogues have also been synthesized,[5] including the recent preparation of furanolactones **2** and **3** (dephenyl goniofufurone analogues).[6] However, data related to their biological activities has not been reported so far. Herein we report an alternative synthesis of the lactones **2** and **3** for evaluation of their antiproliferative activity against human tumour cell lines.



Scheme 1. Goniofufurone (**1**) and analogues.

In 1993 Prakash and Rao[7] reported an efficient synthesis of **1** starting from D-glucose. The key step of the synthesis was a spontaneous lactonisation and Michael ring closure accompanying a *Z*-selective Wittig reaction[8] of ethoxycarbonylmethylenetriphenylphosphorane with a furanose lactol **4** having a free hydroxyl group at C-2. Accordingly it was assumed that a similar Wittig reaction with D-xylo- and D-lyxofuranose lactol derivatives of type **5** and **6** might provide access to the furanolactones **2** and **3**, respectively.

Although the furanolactone **2** can be obtained in a single step by condensation of D-xylose with Meldrum's acid,[9] we envisaged its preparation via the protected D-xylofuranose derivative **7**[10] (Scheme 2) in order to explore a possible divergent route that would be suitable not only for the preparation of **2**, but also for an alternative synthesis of (+)-goniofufurone and 7-*epi*-goniofufurone. The lactol **7** readily reacted with the stabilized ylide

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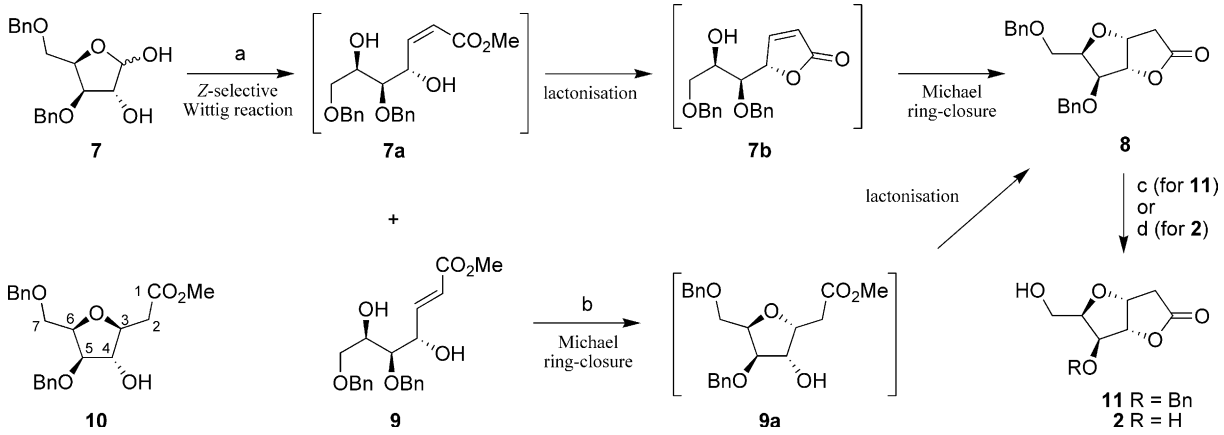
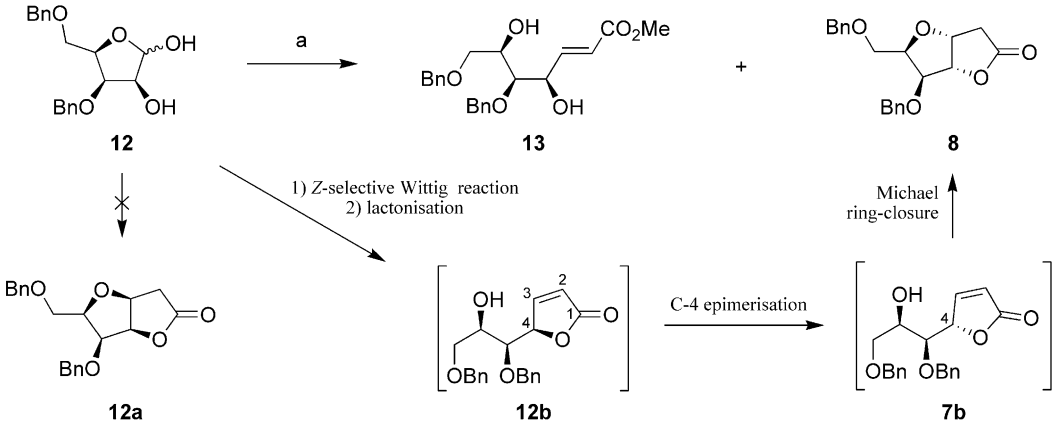


Scheme 2. Reagents and conditions: (a) $Ph_3P{:}CHCO_2Me$, MeOH, rt, 24 h, 61% of **8**, 25% of **9**; (b) imidazole, C_6H_6, reflux, 11 h, 51% of **8**, 17% of **10**; (c) H_2, 10% Pd/C (0.1 equiv), rt, 1.5 h, 84%; (d) H_2, 10% Pd/C (0.2 equiv), rt, 24 h, 87%.

$Ph_3P{:}CHCO_2Me$ in dry methanol to give the expected furanolactone **8** (61%) accompanied by a minor amount of the corresponding *E*-enoate **9** (25%). Both **8** and **9** had been prepared earlier in our laboratory, but in a rather different ratio (12% of **8**; 74% of **9**).[11] The major reaction product **8** was presumably formed from the respective *Z*-enoate **7a** through a sequential lactonisation—Michael cyclisation process.[7,12] If the sequence had proceeded otherwise (with Michael ring closure taking precedence over lactonisation) we would have detected in the reaction mixture at least traces of β-*C*-furanoside **10**, which from steric reasons cannot lactonise. In an alterative sequence, in which Michael ring closure preceded lactonisation, the *E*-enoate **9** was treated with imidazole in boiling benzene for 11 h. Under these reaction conditions the lactone **8** was obtained in 51% yield, along with a minor amount of the β-*C*-glycoside **10**[13] (17%) thus providing an indirect proof for the mechanism of conversion of **7** to **8**. The 'anomeric' configuration of **10** (at C-3) was established by an NOE experiment, which showed a through-space interaction between H-3 and the 4-OH.

Catalytic reduction of **8** over 10% Pd/C (0.1 M equiv of Pd) for 1.5 h at room temperature effected selective removal of the benzyl group from the primary position to afford the alcohol **11**[14] in 84% yield. Compound **11** could presumably be converted to (+)-goniofufurone, or to its 7-epimer, via a three-step sequence that would comprise a selective oxidation of **11** to the corresponding aldehyde, reaction with PhMgBr, followed by OH group deprotection. When the hydrogenolysis of **8** was repeated using an excess of the catalyst (0.2 M equiv of Pd) and prolonging the reaction time to 24 h, the known[6,9,15] goniofufurone analogue **2** was obtained (87%), with physical constants and spectral data in good agreement with those already reported.[6,9,15]

After the successful preparation of the goniofufurone analogue **2**, we were also interested in obtaining the corresponding (3*S*,4*R*)-stereoisomer **3**. This was of interest to us as several goniofufurone stereoisomers have been shown to exhibit moderate to significant antitumour activities.[2,5] In our initial efforts to discover an expedient route to **3**, we first examined a Wittig reaction of the D-lyxofuranose **12**[10] with $Ph_3P{:}CHCO_2Me$ in dry methanol (Scheme 3). Although these conditions were found to be optimal for obtaining *Z*-selectivity in the D-*xylo* series, the major product in the reaction of **12** was the corresponding *E*-alkene **13**, which was isolated in 34%



Scheme 3. Reagents and conditions: (a) $Ph_3P{:}CHCO_2Me$, MeOH, rt, 24 h, 34% of **13**, 14% of **8**.

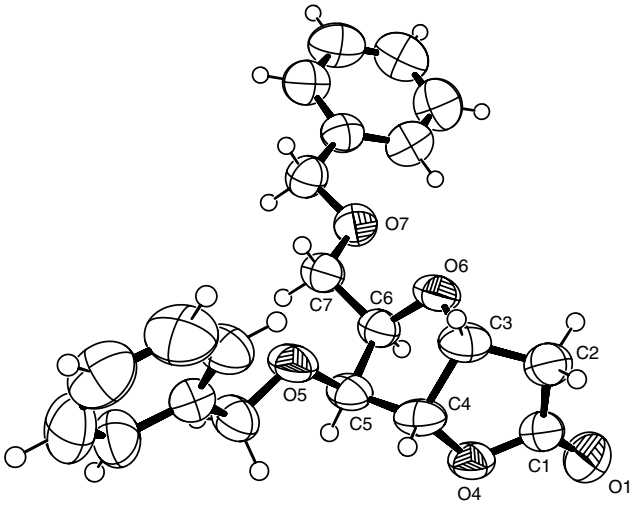


Figure 1. ORTEP drawing of the furanolactone **8** with non-H labelling scheme. The displacement ellipsoids are drawn at 50% probability.

yield, along with the furanolactone **8** (14%) as a minor product. The expected stereoisomer **12a** was not detected in the reaction mixture. Compound **8** could be formed by inversion of configuration at C-4 in the unsaturated lactone **12b**, which is a possible intermediate in this reaction. A similar epimerisation has been observed with D-mannose in the Wittig reaction,[16] as well as with D-galactose[9] and D-mannose[17] in reaction with Meldrum's acid. The structure of compound **8** was confirmed by ^{1}H and ^{13}C NMR spectra, which displayed the same signals as a sample of **8** prepared from the D-*xylo*-lactol **7**, as well as by a single crystal X-ray diffraction analysis.[18]

The X-ray analysis of furanolactone **8** (Fig. 1) obtained from D-lyxofuranose derivative **12**, reveals a *cis* junction between the five-membered rings, as well as a *trans* orientation of the lactone ring with respect to the substituents at C-5 and C-6. The tetrahydrofuran ring exists in an envelope conformation, with C-5 above the plane that contains the C-3, C-4, C-6 and O-6 atoms. The lactone ring also adopts an envelope conformation, but with C-3 below the plane formed by the remaining ring atoms. The values of the torsion angles C2–C3–C4–O4 = −25.2°, O4–C4–C5–O5 = 168.6°, O5–C5–C6–C7 = 168.6° and O5–C5–C6–O6 = 81.6° are consistent with the D-*ido* configuration of **8**.

The undesired outcome of the previous reaction (attempted conversion of **12** to **12a**) forced us to find an alternative route for the preparation of **3**. The synthesis started from the known[19] D-lyxofuranose derivative **14**, which is readily available from D-lyxose in two steps (Scheme 4). Thus, the lactol **14** reacted with $Ph_3P{:}CHCO_2Me$ in refluxing acetonitrile to form an intermediate enoate (not shown in the Scheme), which then underwent a spontaneous Michael ring closure, affording a mixture of α- and β-*C*-glycosides **15**[20] and **16**[21] in a ratio of ca. 1:1. The stereochemistry at C-3 in **15** was assigned on the basis of a NOE interaction between H-2 and H-4. A NOE between the *endo*-orientated methyl group (from the isopropylidene moiety) and H-3, definitely confirmed the D-*talo* configuration of the product. The stereochemistry of **16** at C-3 was resolved on the bases of a NOE interaction between H-2 and *endo*-Me protons. Treatment of purified **15** with NaOMe in MeOH effected equilibration (via a ring-opening and ring-closure mechanism)[22] to give a 10:1 mixture of **16** and **15**, with the β-*C*-glycoside **16** as the major product. The overall yield of **16** was 74% calculated from the starting compound. Hydrolytic removal of the isopropylidene protective group in **16** with aqueous trifluoroacetic acid occurred with concomitant lactonisation to give the target molecule **3** in 80% yield. The physical constants, as well as the ^{1}H and ^{13}C NMR spectral data of product **3** were in agreement with the reported values.[6]

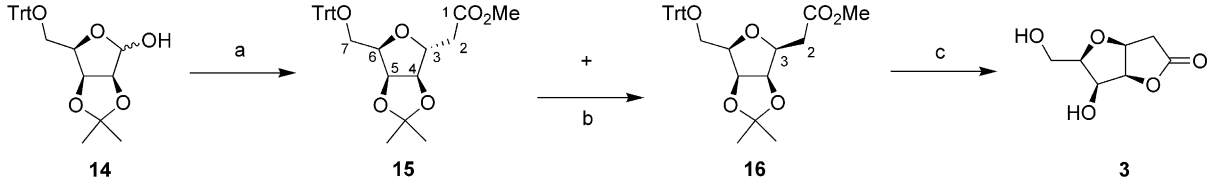
The synthesized analogues **2** and **3** were assayed in vitro for their antiproliferative activity against human myelogenous leukaemia K562, colon adenocarcinoma HT29, estrogen receptor negative breast adenocarcinoma MDA-MB-231, and normal foetal lung MRC-5 cells. In vitro cytotoxicity was evaluated after 24 h cell treatment using the MTT assay.[23] The results, including the data for the reference compound, doxorubicin (DOX), are presented in Table 1.

As shown in the Table 1, the analogue **2** was active only against the K562 cell line, but was more than 6-fold less active than the reference compound, doxorubicin.

Table 1. In vitro cytotoxicity of target compounds

Compds	IC_{50}, μM[a]			
	K562	HT-29	MDA-MB-231	MRC-5
2	5.02	>100	>100	>100
3	0.0074	0.21	0.39	>100
DOX	0.82	0.51	0.29	0.32

[a] IC_{50} is the concentration of compound required to inhibit cell growth by 50% compared to an untreated control.



Scheme 4. Reagents and conditions: (a) $Ph_3P{:}CHCO_2Me$, MeCN, reflux, 24 h, 48% of **15**, 43% of **16**; (b) NaOMe, MeOH, reflux, 48 h, 61% of **16**, 6% of **15**; (c) 2:1 TFA–H_2O, rt, 18 h, 80%.

However, the (3*S*,4*R*)-stereoisomer **3** showed a remarkable cytotoxicity towards the K562 and HT29 cell lines, being approximately 110- and 2.5-fold, respectively, more potent than doxorubicin. In experiments with MDA-MB-231 cells, the analogue **3** exhibited a strong cytotoxicity similar to that observed for the reference compound. Neither furanolactone **2** or **3** exhibited any significant cytotoxicity towards normal foetal lung MRC-5 cells.

In summary, we have synthesized dephenyl goniofufurone analogues **2** and **3**, which have shown significant antiproliferative activity against certain human neoplastic cells. The furanolactone **3** showed strong cytotoxicity towards the K562 and HT29 cell lines, being much more potent than doxorubicin, an approved drug for treatment of these malignant diseases, while both analogues **2** and **3** did not exhibit any cytotoxicity towards the normal MRC-5 cell line. In addition, this approach provided a convenient procedure for selective removal of the primary benzyl group in **8**, thus enabling access to **11**, a postulated intermediate in the synthesis of (+)-goniofufurone **1** or 7-*epi*-goniofufurone, which is also a natural product.[24]

Acknowledgements

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References and notes

1. Blazquez, M. A.; Bermejo, A.; Zafra-Polo, M. C.; Cortes, D. *Phytochem. Anal.* **1999**, *10*, 161–170.
2. Mareyala, H. B.; Joe, M. *Curr. Med. Chem. Anti-Cancer Agents* **2001**, *1*, 293–300.
3. Fang, X. P.; Anderson, J. E.; Chang, C. J.; Fanwick, P. E.; McLaughlin, J. L. *J. Chem. Soc., Perkin Trans. 1* **1990**, 1655–1661.
4. (a) Su, Y. L.; Yang, C. S.; Teng, S. J.; Zhao, G.; Ding, Y. *Tetrahedron* **2001**, *57*, 2147–2153, and references cited therein; (b) Mareyala, H. B.; Gadikota, R. R. *Indian J. Chem. Sect. B* **2000**, *39*, 166–172; (c) Yang, M.; Li, H. M.; Zhao, G.; Yu, Q. S.; Ding, Y. *Chin. J. Chem.* **2000**, *18*, 225–231.
5. Mereyala, H. B.; Gadikota, R. R.; Joe, M.; Arora, S. K.; Dastidar, S. G.; Agarwal, S. *Bioorg. Med. Chem.* **1999**, *7*, 2095–2103.
6. Cagnolini, C.; Ferri, M.; Jones, P. R.; Murphy, P. J.; Ayres, B.; Cox, B. *Tetrahedron* **1997**, *53*, 4815–4820.
7. Prakash, K. R. C.; Rao, S. P. *Tetrahedron* **1993**, *49*, 1505–1510.
8. (a) Mukaiyama, T.; Suzuki, K.; Yamada, T.; Tabusa, F. *Tetrahedron* **1990**, *46*, 265–276; (b) Valverede, S.; Martin-Lomas, M.; Herradon, B.; Garcia-Ochoa, S. *Tetrahedron* **1987**, *43*, 1895–1901.
9. Mata, F. Z.; Martinez, M. B.; Peréz, J. A. G. *Carbohydr. Res.* **1990**, *201*, 223–231.
10. Popsavin, V.; Grabež, S.; Stojanović, B.; Popsavin, M.; Pejanović, V.; Miljković, D. *Carbohydr. Res.* **1999**, *321*, 110–115.
11. Popsavin, V.; Grabež, S.; Krstić, I.; Popsavin, M.; Djoković, D. *J. Serb. Chem. Soc.* **2003**, *68*, 795–804.
12. Prakash, K. R. C.; Rao, S. P. *Tetrahedron Lett.* **1991**, *32*, 7473–7476.
13. Selected data for **10** (syrup): $[\alpha]_D^{23}$ −18.7 (*c* 1.0, $CHCl_3$); ^1H NMR (250 MHz, $CDCl_3$): δ 2.68 (dd, 1H, $J_{2a,3}$ = 9.6, $J_{2a,2b}$ = 17 Hz, H-2a), 2.97 (dd, 1H, $J_{2b,3}$ = 5, $J_{2a,2b}$ = 17 Hz, H-2b), 3.12 (br s, 1H, OH), 3.60–3.80 (m, 5H, 2 × H-7 and CO_2CH_3), 3.93 (m, 1H, H-3), 3.99 (dd, 1H, $J_{4,5}$ = 2.3, $J_{5,6}$ = 5.1 Hz, H-5), 4.08 (dd, 1H, $J_{3,4}$ = 4.9, $J_{4,5}$ = 2.3 Hz, H-4), 4.23 (m, 1H, H-6), 4.48–4.72 (4d, 4H, J_{gem} = 12 Hz, 2 × CH_2Ph), 7.24–7.39 (m, 10H, 2 × Ph); ^{13}C NMR (62.5 MHz, $CDCl_3$): δ 38.3 (C-2), 52.0 (CO_2CH_3), 68.7 (C-7), 71.6 and 73.4 (2 × CH_2Ph), 79.7 (C-6), 80.8 (C-3), 81.1 (C-4), 85.3 (C-5), 127.5, 127.6, 127.8, 128.3, 128.4, 137.9 and 138.1 (Ph), 172.9 (C-1).
14. Selected data for **11** (syrup): $[\alpha]_D^{23}$ +4.3 (*c* 1.0, in $CHCl_3$); ^1H NMR (250 MHz, $CDCl_3$): δ 2.52 (br s, 1H, OH), 2.58–2.78 (m, 2H, 2 × H-2), 3.76 (dd, 1H, $J_{6,7a}$ = 4.3, $J_{7a,7b}$ = 12 Hz, H-7a), 3.84 (dd, 1H, $J_{6,7b}$ = 5.1, $J_{7a,7b}$ = 12 Hz, H-7b), 4.17 (m, 1H, H-6), 4.25 (d, 1H, $J_{5,6}$ = 4.9 Hz, H-5), 4.56 and 4.71 (2d, 2H, J_{gem} = 12 Hz, CH_2Ph), 4.91–5.01 (m, 2H, H-3 and H-4), 7.26–7.42 (m, 5H, Ph); ^{13}C NMR (62.5 MHz, $CDCl_3$): δ 35.8 (C-2), 61.1 (C-7), 72.7 (CH_2Ph), 76.7 (C-3), 80.7 (C-6), 82.1 (C-5), 85.7 (C-4), 127.6, 128.2, 128.6 and 136.7 (Ph), 175.2 (C-1).
15. Dmitriev, B. A.; Chernyak, A. Y.; Kochetkov, N. K. *Zhur. Org. Khim.* **1972**, *41*, 2754–2760.
16. Collins, P. M.; Overend, W. G.; Shing, T. S. *J. Chem. Soc., Chem. Commun.* **1982**, 297–298.
17. Mata, F. Z.; Martinez, M. B.; Peréz, J. A. G. *Carbohydr. Res.* **1992**, *225*, 159–161.
18. Crystallographic data for **8** have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication number CCDC 249584. Copies of the data can be obtained, free of charge, on application to CCDC, 12 Union Road, Cambridge, CB2 1EZ, UK [fax: +44(0)-1223-336033 or e-mail: deposit@ccdc.cam.ac.uk.
19. Lin, C.-C.; Fan, G.-T.; Fang, J.-M. *Tetrahedron Lett.* **2003**, *44*, 5281–5283.
20. Selected data for **15**: mp 138 °C (from MeOH); $[\alpha]_D^{23}$ −21.7 (*c* 0.8, $CHCl_3$); ^1H NMR (250 MHz, $CDCl_3$): δ 1.31 and 1.35 (2s, 3H each, CMe_2), 2.47 (dd, 1H, $J_{2a,3}$ = 7.9, $J_{2a,2b}$ = 15.3 Hz, H-2a), 2.57 (dd, 1H, $J_{2b,3}$ = 7.4, $J_{2a,2b}$ = 15.3 Hz, H-2b), 3.37 (dd, 1H, $J_{7a,7b}$ = 9.5, $J_{6,7a}$ = 6.3 Hz, H-7a), 3.45 (dd, 1H, $J_{7a,7b}$ = 9.5, $J_{6,7b}$ = 5.7 Hz, H-7b), 3.73 (s, 3H, CO_2CH_3), 3.97 (m, 1H, H-6), 4.47 (ddd, 1H, $J_{3,4}$ = 1.1, $J_{2a,3}$ = 7.9, $J_{2b,3}$ = 7.4 Hz, H-3), 4.61 (dd, 1H, $J_{3,4}$ = 1.1, $J_{4,5}$ = 6.1 Hz, H-4), 4.78 (dd, 1H, $J_{4,5}$ = 6.1, $J_{5,6}$ 3.9 Hz, H-5), 7.19–7.54 (m, 15H, 3 × Ph); ^{13}C NMR (62.5 MHz, $CDCl_3$): δ 25.3 and 26.2 (CMe_2), 36.3 (C-2), 51.9 (CO_2CH_3), 61.7 (C-7), 79.5 (C-6), 80.3 (C-3), 81.0 (C-5), 84.8 (C-4), 86.8 (CPh_3), 112.7 (CMe_2), 126.9, 127.7, 128.8 and 144.0 (Ph), 170.8 (C-1).
21. Selected data for **16** (syrup): $[\alpha]_D^{23}$ −26.3 (*c* 1.0, $CHCl_3$); ^1H NMR (250 MHz, $CDCl_3$): δ 1.34 and 1.36 (2 × s, 3H each, CMe_2), 2.78 (dd, 1H, $J_{2a,3}$ = 6.6, $J_{2a,2b}$ = 16.7 Hz, H-2a), 2.83 (dd, 1H, $J_{2b,3}$ = 7, $J_{2a,2b}$ = 16.7 Hz, H-2b), 3.42 (dd, 1H, $J_{7a,7b}$ = 9.5, $J_{6,7a}$ = 6.4 Hz, H-7a), 3.49 (dd, 1H, $J_{7a,7b}$ = 9.5, $J_{6,7b}$ = 6 Hz, H-7b), 3.70 (ddd, 1H, $J_{5,6}$ = 2.9, $J_{6,7a}$ = 6.4, $J_{6,7b}$ = 6 Hz, H-6), 3.72 (s, 3H, CO_2CH_3), 3.94 (ddd, 1H, $J_{3,4}$ = 2.9, $J_{2a,3}$ = 6.6, $J_{2b,3}$ = 7 Hz, H-3), 4.78 (m, 2H, H-4 and H-5), 7.17–7.58 (m, 15H, 3 × Ph); ^{13}C NMR (62.5 MHz, $CDCl_3$): δ 25.2 and 25.8 (CMe_2), 33.4 (C-2), 51.7 (CO_2CH_3), 61.3 (C-7), 77.4 (C-3), 80.6 (C-6), 81.0 and

81.1 (C-4 and C-5), 86.8 (CPh_3), 112.2 (CMe_2), 126.8, 127.6, 128.8 and 144.0 (Ph), 171.5 (C-1).

22. Ohrui, H.; Jones, G. H.; Moffatt, J. G.; Maddox, M. L.; Christensen, A. T.; Byram, S. K. *J. Am. Chem. Soc.* **1975**, *97*, 4602–4613.
23. 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.
24. Fang, X. P.; Anderson, J. E.; Chang, C. J.; McLaughlin, J. L.; Fanwick, P. E. *J. Nat. Prod.* **1991**, *54*, 1034–1043.