Preparation of Phenolic Paclitaxel Metabolites

Haeil Park,[†] Michael Hepperle,[†] Thomas C. Boge,[†] Richard H. Himes,[‡] and Gunda I. Georg*,[†]

Departments of Medicinal Chemistry and Biochemistry, University of Kansas, Lawrence, Kansas 66045

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The synthesis and biological evaluation of the two known phenolic metabolites of paclitaxel are described. The C3′-phenolic metabolite **2** of paclitaxel was prepared from 7-(triethylsilyl)-baccatin III (**8**) and enantioenriched *N*-benzoyl-2-azetidinone **7**. The C2-phenolic metabolite **3** was synthesized from paclitaxel (**1a**) *via* selective C2 debenzoylation and reacylation.

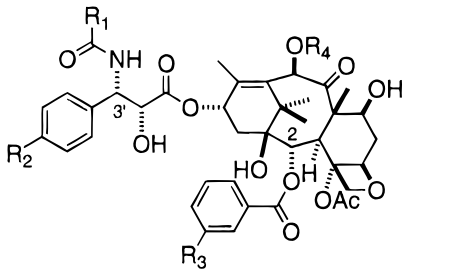
Introduction

Paclitaxel (**1a**; Figure 1) is regarded as one of the most promising new agents for the treatment of drug-refractory ovarian cancer and metastatic breast cancer.[1,2] Clinical evaluations of its potential for the chemotherapy of other cancers have revealed impressive response rates for head and neck cancer, lung cancer, and esophageal carcinomas.[1,2] Paclitaxel is a mitotic spindle poison with a unique mechanism of action, promoting the formation of stable microtubules.[3]

Although paclitaxel has undergone detailed clinical, biochemical, chemical, and structure−activity investigations,[4] its *in vivo* drug disposition is not yet fully understood.[5,6] Since early pharmacological studies revealed insignificant urinary excretion of paclitaxel, it became clear that protein binding, metabolism, and biliary excretion must play a significant role in its disposition.[5]

A paclitaxel metabolism study, conducted in rats, investigated the elimination of nonradioactive paclitaxel in bile and urine.[7] Only about 10% of paclitaxel was recovered in the urine of the rats. Biliary excretion accounted for 12% of unchanged paclitaxel and 29% of metabolites. Nine metabolites were detected, four of which could be identified, but the identity of the other five is still unknown.[6] Two phenolic compounds were found as the principal metabolites. The major metabolite (13%) carries a hydroxyl group at the para position of the 3′-phenyl group in the C13 side chain (**2**; Figure 1). The second phenolic metabolite, accounting for 5% of the administered dose, is hydroxylated at the meta position of the 2-benzoate group (**3**; Figure 1). Baccatin III, which is obtained after hydrolysis of the C13-ester group in paclitaxel, and a C19-hydroxylated product were identified among the minor metabolites accounting for less than 10%. The metabolites **2** and **3**, identified in these *in vivo* studies, were also detected when metabolism assays were carried out with rat hepatocytes.[8] Cytochrome P450 3A enzymes appear to effect the observed aromatic hydroxylation.[8]

Biliary excretion is also the major pathway for human paclitaxel clearance; however, the metabolic products are different. In one metabolism study, 10% of the administered paclitaxel dose was recovered unchanged from urine and 20% from bile.[5] In another disposition study, about 14% of paclitaxel and an unknown major polar metabolite were found in urine, yet the total fecal excretion amounted to over 71%.[9] Examination of human bile provided evidence for the formation of five to seven metabolites in addition to unchanged paclitaxel (3−5%).[5,9] The major human metabolite is 6α-hydroxypaclitaxel (12−16%).[5,9,10] 6α-Hydroxypaclitaxel is also the major metabolite in human liver microsomes.[11−13] Metabolite **2** was found in 2% and other products in about 3% in human bile as percent of the total administered dose, including the dihydroxylated paclitaxel metabolite 3′-dephenyl-3′-(4-hydroxyphenyl)-6α-hydroxypaclitaxel.[5,6] 3′-Hydroxyphenyl metabolite **2** was also identified as a metabolism product in human liver microsomes.[13,14] The identities of other metabolites in human liver microsomes and additional metabolites found in plasma[5,9,15] are not known.



1a R_1=Ph, R_2=R_3=H, R_4=Ac, paclitaxel
1b R_1=*tert*-Butyl, R_2=R_3=H, R_4=H, docetaxel
2 R_1=Ph, R_2=OH, R_3=H, R_4=Ac
3 R_1=Ph, R_2=H, R_3=OH, R_4=Ac

Figure 1. Structures of paclitaxel (**1a**) and docetaxel (**1b**) and the phenolic paclitaxel metabolites **2** and **3**.

It is interesting to note that human and rat biliary metabolites of docetaxel (**1b**; Figure 1), a semisynthetic analogue of paclitaxel, are quite different from metabolites of paclitaxel.[6,16] Apparently, oxidative metabolism takes place exclusively at the *N*-*tert*-butoxycarbonyl group of docetaxel in both humans and rats. No docetaxel metabolites modified at the 3′-phenyl group or at the taxane ring system have been identified.

Although the chemical synthesis of docetaxel metabolites has been described,[17] the preparation of paclitaxel metabolites has not been reported yet. Herein we wish to detail the synthesis of the two known phenolic paclitaxel metabolism products **2** and **3**, through methodology developed in our laboratory for the preparation of 3′-phenyl-modified[18] and 2-benzoate-modified[19] paclitaxel analogues.

Results and Discussion

The 3′-phenyl-hydroxylated paclitaxel metabolite was synthesized by sodium hydride-mediated acylation of

[†] Department of Medicinal Chemistry.
[‡] Department of Biochemistry.
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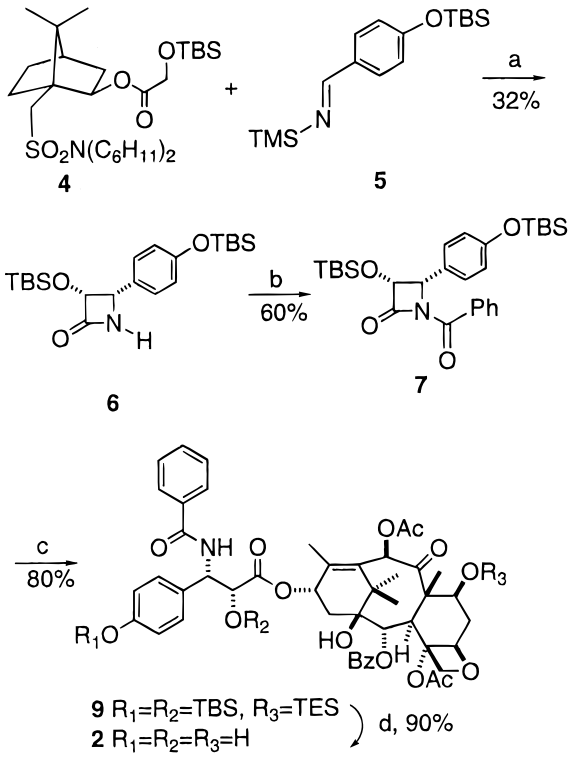
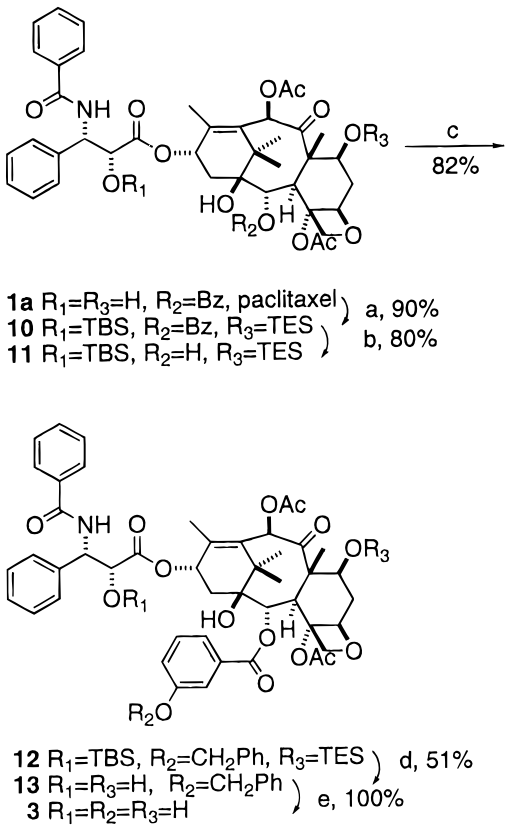
Scheme 1[a]



[a] (a) LDA, THF, −78 °C to room temperature, 12 h; (b) BzCl, Et_3N, DMAP, 1 h; (c) 7-(triethylsilyl)baccatin III (**8**), NaH, THF, 35 °C, 4 h; (d) pyridinium hydrogen fluoride, pyridine, 0 °C to room temperature, 8 h.

Scheme 2[a]



[a] (a) Imidazole, TBSCl, DMF, room temperature, 8 h, then TESCl, 5 h; (b) *t*-BuOK, H_2O, THF, −40 to −15 °C, 24 h; (c) 3-(benzyloxy)benzoic acid, DMAP, DCC, toluene, 55 °C, 20 h; (d) pyridinium hydrogen fluoride, pyridine, 0 °C to room temperature, 6 h; (e) Pd/C, cyclohexene:ethanol = 1:1, 60 °C, 2 h.

protected baccatin III **8** with enantioenriched (3*R*,4*S*)-*N*-benzoyl-2-azetidinone **7** (Scheme 1). The asymmetric ester enolate−imine cyclocondensation reaction of glycolate **4**[20] and *N*-(trimethylsilyl)benzaldimine **5** resulted in the formation of *β*-lactam **6** in 32% yield and 80% ee. Acylation of **6** with benzoyl chloride in the presence of triethylamine and a catalytic amount of *N*,*N*-(dimethylamino)pyridine (DMAP) provided *N*-benzoyl *β*-lactam **7** in 60% yield. Reaction of the sodium alkoxide of 7-(triethylsilyl)baccatin III (**8**)[21] with compound **7** afforded protected taxane **9** in 80% yield. Due to kinetic resolution in the acylation step, only one diastereoisomer of **9** was formed. Simultaneous deprotection of all three silyl groups with pyridinium hydrofluoride produced a 90% yield of paclitaxel metabolite **2**.

The paclitaxel metabolite **3**, hydroxylated at the 2-benzoyl group, was prepared *via* DCC/DMAP-promoted coupling of 2-debenzoylpaclitaxel **11** with 3-(benzyloxy)benzoic acid (Scheme 2). The two secondary hydroxyl groups of paclitaxel were selectively protected in a one-flask procedure to give compound **10** in excellent yield by adding first *tert*-butyldimethylsilyl chloride to silylate the C2′-hydroxyl group followed by triethylsilyl chloride to silylate the C7-hydroxyl group. Protected paclitaxel **10** could now easily be debenzoylated at C2 with anhydrous KOH to give **11** in high yield.[22] The bulky *tert*-butyldimethylsilyl protecting group at C2′ is necessary to prevent the hydrolytic cleavage of the C13 side chain under the strong basic reaction conditions,[23] and the C7-hydroxyl group must be protected to avoid base-catalyzed C7 isomerization as well as acylation in the following step.[24] Esterification of compound **11** and 3-(benzyloxy)benzoic acid[25] in the presence of DCC and DMAP in refluxing methylene chloride gave reacylated **12** in 82% yield. Treatment of **12** with pyridinium hydrofluoride in pyridine removed the silyl protecting groups and provided C2-modified paclitaxel **13** in 51% yield. Hydrogen transfer conditions, generated by cyclohexene and 10% Pd/C in ethanol, converted compound **13** into metabolite **3**.

Metabolites **2** and **3** were characterized by positive FAB mass spectrometry as well as ^1H and ^{13}C NMR spectroscopy. Both derivatives show a molecular ion peak at $m/z = 870$ (MH^+), consistent with the addition of one hydroxyl group to the paclitaxel molecule ($m/z = 853$). The ^1H NMR spectral data are very similar to the ones reported by Monsarrat *et al.* for **2** and **3**.[7] Since the para position of the 3′-phenyl group in **2** is hydroxylated, two doublets for the ortho and meta protons at 7.23 and 6.75 ppm are observed. The introduction of a *m*-hydroxyl group at the C2-benzoate moiety causes the resonance of one of the ortho protons to appear as a singlet at 7.73 ppm. The resonance of the phenolic proton in **3** is detected at 9.05 ppm. The benzylic methylene protons in **13** are identified as a nicely separated quartet at 5.16 ppm.

Biological Evaluation. The paclitaxel metabolites **2** and **3** as well as the benzylated precursor **13** of metabolite **3** were tested and compared with paclitaxel for their ability to initiate the polymerization of tubulin in the microtubule assembly assay and for their cytotoxicity against B16 melanoma cells (Table 1).[26] Metabolite **2** was about 2 times as active in the microtubule assembly assay but about 7 times less cytotoxic against B16 melanoma cells than paclitaxel. This result sug-

Table 1. *In Vitro* Biological Evaluation of Paclitaxel Metabolites **2** and **3** and Precursor **13**

	ED_{50}/ED_{50} (paclitaxel)	
compd	microtubule assembly assay[a]	B16 melanoma[b]
2	0.60	6.8
3	2.0	>20
13	24	not evaluated

[a] ED_{50} is the concentration which causes polymerization of 50% of the tubulin present in 15 min at 37 °C. [b] ED_{50} refers to the concentration which produces 50% inhibition of proliferation after 40 h incubation.

gests that the phenolic hydroxyl group does not interfere with the binding of **2** to microtubules but may reduce cellular uptake because of the increased hydrophilicity. Similar bioactivities were obtained by Monsarrat *et al.*, who reported that **2** was as active as paclitaxel in a microtubule disassembly assay and about 9 times less cytotoxic than the parent against L1210 leukemia cells.[7] Metabolite **3** was reported to have greatly reduced activity (39 times) on L1210 leukemia cell growth but showed paclitaxel-like activity in the microtubule disassembly assay.[7] The reported data are in accordance with the results from our investigations, which demonstrate paclitaxel-like activity of **3** in the microtubule assembly assay yet significant loss of cytotoxicity against B16 melanoma cells. The poor activity of **13** was unexpected because other meta-substituted 2-aroyl-2-debenzoylpaclitaxel analogues, carrying a 3-methoxy,[27] 3-chloro,[19] or other meta substituents, display improved or paclitaxel-like activity in tubulin assays.[19,27] The greatly reduced activity of **13** points to a severe interference of the benzyl group with the paclitaxel binding site.

Summary

The chemical syntheses of the two known phenolic paclitaxel metabolites **2** and **3** are reported for the first time. The compounds were prepared in good overall yields by semisynthesis from baccatin III and an *N*-acyl β-lactam and by a selective deacylation−reacylation protocol. The availability of these metabolites through synthesis should facilitate further studies of the disposition of paclitaxel.

Experimental Section

General. For general synthetic procedures, see ref 28. 4-[(*tert*-Butyldimethylsilyl)oxy]benzaldehyde was prepared by standard silylation of 4-hydroxybenzaldehyde. All other starting materials are commercially available, or their syntheses have been described in the literature. For a description of the biological assays, see refs 26 and 29.

4-[(*tert*-Butyldimethylsilyl)oxy]-*N*-(trimethylsilyl)-benzaldimine (5).[30] To 1,1,1,3,3,3-hexamethyldisilazane (57.5 mL, 27.5 mmol) was added slowly *n*-butyllithium (100 mL, 25 mmol, 2.5 M hexane solution). The reaction mixture was cooled to 0 °C and diluted with THF (100 mL). After 20 min, 4-[(*tert*-butyldimethylsilyl)oxy]benzaldehyde (5.9 g, 25 mmol) was added dropwise to the reaction mixture over 10 min. The reaction was continued for 30 min at 0 °C and quenched with trimethylsilyl chloride (23.5 mL). After 30 min, the solvent was evaporated to one-half of the original volume. *n*-Pentane (500 mL) was then added to the reaction mixture. The precipitate was filtered using Celite, and removal of the solvent *in vacuo* provided the imine in 95% (7.3 g) as a colorless liquid, which was used without further purification: ^{1}H NMR (300 MHz, $CDCl_3$) δ −0.06 (s, 3H, $SiCH_3$), −0.02 (s, 3H, $SiCH_3$), 0.01 (s, 9H, $Si(CH_3)_3$), 0.74 (s, 9H, $C(CH_3)_3$), 6.63 (d, J = 7.8 Hz, 2H, aryl), 7.46 (d, J = 7.8 Hz, 2H, aryl), 8.67 (s, 1H, CH═N).

(3*R*,4*S*)-3-[(*tert*-Butyldimethylsilyl)oxy]-4-[4-[(*tert*-butyldimethylsilyl)oxy]phenyl]-2-azetidinone (6). To a stirred solution of diisopropylamine (0.38 mL, 2.69 mmol) in THF (1 mL) was added a 2.5 M solution of *n*-butyllithium (1.08 mL, 2.69 mmol) in hexanes at −78 °C under argon gas. The reaction mixture was stirred at 0 °C for 30 min and then recooled to −78 °C. To the cold mixture was then added a solution of the chiral glycolate **4**[20] (1180 mg, 2.07 mmol) in THF (2 mL) *via* cannula under argon gas. The reaction mixture was stirred for 1 h at −78 °C followed by the addition of benzaldimine **5** (826 mg, 2.69 mmol). The mixture was stirred at −78 °C for 4 h and then at room temperature for 8 h. The reaction was quenched with the addition of a saturated NH_4Cl solution (5 mL); then the mixture was diluted with diethyl ether (100 mL) and washed with brine (100 mL). The organic layer was dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. Purification of the crude material by silica gel flash chromatography (EtOAc:hexane = 1:9) gave 266 mg (32%) of β-lactam **6** as a white solid. HPLC analysis for the determination of the enantiomeric excess of azetidinone **6** was performed on a Waters Model 481 LC spectrometer detector set at 254 nm equipped with a Waters 740 data module integrator using a chiral column (Diacel Chiracel OD-H). The flow rate was 0.5 mL/min, utilizing a 20:1 hexane:2-propanol mobile phase; ee = 80%: mp = 97−103 °C; $[\alpha]_D$ +36° (c = 0.32, $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$) δ −0.16 (s, 3H, $SiCH_3$), 0.04 (s, 3H, $SiCH_3$), 0.17 (s, 6H, 2 × $SiCH_3$), 0.67 (s, 9H, $(CH_3)_3$), 0.98 (s, 9H, $(CH_3)_3$), 4.73 (d, J = 4.7 Hz, 1H, H4), 5.01 (dd, J = 2.8, 4.6 Hz, 1H, H3), 6.10 (br s, 1H, NH), 6.82 (d, J = 8.4 Hz, 2H, aryl), 7.17 (d, J = 8.4 Hz, 2H, aryl); ^{13}C NMR (125 MHz, $CDCl_3$) δ −5.4, −4.9, −4.5, 25.4, 25.7, 58.8, 79.5, 119.8, 128.9, 129.2, 155.7, 169.7; FAB HRMS m/z calcd for $(M + H)^+$ $C_{21}H_{38}NO_3Si_2$ 408.2387, found 408.2366.

(3*R*,4*S*)-*N*-Benzoyl-3-[(*tert*-butyldimethylsilyl)oxy]-4-[4-[(*tert*-butyldimethylsilyl)oxy]phenyl]-2-azetidinone (7). To a cold (0 °C) solution of β-lactam **6** (220 mg, 0.54 mmol), triethylamine (0.5 mL, 2.5 mmol), and DMAP (catalytic amount) in anhydrous CH_2Cl_2 (2 mL) was added benzoyl chloride (0.3 mL, 1.8 mmol). The reaction mixture was stirred for 1 h at room temperature, and then the reaction was quenched with saturated aqueous NH_4Cl solution (10 mL); the mixture was diluted with CH_2Cl_2 (50 mL), washed with a saturated aqueous $NaHCO_3$ solution (20 mL) and brine (50 mL), and dried over anhydrous $MgSO_4$. The desiccant was removed by filtration and the solvent evaporated under reduced pressure. Purification of the crude product by silica gel flash chromatography (EtOAc:hexane = 5:95) gave 60% yield of compound **7** as a white solid: mp = 85−88 °C; $[\alpha]_D$ +130° (c = 0.70, $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$) δ −0.11 (s, 3H, $SiCH_3$), −0.08 (s, 3H, $SiCH_3$), 0.17 (s, 6H, 2 × $SiCH_3$), 0.72 (s, 9H, $(CH_3)_3$), 0.97 (s, 9H, $(CH_3)_3$), 5.09 (d, J = 6.1 Hz, 1H, H4), 5.36 (d, J = 6.1 Hz, 1H, H3), 6.82 (d, J = 8.5 Hz, 2H, aryl), 7.22 (d, J = 8.5 Hz, 2H, aryl), 7.50 (t, J = 7.8 Hz, 2H, aryl), 7.59 (t, J = 7.8 Hz, 1H, aryl), 8.00 (d, J = 7.1 Hz, 2H, aryl); ^{13}C NMR (125 MHz, $CDCl_3$) δ −5.4, −5.0, −4.5, −4.5, 17.9, 18.2, 25.3, 25.7, 60.4, 76.4, 119.9, 126.5, 128.1, 129.3, 129.9, 132.1, 133.3, 155.8, 165.4, 166.3; FAB HRMS m/z calcd for $(M + H)^+$ $C_{28}H_{41}NO_4Si_2$ 511.2574, found 511.2551.

2′-*O*-(*tert*-Butyldimethylsilyl)-3′-[4-[(*tert*-butyldimethylsilyl)oxy]phenyl]-3′-dephenyl-7-*O*-(triethylsilyl)paclitaxel (9). 7-(Triethylsilyl)baccatin III (**8**; 44 mg, 0.063 mmol) was dissolved in dry THF (1.5 mL). The solution was cooled to 0 °C, and NaH (63 mg, 60% mineral oil dispersion, 1.57 mmol) was added. The suspension was stirred for 30 min at 0 °C, and then a solution of *N*-benzoyl-2-azetidinone **7** (89 mg, 0.18 mmol) in THF (2 mL) was added through a cannula. The reaction mixture was stirred at 35 °C for 4 h, and then the reaction was quenched with cold brine (5 mL); the mixture was diluted with diethyl ether (70 mL), washed with brine (2 × 50 mL), and dried over anhydrous $MgSO_4$. The suspension was filtered and the filtrate evaporated to dryness under reduced pressure. Purification by silica gel flash column chromatography (EtOAc:hexane = 1:9) provided 61 mg (80% yield) of compound **9** as a white solid: mp = 105−115 °C; $[\alpha]_D$ −48° (c = 2.0, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ −0.24 (s, 3H, $SiCH_3$), −0.01 (s, 3H, $SiCH_3$), 0.18 (s, 6H, 2 × $SiCH_3$),

0.55−0.62 (m, 6H, Si(CH_2CH_3)$_3$), 0.82 (s, 9H, Si(CH_3)$_3$), 0.93 (t, J = 7.8 Hz, 9H, Si(CH_2CH_3)$_3$), 0.99 (s, 9H, Si(CH_3)$_3$), 1.17 (s, 3H, H17), 1.22 (s, 3H, H16), 1.70 (s, 3H, H19), 1.91 (m, 1H, H6), 2.02 (s, 3H, H18), 2.13 (dd, J = 9.4, 15.5 Hz, 1H, H14), 2.17 (s, 3H, 10-OAc), 2.40 (dd, J = 9.6, 15.2 Hz, 1H, H14), 2.49−2.56 (m, 1H, H6), 2.57 (s, 3H, 4-OAc), 3.83 (d, J = 6.9 Hz, 1H, H3), 4.22 (d, J = 8.3 Hz, 1H, H20), 4.32 (d, J = 8.3 Hz, 1H, H20), 4.47 (dd, J = 6.8, 10.6 Hz, 1H, H7), 4.62 (d, J = 2.0 Hz, 1H, H2′), 4.95 (d, J = 9.2 Hz, 1H, H5), 5.7 (m, 2H, H3′, H2), 6.23 (t, J = 9.1 Hz, 1H, H13), 6.45 (s, 1H, H10), 6.84 (d, J = 8.4 Hz, 2H, *m*-aryl), 7.04 (d, J = 9.0 Hz, 1H, NH), 7.17 (d, J = 8.3 Hz, 2H, *o*-aryl), 7.39 (t, J = 7.7 Hz, 2H, *m*-PhCON), 7.49 (t, J = 7.4 Hz, 1H, *p*-PhCON), 7.51 (t, J = 7.7 Hz, 2H, *m*-PhCO$_2$), 7.60 (t, J = 7.3 Hz, 1H, *p*-PhCO$_2$), 7.72 (d, J = 7.0 Hz, 2H, *o*-PhCON), 8.13 (d, J = 7.0 Hz, 2H, *o*-PhCO$_2$); ^{13}C NMR (125 MHz, $CDCl_3$) δ −5.3, −4.7, −3.9, 5.3, 6.7, 10.1, 14.2, 18.2, 18.2, 20.8, 21.5, 23.1, 25.6, 26.7, 26.6, 35.7, 37.3, 43.4, 46.7, 55.1, 58.5, 71.3, 72.3, 75.0, 75.2, 78.9, 81.27, 84.3, 120.3, 127.0, 127.5, 128.7, 128.7, 129.4, 130.2, 131.1, 131.7, 133.5, 133.8, 134.3, 140.2, 155.5, 166.9, 167.0, 169.2, 170.1, 171.4, 201.6; FAB HRMS m/z calcd for (M + H)$^+$ $C_{65}H_{94}NO_{15}Si_3$ 1212.5931, found 1212.5946.

3′-Dephenyl-3′-(4-hydroxyphenyl)paclitaxel (2). The protected taxane **9** (30 mg, 0.025 mmol) was dissolved in cold (0 °C) pyridine (1 mL). To that solution was added pyridinium hydrogen fluoride (0.2 mL) at 0 °C. The reaction mixture was stirred for 3 h at 0 °C and for 5 h at room temperature. Excess reagent was destroyed with saturated aqueous $NaHCO_3$ (15 mL), and the reaction mixture was diluted with diethyl ether (50 mL) and washed with an aqueous HCl solution (3%, 5 mL) and brine (30 mL). The organic layer was separated, dried over anhydrous $MgSO_4$, filtered, and evaporated under reduced pressure. Recrystallization (CH_2Cl_2/*n*-pentane) of the crude product gave compound **2** in 91% yield (19 mg) as a colorless solid: mp = 184−189 °C; $[\alpha]_D$ −54° (c = 0.7, $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$ + D_2O) δ 1.13 (s, 3H, H17), 1.20 (s, 3H, H16), 1.67 (s, 3H, H19), 1.76 (s, 3H, H18), 1.79−1.94 (m, 1H, H6), 2.22 (s, 3H, 10-OAc), 2.24−2.34 (m, 2H, H14), 2.35 (s, 3H, 4-OAc), 2.45−2.56 (m, 1H, H6), 3.76 (d, J = 6.8 Hz, 1H, H3), 4.18 (d, J = 8.3 Hz, 1H, H20), 4.28 (d, J = 8.3 Hz, 1H, H20), 4.36 (dd, J = 6.8, 10.7 Hz, 1H, H7), 4.71 (d, J = 2.6 Hz, 1H, H2′), 4.92 (d, J = 9.3 Hz, 1H, H5), 5.63−5.67 (m, 2H, H3′, H2), 6.17 (t, J = 8.4 Hz, 1H, H13), 6.26 (s, 1H, H10), 6.75 (d, J = 7.9 Hz, 2H, *m*-aryl), 7.23 (d, J = 8.3 Hz, 2H, *o*-aryl), 7.38 (t, J = 7.4 Hz, 2H, *m*-PhCON), 7.49 (t, J = 7.8 Hz, 2H, *m*-PhCO$_2$, 1H from *p*-PhCON), 7.59 (t, J = 8.8 Hz, 1H, *p*-PhCO$_2$), 7.73 (d, J = 7.3 Hz, 2H, *o*-PhCON), 8.10 (d, J = 7.4 Hz, 2H, *o*-PhCO$_2$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 9.6, 14.8, 20.8, 21.7, 22.6, 26.9, 35.7, 35.8, 43.2, 45.8, 54.9, 58.6, 72.1, 72.2, 73.4, 75.0, 75.6, 79.0, 81.3, 84.4, 115.9, 127.1, 128.4, 128.7, 129.2, 129.5, 130.2, 132.0, 133.3, 133.6, 133.7, 141.9, 156.1, 167.0, 167.4, 170.6, 171.2, 172.6, 203.6; FAB HRMS m/z calcd for (M + H)$^+$ $C_{47}H_{52}NO_{15}$ 870.3337, found 870.3327.

2′-*O*-(*tert*-Butyldimethylsilyl)-7-*O*-(triethylsilyl)paclitaxel (10). Paclitaxel (500 mg, 0.58 mmol), DMAP (720 mg, 5.8 mmol), and TBSCl (883 mg, 5.8 mmol) were dissolved in cold (0 °C) CH_2Cl_2 (5 mL) and stirred overnight at room temperature. The solution was cooled in an ice bath while TESCl (0.98 mL, 5.8 mmol) and DMAP (720 mg, 5.8 mmol) were added. After 30 min at room temperature, the reaction was quenched with brine (5 mL). The organic layer was collected and washed with dilute HCl (10%, 5 mL) and saturated aqueous $NaHCO_3$ solution (5 mL) and dried (Na_2SO_4). The crude residue, obtained by removal of solvent under reduced pressure, was purified by radial chromatography (silica gel, 2 mm, 7:3 hexane:EtOAc) to give 565 mg (90%) of the title compound as a colorless viscous oil: $[\alpha]_D$ −54° (c = 1.1, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ −0.32(s, 3H, SiCH$_3$), −0.22 (s, 3H, SiCH$_3$), 0.55−0.61 (m, 6H, Si(CH_2CH_3)$_3$), 0.80 (s, 9H, Si(CH_3)$_3$), 0.93 (t, J = 8.1 Hz, 9H, Si(CH_2CH_3)$_3$), 1.16 (s, 3H, H17), 1.22 (s, 3H, H16), 1.70 (s, 3H, H19), 1.89−1.94 (m, 1H, H6), 2.02 (s, 3H, H18), 2.10 (dd, J = 9.0, 15.4 Hz, 1H, H14), 2.17 (s, 3H, 10-OAc), 2.40 (dd, J = 9.7, 15.3 Hz, 1H, H14), 2.50−2.55 (m, 1H, H6), 2.58 (s, 3H, 4-OAc), 3.84 (d, J = 7.0 Hz, 1H, H3), 4.21 (d, J = 8.4 Hz, 1H, H20), 4.32 (d, J = 8.4 Hz, 1H, H20), 4.47 (dd, J = 6.6, 10.4 Hz, 1H, H7), 4.67 (d, J = 2.0 Hz, 1H, H2′), 4.95 (d, J = 8.3 Hz, 1H, H5), 5.70 (d, J = 7.1 Hz, 1H, H2), 5.74 (d, J = 8.8 Hz, 1H, H3′), 6.26 (t, J = 8.9 Hz, 1H, H13), 6.45 (s, 1H, H10), 7.07 (d, J = 8.9 Hz, NH), 7.30−7.61 (m, 10H, aryl), 7.74 (d, J = 7.7 Hz, 2H, aryl), 8.13 (d, J = 7.4 Hz, 2H, aryl); ^{13}C NMR (125 MHz, $CDCl_3$) δ −5.9, −5.2, 5.3, 6.7, 10.1, 14.3, 18.1, 20.9, 21.5, 23.1, 25.5, 25.7, 26.6, 35.6, 37.2, 43.3, 46.7, 55.6, 58.4, 71.4, 72.2, 74.9, 75.0, 75.1, 76.6, 78.8, 81.2, 84.2, 126.4, 127.0, 127.9, 128.7, 128.7, 129.2, 130.2, 131.8, 133.6, 133.7, 134.1, 138.3, 140.2, 166.9, 167.1, 169.3, 170.2, 171.4, 201.6; FAB HRMS m/z calcd for (M + Li)$^+$ $C_{59}H_{79}NO_{14}Si_2Li$ 1088.5199, found 1088.5243.

2′-*O*-(*tert*-Butyldimethylsilyl)-2-*O*-debenzoyl-7-*O*-(triethylsilyl)paclitaxel (11). A solution of anhydrous KOH was prepared immediately prior to use by adding H_2O (4 μL) to a 20-fold dilution of commercial *t*-BuOK in THF solution (0.05 M *t*-BuOK, 5 mL).[31] An aliquot of this solution (1.6 mL, 0.072 M KOH) was then added to a solution of **10** (65 mg, 0.06 mmol) in THF (4 mL) immersed in an CH_3CN/CO_2 bath. After the addition, the temperature was raised to −15 °C for 12 h, the reaction was quenched with a saturated aqueous NH_4Cl solution (5 mL), and then the mixture was warmed to room temperature. Extraction between diethyl ether (50 mL) and brine (30 mL) followed by drying (Na_2SO_4) of the collected organic phase resulted in crude product which was subjected to silica gel flash column chromatography (hexane:EtOAc = 4:1 to 1:1). Along with 15% (8 mg) of the dideacylated product 2′-*O*-(*tert*-butyldimethylsilyl)-4-*O*-deacetyl-2-*O*-debenzoyl-7-*O*-(triethylsilyl)paclitaxel,[23] compound **11** was isolated in 81% yield (47 mg) as an amorphous solid: $[\alpha]_D$ −54° (c = 0.67, CH_2Cl_2); ^{1}H NMR (500 MHz, CD_3OD) −0.12 (s, 3H, SiCH$_3$), −0.01 (s, 3H, SiCH$_3$), 0.56−0.66 (m, 6H, SiCH_2CH$_3$), 0.83 (s, 9H, C(CH$_3$)$_3$), 0.95 (t, J = 7.9 Hz, 9H, SiCH$_2$CH_3), 1.07 (s, 3H, H17), 1.14 (s, 3H, H16), 1.63 (s, 3H, H19), 1.80−1.87 (m, 1H, H6), 1.85 (s, 3H, H18), 1.99 (dd, J = 8.8, 15.0 Hz, 1H, H14), 2.13 (s, 3H, 10-OAc), 2.24 (dd, J = 9.5, 15.3 Hz, 1H, H14), 2.46 (s, 3H, 4-OAc), 2.52−2.58 (m, 1H, H6), 3.50 (d, J = 6.7 Hz, 1H, H2), 3.88 (d, J = 6.8 Hz, 1H, H3), 4.50 (dd, J = 10.5, 6.6 Hz, 1H, H7), 4.63 (q_{ab}, J = 8.9, 26 Hz, 2H, H20), 4.78 (d, J = 4.1 Hz, 1H, H2′), 5.01 (d, J = 8.0 Hz, 1H, H5), 5.72 (d, J = 4.0 Hz, 1H, H3′), 6.13 (t, J = 9.1 Hz, 1H, H13), 6.39 (s, 1H, H10), 7.32−7.58 (m, 8H, Bz, Ph), 7.77−7.80 (m, 2H, Bz); FAB HRMS m/z calcd for (M + H)$^+$ $C_{52}H_{75}NO_{13}Si_2$ 978.4855, found 978.4870.

2-*O*-[3-(Benzyloxy)benzoyl]-2-*O*-(debenzoyl)paclitaxel (13). To a stirring solution of **11** (43 mg, 0.044 mmol) in methylene chloride (2 mL) was added, in the following order, (3-benzyloxy)acetic acid (120 mg, 0.528 mmol), DMAP (64 mg, 0.528 mmol), and DCC (109 mg, 0.528 mmol). This solution was refluxed for 5 h or until the disappearance of the starting material. After cooling, the reaction mixture was diluted with methylene chloride (50 mL) and filtered. The filtrate was washed consecutively with 1 N HCl (10 mL), $NaHCO_3$ (10 mL), and brine (10 mL) and evaporated under reduced pressure. The crude product was dissolved in pyridine (0.5 mL), and the solution was cooled to 0 °C. To that mixture was added pyridinium hydrogen fluoride (0.1 mL). The reaction mixture was stirred at 0 °C for 3 h and at room temperature for another 5 h. Excess reagent was destroyed with saturated aqueous $NaHCO_3$ (5 mL), and the reaction mixture was diluted with diethyl ether (50 mL) and washed with 3% aqueous HCl (10 mL) and brine (10 mL). The organic layer was separated, dried over anhydrous $MgSO_4$, filtered, and evaporated under reduced pressure. Recrystallization (CH_2Cl_2/*n*-pentane) of the crude product gave compound **13** in 51% yield as a white solid: mp = 151−155 °C; $[\alpha]_D$ −37° (c = 0.4, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ 1.15 (s, 3H, H17), 1.23 (s, 3H, H16), 1.69 (s, 3H, H19), 1.80 (s, 3H, H18), 1.86−1.91 (m, H6), 2.22−2.38 (m, 2H, H14), 2.24 (s, 3H, 10-OAc), 2.36 (s, 3H, 4-OAc), 2.52−2.58 (m, 1H, H6), 3.80 (d, J = 7.0 Hz, 1H, H3), 4.20 (d, J = 8.5 Hz, 1H, H20), 4.35 (d, J = 8.5 Hz, 1H, H20), 4.38−4.46 (m, 1H, H7), 4.79 (dd, J = 2.5, 4.9 Hz, 1H, H2′), 4.95 (d, J = 8.5 Hz, 1H, H5), 5.11 (d, J = 11.3 Hz, 1H, CH$_2$Ph), 5.21 (d, J = 11.3 Hz, 1H, CH$_2$Ph), 5.67 (d, J = 7.0 Hz, 1H, H2), 5.79 (d, J = 8.9 Hz, 1H, H3′), 6.24 (t, J = 9.0 Hz, 1H, H13), 6.27 (s, 1H, H10), 6.93 (d, J = 8.9 Hz, NH), 7.21−7.23 (m, 1H, aryl), 7.34−7.43 (m, 14H, aryl), 7.71−7.79 (m, 4H, aryl); ^{13}C NMR (125 MHz,

$CDCl_3$) δ 9.6, 14.9, 20.9, 21.8, 22.6, 26.9, 35.7, 43.2, 45.6, 54.9, 58.6, 70.2, 72.2, 72.4, 73.0, 75.1, 75.6, 77.3, 79.0, 81.1, 84.4, 115.4, 121.2, 127.0, 127.6, 128.2, 128.3, 128.6, 128.7, 129.0, 129.8, 130.4, 132.0, 133.2, 133.6, 136.4, 137.9, 142.0, 158.9, 166.8, 167.0, 170.3, 171.3, 172.8, 203.6; FAB HRMS *m/z* calcd for $(M + H)^+$ $C_{54}H_{58}N_1O_{15}$ 960.3806, found 960.3850.

2-*O*-Debenzoyl-2-*O*-(3-hydroxybenzoyl)paclitaxel (3). To a solution of **13** (7 mg, 0.007 mmol) in a mixture of cyclohexene (0.5 mL) and ethanol (0.5 mL) was added 10% Pd/C (5 mg). The suspension was brought up to 60 °C and stirred for 2 h. After cooling, the Pd/C was removed by filtration through Celite and the solvent evaporated under reduced pressure. Purification of the reaction mixture by flash column chromatography on silica gel (EtOAc:hexane = 6:4) gave the final product **13** in quantitative yield (6.3 mg) as a white solid: mp = 169−172 °C; $[\alpha]_D$ +56° (c = 0.2, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ 1.09 (s, 3H, H17), 1.25 (s, 3H, H16), 1.71 (s, 3H, H19), 1.90 (s, 3H, H18), 2.07−2.12 (m, H6), 2.25 (s, 3H, 10-OAc), 2.46 (s, 3H, 4-OAc), 2.50−2.61 (m, 3H, H6, H14), 3.83 (d, J = 7.8 Hz, 1H, H3), 4.24 (d, J = 8.4 Hz, 1H, H20), 4.32 (d, J = 8.4 Hz, 1H, H20), 4.44−4.46 (m, 1H, H7), 4.90 (d, J = 7.7 Hz, 1H, H5), 4.95 (s, 1H, H2′), 5.54 (d, J = 7.8 Hz, 1H, H2), 6.05 (d, J = 10.1 Hz, 1H, H3′), 6.25 (s, 1H, H10), 6.48 (t, J = 9.1 Hz, 1H, H13), 7.09 (dd, J = 1.7, 8.1 Hz, 1H, 4-arylCO_2), 7.13 (d, J = 9.9, NH), 7.33 (t, J = 8.0 Hz, 1H, 5-arylCO_2), 7.38 (t, J = 8.5 Hz, 1H, *p*-Ph), 7.40−7.45 (m, 4H, *o*- and *m*-Ph), 7.47 (t, J = 7.3 Hz, 2H, *m*-PhCON), 7.50−7.57 (m, 2H, 6-arylCO_2, *p*-PhCON), 7.67 (d, J = 7.2 Hz, 2H, *o*-PhCON), 7.73 (s, 1H, 2-arylCO_2), 9.05 (br s, 1H, aryl OH); ^{13}C NMR (125 MHz, $CDCl_3$) δ 9.8, 14.7, 20.9, 22.6, 23.0, 26.6, 29.7, 35.3, 36.3, 43.0, 45.7, 53.7, 58.3, 72.0, 72.3, 72.4, 75.3, 75.5, 80.1, 80.8, 84.4, 116.5, 121.4, 121.5, 126.5, 127.0, 128.4, 129.1, 130.0, 130.6, 132.6, 133.1, 133.4, 137.4, 141.5, 157.3, 1667.0, 169.7, 170.4, 171.3, 172.8, 203.5; HRMS *m/z* calcd for $(M + H)^+$ $C_{47}H_{52}N_1O_{15}$: 870.3337, found 870.3325.

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Supporting Information Available: ^{1}H NMR spectra for compounds **2**, **3**, **5**−**7**, **9**−**11**, and **13** (9 pages). Ordering information is given on any current masthead page.

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