Chemical Synthesis and Biological Evaluation of C-2 Taxoids

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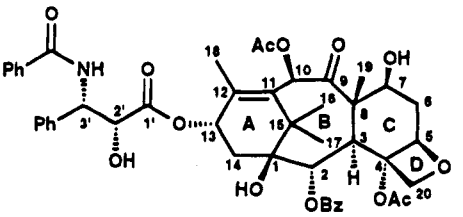
Received November 2, 1994[⊗]

Abstract: An efficient, general method for the synthesis of 1,2-hydroxy esters by regioselective nucleophilic opening of 1,2-cyclic carbonate systems has been developed. A reliable and practical route giving access to the taxoid carbonate **2** from the readily available 10-deacetylbaccatin III (**13**) has been established. Nucleophilic opening of the carbonate **2** with a variety of nucleophiles provided a number of novel C-2 Taxols. Water-soluble taxoid onium salts (e.g., **31e**, **31n**, **32**, and **34b**) were also synthesized and studied. A selected number of taxoids were subjected to cytotoxicity and tubulin polymerization assays as well as *in vivo* studies. The results of these studies are reported herein.

Introduction

Taxol (**1**, Figure 1), a highly functionalized diterpene originally isolated from the Pacific yew *Taxus brevifolia*,[1] is rapidly emerging as one of the most exciting anticancer drugs of the decade.[2] This compound is already in clinical usage for the treatment of breast and ovarian cancers[3] and is also showing promise for the treatment of lung,[4] skin,[5] and head and neck[6] cancers. Taxol exerts its action through a unique mechanism: it enhances the polymerization of tubulin into microtubules and stabilizes the resulting polymers.[7] The combination of these important and intriguing biological effects with Taxol's unusual molecular architecture created a high level of interest within the synthetic chemical community.[8] This interest culminated early in 1994 with reports by this group[9] and another[10] of two successful total syntheses of Taxol.

A number of the chemical studies of the taxoids have focused on elucidating the structure–activity relationship (SAR) of Taxol.[8,11] In general, those studies have shown that substitutions



1: Taxol

Figure 1. Structure of Taxol (**1**).

in the northern region of the molecule (C-7, -9, -10) have little effect upon its biological action, while those in the southern region (C-4, -2) and side chain (C-2′, 3′-aryl, -3′-acyl) can dramatically alter activity. Precise interpretation of these results is made more difficult by the lack of a clear conformational model for Taxol's structure with competing models existing for the structure of Taxol in aprotic media,[12] aqueous DMSO solution,[13] and water.[14]

One area of Taxol's SAR that has only recently been probed is the significance of the C-2 benzoate. Deletion of the C-2 oxygen[15] provides inactive compounds. However, recent studies by this group[16] and others[17] have shown that this position, while critical to activity, is tolerant of subtle changes. During the

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⊗ Abstract published in *Advance ACS Abstracts,* February 15, 1995.

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(10) Holton, R. A.; Somoza, C.; Kim, H.-B.; Liang, F.; Biediger, R. J.; Boatman, P. D.; Shindo, M.; Smith, C. C.; Kim, S.; Nadizadeh, H.; Suzuki, Y.; Tao, C.; Vu, P.; Tang, S.; Zhang, P.; Murthi, K. K.; Gentile, L. N.; Liu, J. H. *J. Am. Chem. Soc.* **1994**, *116*, 1597–1598. Holton, R. A.; Kim, H.-B.; Somoza, C.; Liang, F.; Biediger, R. J.; Boatman, P. D.; Shindo, M.; Smith, C. C.; Kim, S.; Nadizadeh, H.; Suzuki, Y.; Tao, C.; Vu, P.; Tang, S.; Zhang, P.; Murthi, K. K.; Gentile, L. N.; Liu, J. H. *J. Am. Chem. Soc.* **1994**, *116*, 1599–1600.

(11) Kingston, D. G. I *TiBTech* **1994**, *12*, 222–227.

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(13) Williams, H. J.; Scott, A. I.; Dieden, R. A.; Swindell, C. S.; Chirlian, L. E.; Francl, M. M.; Heerding, J. M.; Krauss, N. E. *Tetrahedron* **1993**, *49*, 6545–6560. Vander Velde, D. G.; Georg, G. I.; Grunewald, G. L.; Gunn, C. W.; Mitscher, L. A. *J. Am. Chem. Soc.* **1993**, *115*, 11650–11651.

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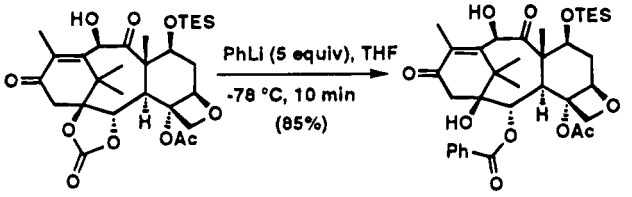
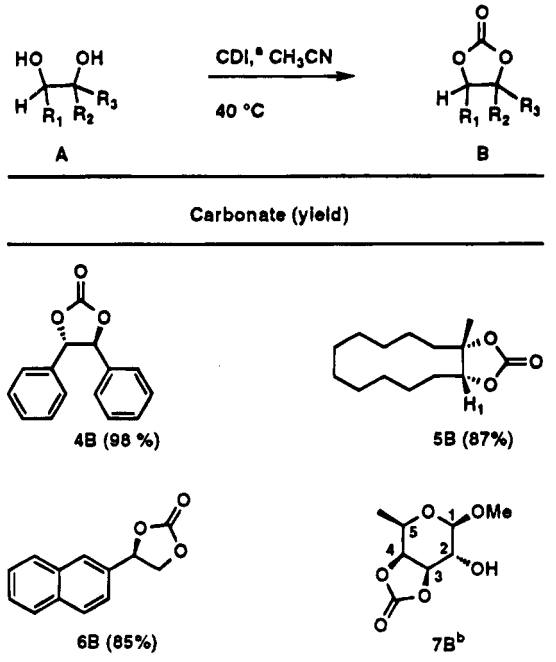


Figure 2. Reaction of PhLi with carbonate **2**. TES = $SiEt_3$.

Table 1. Preparation of Carbonates **4B**–**7B**



[a] 1,1′-Carbonyldiimidazole. [b] Groneberg, R. D.; Miyazaki, T.; Stylianides, N. A.; Schulze, T. J.; Stahl, W.; Schreiner, E. P.; Suzuki, T.; Iwabuchi, Y.; Smith, A. L.; Nicolaou, K. C. *J. Am. Chem. Soc.* **1993**, *115*, 7593.

course of our studies on the chemistry of Taxol, it was discovered that treating cyclic carbonate **2** with phenyllithium provided hydroxy benzoate **3** with high regio- and chemoselectivity (Figure 2). Given the paucity of reports of nucleophilic opening of carbonates,[18] this was a somewhat surprising but fortunate result. We disclose herein the full details of our studies on the exploration of the nucleophilic opening of cyclic carbonates and its application to the synthesis of C-2 derivatives of Taxol. This report also addresses preliminary studies on the generality of nucleophilic addition to cyclic carbonates and details biological studies with a number of selected taxoids.

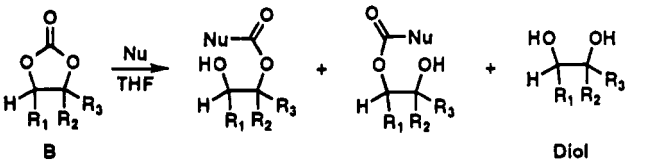
Results and Discussion

This program centered upon developing methodology to access new Taxol analogs with modified C-2 substituents in a manner that would afford such taxoids from readily available precursors. Below we describe the evolution of the necessary methodology and the construction of a series of novel taxoids for biological evaluation.

Establishing the Methodology. In order to explore the generality and scope of the carbonate ring opening reaction encountered in the Taxol project, a series of five-membered cyclic carbonates covering a diverse range of structures were synthesized from the corresponding diols as summarized in Table 1.

(18) Satyanarayana, G.; Sivaram, S. *Synth. Commun.* **1990**, *20*, 3273–3276. Wender, P. A.; Kogen, H.; Lee, H. Y.; Munger, J. D.; Wilhelm, R. S.; Williams, P. D. *J. Am. Chem. Soc.* **1989**, *111*, 8957–8958.

Table 2. Regioselective Synthesis of Esters via Nucleophilic Addition to Cyclic Carbonates



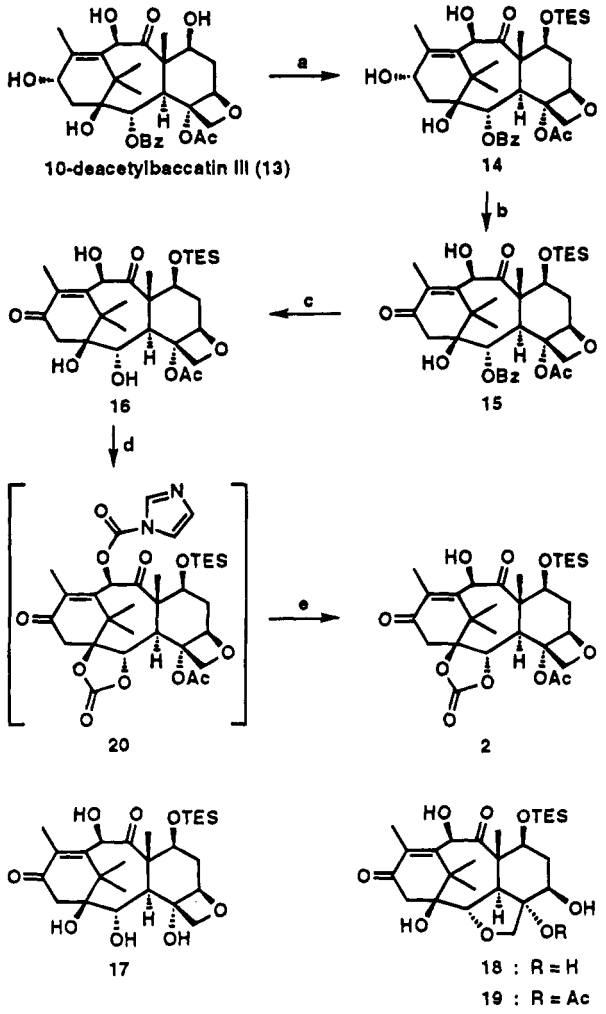
Entry	Carbonate	Products	Nucleophile (NuLi)	Yield Ester : Diol
1	4B		a	78 : 16[a] 70 : 26[b]
2	4B	8a-d	b	98 : 0
3	4B		c	87 : 0
4	4B		d Ph–≡–Li	100 : 0
5	5B		a	85 : 12[c]
6	5B	9a,b	b	96 : 2[d]
7	6B	10 + 11	b	68 : 0 (10) 17 : 0 (11)
8	7B	12	a	93 : 0

[a] Reaction carried out at −78 °C, using 1.25 equiv of PhLi; the yield is based on 92% conversion. [b] Reaction carried out at −98 °C, using 1.5 equiv of PhLi. [c] Yield based on 58% conversion. [d] Yield based on 91% conversion.

Carbonates **4B**–**7B** were then subjected to ring opening with a variety of organolithium reagents (Table 2). Thus, compound **4B** reacted smoothly with reagents **a**–**d** (Table 2) to afford **8a**–**d** as the major products (Table 2, entries 1–4), demonstrating the generality of the method with respect to nucleophiles. In some instances, the diol was formed as a minor product (e.g., entry 1), presumably via double nucleophilic attack or hydroxide ion-induced hydrolysis.

The regiochemistry of the opening was examined by treating compounds **5B**–**7B** with phenyllithium and 2-thienyllithium as shown in Table 2. In the case of carbonate **5B** (entries 5 and

Scheme 1[a]



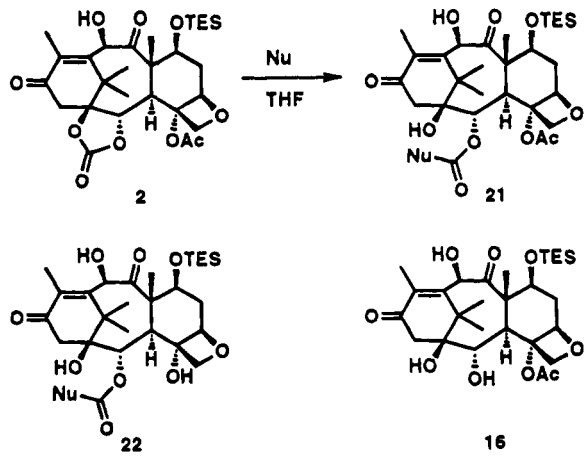
[a] Synthesis of taxoid carbonate **2**. Reagents and conditions: (a) 20 equiv of Et_3SiCl, pyridine, 25 °C, 17 h, 91%; (b) 1.0 equiv of 4-methylmorpholine *N*-oxide (NMO), 0.05 equiv of tetrapropylammonium perruthenate (TPAP), CH_2Cl_2, 25 °C, 2 h, 96%; (c) 10 equiv of K_2CO_3, MeOH, H_2O, 0 °C, 2.5 h, 93% based on 81% conversion; (d) 6.0 equiv of 1,1′-carbonyldiimidazole (CDI), THF, 40 °C, 0.5 h; (e) 1 N aqueous HCl, THF, 25 °C, 15 min, 93%. TES = $SiEt_3$, Bz = COPh.

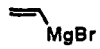
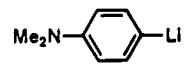
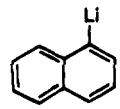
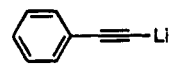
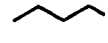
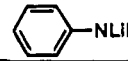
6), the reaction proceeded with high regiochemical control leading to the secondary esters as the predominant products. Entry 7 (Table 2) demonstrated the preference for the primary ester while entry 8 showed that the ring opening proceeded with remarkable selectivity to afford a single ester in high yield. It is of note that the latter example from the carbohydrate field represents a net result of forming, regioselectively, a monobenzoate from a triol in two steps.

Synthesis and Opening Reactions of a Taxoid Cyclic Carbonate. Having established the generality and scope of the carbonate ring opening in model systems, we then proceeded to apply it to the taxoid field. The first objective was to secure a reliable and practical route to a suitable taxoid C-1,2-carbonate. To this end, compound **2** was chosen as a suitable candidate since both its behavior toward phenyllithium and accessibility from natural sources were already established.[16] A new, more efficient route to access compound **2** from 10-deacetylbaccatin III (10-DAB, **13**) was sought and found as summarized in Scheme 1. Thus, selective protection of the C-7 hydroxyl group with a triethylsilyl (TES) group according to Greene's procedure[19] proceeded smoothly to afford compound **14** (91%), which

(19) Denis, J. N.; Greene, A. E.; Guénard, D.; Guéritte-Voegelein, F.; Mangatal, L.; Potier, P. *J. Am. Chem. Soc.* **1988**, *110*, 5917–5919.

Table 3. Synthesis of C-2 Taxol Analogs



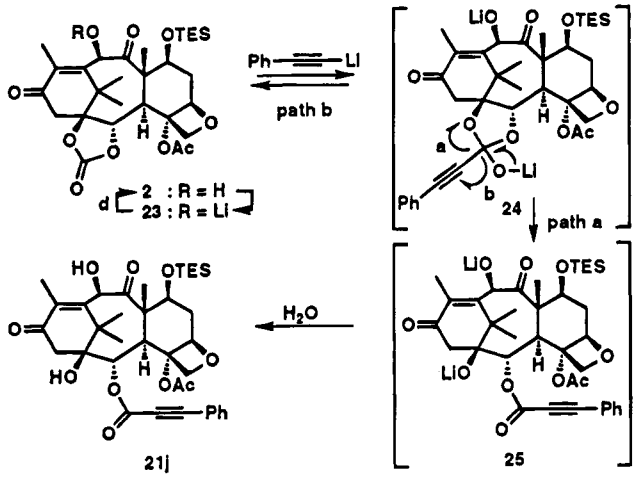
Entry	Nucleophile		Yield of products 21 : 22 : 16
1		a	72 : 19 : 0
2		b	52 : 24 : 0 74[e]: 0 : 0
3		b	60 : 0 : 0
4		c	78[b]: 0 : 0
5		d	96[c]: 0 : 0
6		e	83[c]: 10: 0
7		f	39 : 13 : 27
8		g	57[c]: 0 : 0
9		h	30 : 29 : 0
10		i	89[b]: 0 : 0
11		j	95[d]: 0 : 0
12		k	92 : 0 : 0
13		l	93[c]: 0 : 0

[a] Reaction conducted on 0.01 mmol scale at 70% conversion. [b] Crude yield. [c] Yield based on 50–85% conversion. [d] Yield based on 10–20% conversion.

in turn underwent chemoselective oxidation at C-13 with TPAP–NMO,[20] leading to diketone **15** (96%). Hydrolysis of

(20) Ley, S. V.; Norman, J.; Griffith, W. P.; Marsden, S. P. *Synthesis* **1994**, 639–666.

Scheme 2[a]



[a] Reversibility of the lithium phenylacetylide addition to carbonate **2**.

the C-2 benzoate group of **15** was achieved using K_2CO_3 in wet methanol at 0 °C furnishing triol **16** in 93% yield (81% conversion). Higher temperatures led to byproducts such as compounds **17** (deacetylation), **18** (intramolecular oxetane opening), and **19** (intramolecular oxetane opening and deacetylation) (Scheme 1).[21] Exposure of triol **16** to 1,1′-carbonyldiimidazole led to the formation of the cyclic carbonate–imidazolide **20**, which was smoothly and selectively hydrolyzed under mildly acidic conditions to the targeted compound **2**, in 93% overall yield from **16** (Scheme 1). Thus, **2** is available in approximately 60% overall yield from commercially available 10-DAB (**13**).

Having secured a practical route for large scale synthesis of carbonate **2**, we next examined its reaction with a variety of nucleophilic reagents. Table 3 lists the results of this study. In general, these reactions proved to be high-yielding and regiospecific, leading to the C-2 ester derivatives (**21**, Table 3). Occasional byproducts included the C-4 deacetyl system **22** (entries 1, 2, 6, 7 and 9, Table 3) and the triol **16** (entry 7, Table 3).

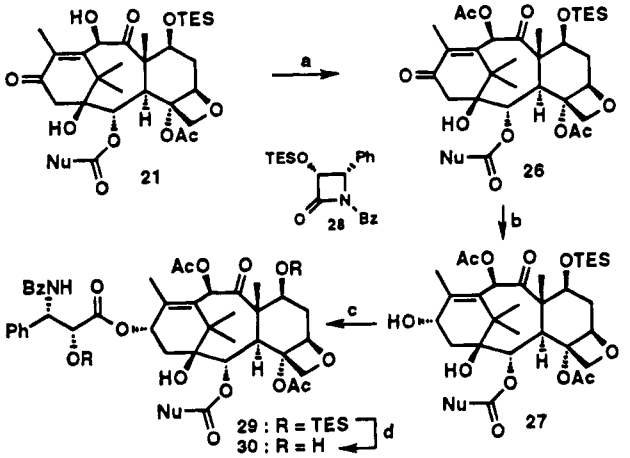
Some specific entries of Table 3 warrant additional comments. Reaction of 2-pyridyllithium with **2** resulted in a mixture of products and a modest yield of the desired ester **21f** (entry 7, Table 3) due to the latter's instability (hydrolysis). The synthesis of the much more stable 3-pyridyl ester **21g** (entry 8, Table 3) was also somewhat problematic due to instability of the required organolithium reagent at −78 °C. Lastly, the addition of lithium phenylacetylide to carbonate **2** proved quite interesting, in that only 10–20% conversion to product **21j** could be achieved. This rather clean reaction presumably suffers from reversibility, due to the highly stabilized nature of the acetylide anion involved (see Scheme 2).

Synthesis of New, C-2-Substituted Taxoids. With a variety of novel C-2-substituted taxoid intermediates in hand through the above chemistry, we undertook the conversion of a selected number of them to taxoids in order to evaluate their biological properties. The main task, of course, was the introduction of Taxol's side chain, after appropriate protection and reduction of the C-13 carbonyl.

Following a previously established route,[16] **21c–i** and **21m** were converted into taxoids **30c–i** and **30m** (see Scheme 3 and Table 4). Thus, selective acetylation of diols **21c–i** and **21m** under standard conditions (Ac_2O, DMAP) provided monoacetates **26c–i** and **26m** in good to excellent yields (Table 4). Subsequent reduction of the C-13 carbonyl group proceeded both chemo- and stereoselectively with $NaBH_4$ in methanol or methanol–THF to afford good to excellent yields of the corresponding allylic alcohols **27c–j** and **27m** (Scheme 3 and

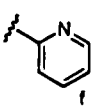
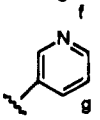
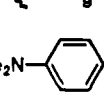
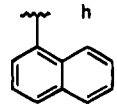
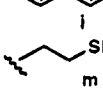
(21) Wahl, A.; Guéritte-Voegelein, F.; Guénard, D.; Le Goff, M.-T.; Potier, P. *Tetrahedron* **1992**, *48*, 6965–6974. Klein, L. L. *Tetrahedron Lett.* **1993**, *34*, 2047–2050. Farina, V.; Huang, S. *Tetrahedron Lett.* **1992**, *33*, 3979–3982.

Scheme 3[a]



[a] Synthesis of Taxols **30c–i** and **30m**. Reagents and conditions: (a) 36–98 equiv of Ac_2O, 3–10 equiv of 4-(dimethylamino)pyridine (DMAP), CH_2Cl_2, 25 °C, 1–3 h; (b) 15–100 equiv of $NaBH_4$, MeOH or MeOH–THF, 0 or 25 °C, 1–6 h; (c) 2.4–3 equiv of $NaN(SiMe_3)_2$, 1.7–3.5 equiv of β-lactam **28**, THF, 0 or −78 °C, 5–30 min; (d) HF·pyridine, THF, 25 °C, 1.0–3.75 h. TES = $SiEt_3$, Bz = COPh.

Table 4. Preparation of Taxols **30c–i** and **30m** from Diols **21c–i** and **21m**

Entry	Nu	Yield (%) of acetylation **21** ➤ **26**[a]	Yield (%) of reduction **26** ➤ **27**[a]	Yield (%) of coupling **27** ➤ **29**[a]	Yield (%) of deprotection **29** ➤ **30**[a]
1	 c	66[b]	83	26	64
2	 d	77	59[c]	43[c]	71
3	 e	91	89[c]	61[c]	56
4	 f	77	59	17	38
5	 g	95	67[c]	48[c]	75
6	Me_2N—  h	77	88[c]	57	84
7	 i	89[b]	48	62	86
8	 m	95	60	42	49

[a] See Scheme 3. [b] Overall yield from **2**. [c] Yield based on 79–94% conversion.

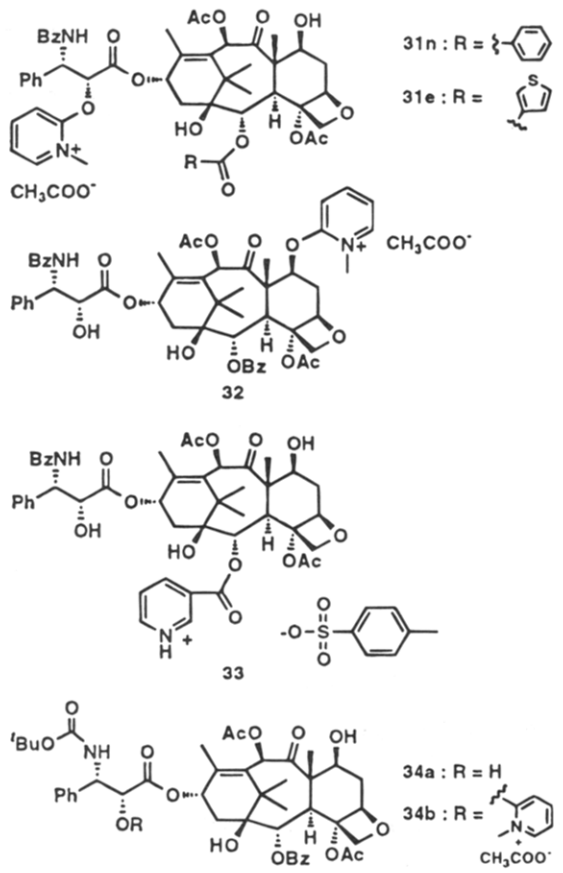


Figure 3. Structures of taxoid onium salts **31**−**34**.

Table 4). These alcohols were subjected to the Ojima−Holton protocol[22] with β-lactam **28** (Scheme 3) furnishing compounds **29c**−**i** and **29m**, respectively (Table 4). Finally, desilylation of these compounds with HF·pyridine resulted in the formation of the targeted taxoids **30c**−**i** and **30m**.

The phenylthio derivative **30m** (Scheme 3, Table 4) was smoothly prepared by conjugate addition of phenylthio to vinyl ester **21b** (Table 3). While the acetylation of the vinyl ester **21b** could be carried out, albeit in low yield, it was not possible to reduce the C-13 carbonyl group of the corresponding acetate due to competitive conjugate reduction of the vinyl group.

Synthesis of Water-Soluble Taxoid Onium Salts. In order to produce water-soluble taxoids,[23] the compounds shown in Figure 3 were targeted for synthesis. Compounds **31n**, **32**, and **34b** have already been reported from these laboratories.[14,23] Following similar procedures, compound **31e** was synthesized from **30e** by exposure to excess 2-fluoro-1-methylpyridinium *p*-toluenesulfonate in CH_2Cl_2 in the presence of Et_3N, followed by HPLC purification using an ammonium acetate buffer solution as eluent (anion exchange). The 3-pyridine ester derivative **33** was prepared by treatment of **30g** with stoichiometric amounts of TsOH.

As was the case with onium salts **31n**[23] and **32**,[14] compound **31e** self-assembles in aqueous solution to provide filamentous helical structures (Figure 4). This structural motif may lead to the high chemical stability that these highly water-soluble (>5 mM) compounds exhibit in aqueous solution.

(22) Holton, R. A. *Chem. Abstr.* **1990**, *114*, 164568q. Ojima, I.; Habus, I.; Zhao, M.; Georg, G. I.; Jayasinghe, L. R. *J. Org. Chem.* **1991**, *56*, 1681−1683. Ojima, I.; Habus, I.; Zhao, M.; Zucco, M.; Park, Y. H.; Sun, C. M.; Brigaud, T. *Tetrahedron* **1992**, *48*, 6985−7012.

(23) Nicolaou, K. C.; Guy, R. K.; Pitsinos, E. N.; Wrasidlo, W. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 1583−1587.

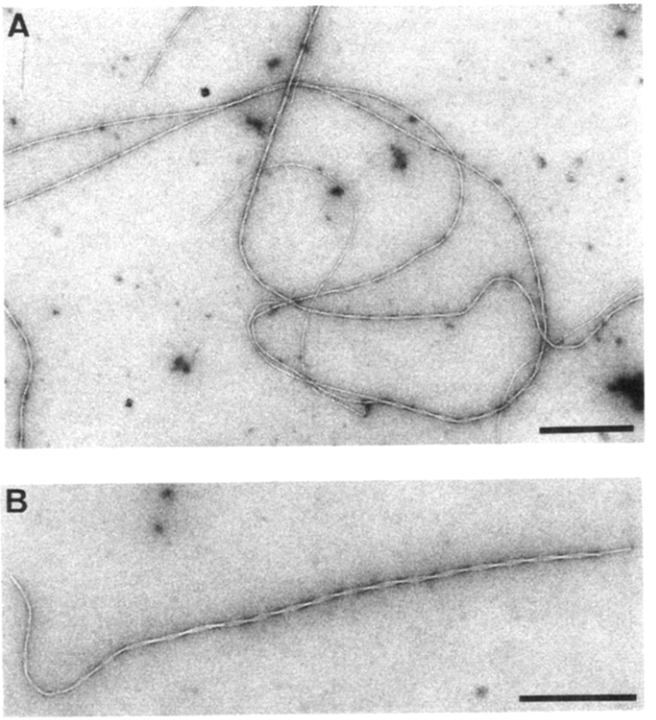


Figure 4. TEM (transmission electron microscope) image of negatively stained 3-thiophene 2′-MPA **31e**. Negative staining is performed by adding one drop of a solution of **31e** in 100 mM PBS (phosphate-buffered saline) onto a parlodion−carbon coated grid, followed by fixing in 2% glutaraldehyde in PBS, washing in water, and staining in 2% aqueous uranyl acetate. Panel A shows the filamentous form of **31e** and different degrees of aggregation as revealed by various thicknesses of the filaments. Panel B shows the helical nature (arrangement) within aggregates. Lines (scale) indicate 400 nm in both A and B.

Biological Results

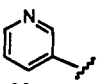
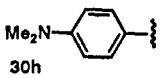
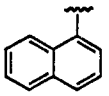
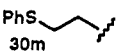
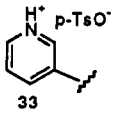
Microtubule Binding and Cytotoxicity Assays. The taxoids synthesized during this project were subjected to two types of biological evaluation: cytotoxicity assays and microtubule binding.

Cytotoxicities of the taxoids were determined using a panel of eight cell lines (Table 5). In general, the compounds can be grouped in three sets. Those lacking a simple unsubstituted aromatic system (**30h**,**i**,**m**) at the C-2 position show poor cytotoxicities. Of the aromatic esters, one set of compounds (**30c**,**d**,**f**,**g**) shows reasonable activity, relative to Taxol, but has cytotoxicities attenuated by 10−100-fold. Finally, the 3-thiophene ester (**30e**) consistently shows cytotoxicities as high as or higher than those of Taxol. The pyridinium salt **31e**, functioning as a prodrug in this assay by discharging its parent **30e**, gave the expected cytotoxicity profile. One should note that these data exhibit significant variability between both cell lines and compounds. These results may indicate subtle variations in receptor structure (i.e., microtubule isotype)[24] between cell lines.

Microtubule binding was established by challenging GTP-induced microtubules with calcium chloride in the presence of the test compound. With Taxol present, these microtubules are stable, whereas with no drug present they will depolymerize. In this study (Figure 5) those compounds which were modified on the side chain (**31e**, **31n**) did not prevent depolymerization.

(24) Ludueña, R. F. *Mol. Biol. Cell* **1993**, *4*, 445−457. Murphy, D. B. *Curr. Opin. Cell Biol.* **1991**, *3*, 43−51. Joshi, H. C.; Cleveland, D. W. *Motil. Cytoskel.* **1990**, *16*, 159−163. Lu, Q.; Ludueña, R. F. *Cell Struct. Funct.* **1993**, *18*, 173−182.

Table 5. Cytotoxicities of C-2 Taxol Analogs (**30c–i,m**)[a]

	IC_{50} Values [M] from XTT Assays							
Cell Type	Human T-cell leukemia	Mouse leukemia	Melanoma	Lung adenocarcinoma	Human promyelocytic leukemia	Human prostate adenocarcinoma	Human ovarian carcinoma	Human perirenal cell carcinoma
Cell Line	MOLT-4	L1210	SK-MEL 28	UCLA-P3	HL-60	PC-3	OVCAR-3	786-O
 Taxol (1)	6.21×10^{-10}	1.3×10^{-9}	1.39×10^{-9}	3.3×10^{-9}	1.8×10^{-9}	7.03×10^{-10}	7.13×10^{-9}	6.3×10^{-8}
 30c	4.5×10^{-8}	4.7×10^{-7}	8.4×10^{-8}	6.9×10^{-8}	2.5×10^{-8}	1.0×10^{-7}	9.9×10^{-4}	5.2×10^{-6}
 30d	3.7×10^{-9}	1.8×10^{-8}	1.2×10^{-8}	9.3×10^{-8}	1.2×10^{-8}	1.8×10^{-8}	5.8×10^{-9}	4.3×10^{-7}
 30e	6.7×10^{-10}	7.6×10^{-9}	3.8×10^{-9}	6.9×10^{-9}	1.2×10^{-10}	3.8×10^{-9}	2.5×10^{-9}	1.5×10^{-9}
 30f	4.24×10^{-8}	1.94×10^{-9}	2.33×10^{-8}	9.02×10^{-8}	2.47×10^{-7}	3.09×10^{-9}	9.9×10^{-9}	4.05×10^{-6}
 30g	7.84×10^{-9}	2.98×10^{-6}	3.04×10^{-7}	1.02×10^{-7}	8.00×10^{-7}	4.54×10^{-7}	4.29×10^{-7}	5.00×10^{-7}
 30h	2.2×10^{-6}	1.6×10^{-6}	1.2×10^{-4}	9.8×10^{-6}	1.3×10^{-5}	3.4×10^{-5}	1.1×10^{-5}	5.0×10^{-6}
 30i	1×10^{-4}	2.9×10^{-5}	1.0×10^{-4}	1.0×10^{-4}	1.0×10^{-4}	1.0×10^{-4}	1.0×10^{-4}	1.0×10^{-4}
 30m	1.37×10^{-6}	1.14×10^{-6}	8.50×10^{-6}	3.20×10^{-6}	1.0×10^{-4}	6.33×10^{-6}	3.08×10^{-5}	6.34×10^{-6}
C-2' MPA of Taxol 31n	7.07×10^{-9}	1.28×10^{-8}	8.79×10^{-7}	2.44×10^{-9}	2.46×10^{-10}	6.47×10^{-9}	7.49×10^{-9}	2.74×10^{-7}
C-2' MPA of 3-thiophene 31e	1.05×10^{-10}	1.07×10^{-9}	8.29×10^{-9}	1.67×10^{-9}	2.00×10^{-9}	6.07×10^{-9}	2.53×10^{-9}	9.07×10^{-9}
C-7 MPA of taxol 32	9.94×10^{-9}	3.53×10^{-8}	7.02×10^{-6}	1.28×10^{-8}	5.85×10^{-9}	7.07×10^{-8}	8.42×10^{-8}	4.54×10^{-6}
 33	2.72×10^{-8}	6.70×10^{-6}	1.64×10^{-7}	2.05×10^{-7}	3.47×10^{-7}	2.56×10^{-7}	1.23×10^{-7}	4.99×10^{-6}
Taxotere 34a	1.00×10^{-12}	5.90×10^{-10}	6.14×10^{-6}	1.15×10^{-10}	3.50×10^{-11}	4.06×10^{-10}	1.10×10^{-10}	4.15×10^{-8}
C-2' MPA of Taxotere 34b	3.22×10^{-8}	1.88×10^{-11}	6.82×10^{-8}	1.99×10^{-8}	8.99×10^{-10}	—	8.18×10^{-8}	5.55×10^{-10}

[a] See Experimental Section for further details.

Those compounds which contained simple aromatic esters at C-2 (**30d,e**, **32**) stabilized microtubules to calcium chloride induced depolymerization. Due to its stability in this assay, compound **31e** did not inhibit the depolymerization of microtubules. This is the expected behavior of a stable C-2 derivative. In this assay, the 3-thiophene ester **30e** registered a slight advantage, both in kinetics of assembly and in stability of microtubule (Figure 5), over the 2-thiophene ester **30d**.

***In Vivo* Studies.** Distal tumor model studies using athymic nude mice inflicted with PC-3 prostate carcinoma were carried out for the 2- and 3-thiophene ester derivatives **30d** and **30e**. Both compounds caused tumor growth inhibition significantly higher than controls and approximately equivalent to that normally exhibited by Taxol (Figure 6). Visual examination of test subjects also indicated toxic effects similar to those seen with Taxol. These studies also revealed that the 3-substituted

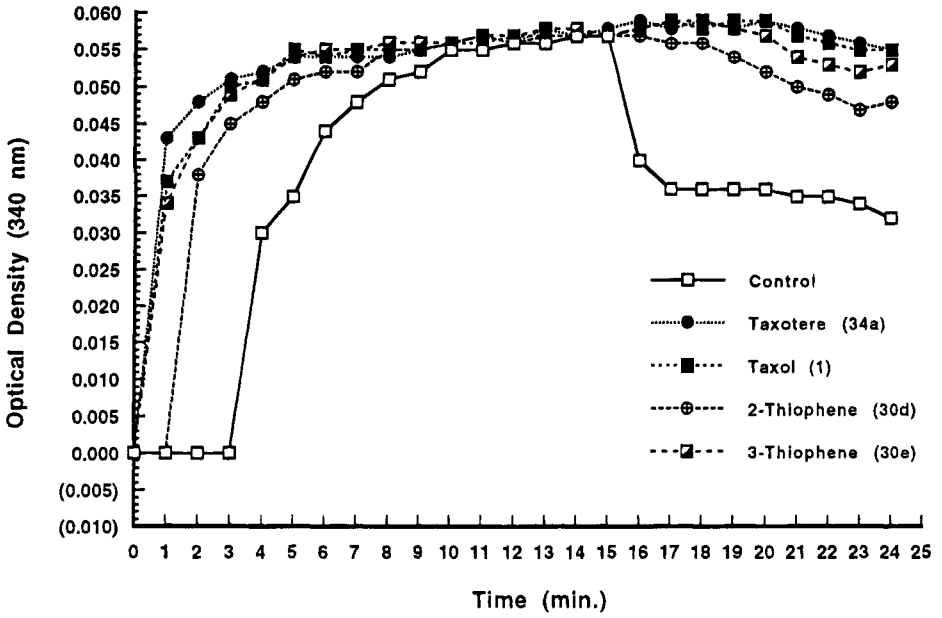


Figure 5. A comparison of the microtubule depolymerization inhibition ability of selected taxoids (see Experimental Section for further details).

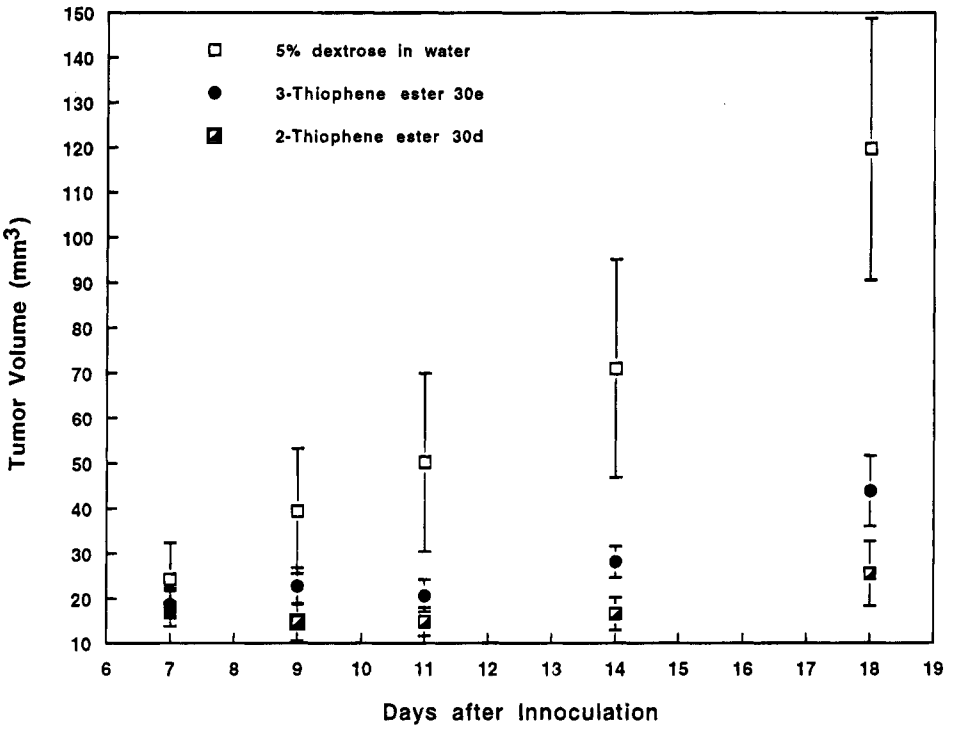


Figure 6. A comparison of the *in vivo* tumor inhibition efficacy of the thiophene esters **30d** and **30e** (see Experimental Section for further details).

derivative (**30e**) was significantly (at the 0.05 confidence limit) less active than the 2-substituted derivative (**30d**). The fact that this result is antipodal to the cytotoxicity and microtubule results, both in general and for the specific cell line, indicates that one must take extreme care in extrapolating from tubulin or *in vitro* cytotoxicity data to *in vivo* efficacy.

Of the various water-soluble taxoids synthesized during this study, the tosylate salt **33** had sufficiently low cytotoxicity to rule out further study. However, the 2′-MPA salt **31e**, prepared from **30e** according to a similar derivatization of Taxol,[23] exhibited promising *in vitro* results including high aqueous solubility (>5 mM), high aqueous stability (>10 days), and cytotoxicities equivalent to the parent compound **30e**. Unfortunately, distal tumor models (OVCAR-3, PC-3) revealed no *in vivo* activity for this compound, presumably due to a rather slow release of **30e** *in vivo* resulting in excretion.

Conclusion

The systematic studies described in this article establish the ring opening of cyclic carbonates by nucleophilic reagents as a viable and useful method for the regioselective synthesis of esters starting with 1,2-diol systems. This method found application in the total synthesis of Taxol[9] and a series of designed Taxols including water-soluble versions. These designed taxoids are now readily available from a common precursor (compound **2**) which can be obtained from 10-DAB (**13**) via semisynthesis.

The work presented in this report also provides some important insight into the character of Taxol's binding site. While the binding pocket is loose enough to allow the substitution of the phenyl ester with other aromatic groups, bulky substituents in the para position[17] or removal of the aromatic

group cause problematic interactions. These results, as well as subtle differences between the various aromatic esters, may point toward specific electrostatic and spacial interactions. Presumably, the preparation of other C-2 taxoids will allow a finer appraisal of the character of Taxol's receptor. In conclusion, the lack of congruity between our microtubule binding assays, cytotoxicity tests, and *in vivo* models strongly suggests that one must carry out *in vivo* studies in order to make meaningful claims about efficacy. This newly found understanding has also been supported by the recent results of Chen's group[25] with C-7 derivatives of Taxol. Thus, supposedly "set" views of Taxol's SAR, based solely on microtubule binding or cytotoxicity, are due some serious reconsideration.

Experimental Section

General Techniques. All reactions were carried out under an argon atmosphere with dry, freshly distilled solvents under anhydrous conditions, unless otherwise noted. Tetrahydrofuran (THF) and ethyl ether (Et_2O) were distilled from sodium–benzophenone; methylene chloride (CH_2Cl_2), benzene (PhH), and toluene from calcium hydride. Yields refer to chromatographically and spectroscopically (^1H NMR) homogeneous materials, unless otherwise stated. All solutions used in workup procedures are saturated unless otherwise noted. All reagents were purchased at highest commercial quality and used without further purification unless otherwise stated.

All reactions were monitored by thin-layer chromatography carried out on 0.25 mm E. Merck silica gel plates (60F-254) using UV light as visualizing agent and 7% ethanolic phosphomolybdic acid or *p*-anisaldehyde solution and heat as developing agents. E. Merck silica gel (60, particle size 0.040–0.063 mm) was used for flash column chromatography. Preparative thin-layer chromatography (PTLC) separations were carried out on 0.25 or 0.50 mm E. Merck silica gel plates (60F-254).

NMR spectra were recorded on Bruker AMX-500 or AM-300 instruments and calibrated using residual undeuterated solvent as an internal reference. The following abbreviations were used to explain the multiplicities: s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; band, several overlapping signals; br, broad. The carbon numbering of Taxol was used to assign protons. IR spectra were recorded on a Perkin-Elmer 1600 series FT-IR spectrometer. High-resolution mass spectra (HRMS) were recorded on a VG ZAB-ZSE mass spectrometer under fast atom bombardment (FAB) conditions.

Silyl Ether 14. To a solution of 10-deacetylbaccatin III (**13**, 3.0 g, 5.51 mmol, Indena, Italy) in pyridine (250 mL) was added chlorotriethylsilane (18.5 mL, 110 mmol) dropwise. The resulting solution was stirred at 25 °C for 17 h. After dilution with Et_2O (750 mL), the solution was washed with aqueous $CuSO_4$ (3 × 200 mL) and brine (200 mL). The organic layer was dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 35 → 50% EtOAc in petroleum ether) to give alcohol **14** (3.39 g, 91%) as a white solid: R_f = 0.32 (silica, 50% EtOAc in hexanes); IR (thin film) ν_{max} 3464, 2954, 2282, 1710, 1453, 1362, 1271, 1242, 1105, 994 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.06 (dd, J = 8.0, 0.9 Hz, 2H, Bz), 7.57 (t, J = 7.9 Hz, 1H, Bz), 7.44 (t, J = 7.9 Hz, 2H, Bz), 5.56 (d, J = 7.0 Hz, 1H, 2-H), 5.14 (d, J = 1.9 Hz, 1H, 10-H), 4.92 (d, J = 9.5 Hz, 1H, 5-H), 4.84–4.78 (m, 1H, 13-H), 4.37 (dd, J = 10.6, 7.0 Hz, 1H, 7-H), 4.27 (d, J = 8.5 Hz, 1H, 20-H), 4.25 (d, J = 1.9 Hz, 1H, 10-OH), 4.12 (d, J = 8.5 Hz, 1H, 20-H), 3.91 (d, J = 7.0 Hz, 1H, 3-H), 2.48–2.40 (m, 1H, 6-H), 2.25 (s, 3H, Me), 2.25–2.17 (m, 2H, 14-CH_2), 2.04 (s, 3H, Me), 1.90–1.82 (m, 1H, 6-H), 1.70 (s, 3H, Me), 1.03 (s, 6H, Me, Me), 0.90 (t, J = 8 Hz, 9H, $Si(CH_2CH_3)_3$), 0.58–0.42 (band, 6H, $Si(CH_2CH_3)_3$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 210.3, 170.8, 167.0, 141.8, 135.1, 133.6, 130.1, 129.4, 128.6, 84.2, 80.7, 78.8, 76.5, 74.8, 74.6, 72.9, 67.9, 57.9, 47.0, 42.7, 38.6, 37.2, 26.8, 22.6, 19.5, 15.2, 9.9, 6.7, 5.1; FAB HRMS (NBA/CsI) *m/e* 791.2251, M + Cs^+ calcd for $C_{35}H_{50}O_{10}Si$ 791.2228.

(25) Chen, S.-H.; Kant, J.; Mamber, S. W.; Roth, G. P.; Wei, J.-M.; Marshall, D.; Vyas, D. M.; Farina, V. *Bio. Med. Chem. Lett.* **1994**, *4*, 2223–2228.

Enone 15. To a solution of 7-TES deacetylbaccatin III (**14**, 1.5 g, 2.28 mmol) and 4-methylmorpholine *N*-oxide (NMO, 240 mg, 2.05 mmol) in CH_2Cl_2 (5 mL) were added 4 Å molecular sieves (200 mg), and the suspension was stirred at 25 °C for 10 min. A catalytic amount of tetrapropylammonium perruthenate (TPAP, 40 mg, 0.11 mmol) was added by portions, and the reaction mixture was stirred at 25 °C for 0.5 h. Small amounts of 4-methylmorpholine *N*-oxide and TPAP were added alternatively at 0.5 h intervals until the starting material was consumed to the extent of *ca.* 95% by TLC. The reaction mixture was filtered through silica gel, eluted with CH_2Cl_2 (100 mL), and concentrated to give enone **15** (1.44 g, 96%) as a white solid: R_f = 0.5 (silica, 50% EtOAc in hexanes); IR (thin film) ν_{max} 3446, 2957, 2882, 1726, 1672, 1456, 1367, 1243, 1106 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.05 (dd, J = 8.0, 1.0 Hz, 2H, Bz), 7.61 (t, J = 7.5 Hz, 1H, Bz), 7.45 (t, J = 7.5 Hz, 2H, Bz), 5.63 (d, J = 7.5 Hz, 1H, 2-H), 5.30 (d, J = 2.0 Hz, 1H, 10-H), 4.90 (d, J = 8.0 Hz, 1H, 5-H), 4.36 (dd, J = 10.5, 7.0 Hz, 1H, 7-H), 4.31 (d, J = 8.5 Hz, 1H, 20-H), 4.30 (d, J = 2.0 Hz, 1H, 10-OH), 4.11 (d, J = 8.5 Hz, 1H, 20-H), 3.93 (d, J = 7.5 Hz, 1H, 3-H), 2.92 (d, J = 19.5 Hz, 1H, 14-H), 2.62 (d, J = 19.5 Hz, 1H, 14-H), 2.50–2.42 (m, 1H, 6-H), 2.17 (s, 3H, Me), 2.08 (s, 3H, Me), 1.90–1.82 (m, 1H, 6-H), 1.77 (s, 1H, 1-OH), 1.70 (s, 3H, Me), 1.21 (s, 3H, Me), 1.14 (s, 3H, Me), 0.90 (t, J = 8.0 Hz, 9H, $Si(CH_2CH_3)_3$), 0.60–0.42 (band, 6H, $Si(CH_2CH_3)_3$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 208.2, 198.1, 170.2, 166.8, 156.6, 139.1, 134.0, 130.0, 128.8, 128.8, 84.0, 80.4, 78.5, 76.2, 75.7, 72.9, 72.8, 58.8, 45.9, 43.4, 42.5, 37.2, 33.0, 21.7, 17.5, 13.6, 9.6, 6.7, 5.1; FAB HRMS (NBA/NaI) *m/e* 657.3070, M + Na^+ calcd for $C_{35}H_{48}O_{10}Si$ 657.3095.

Triol 16. To a solution of enone **15** (1.44 g, 2.19 mmol) in MeOH (300 mL) at 0 °C was slowly added an aqueous solution of K_2CO_3 (3.0 g in 32 mL of H_2O). The solution was stirred at 0 °C for 2.5 h. The reaction was quenched with aqueous NH_4Cl (150 mL), and the resulting mixture was extracted with CH_2Cl_2 (2 × 200 mL). The organic layer was dried (Na_2SO_4), concentrated, and purified by flash chromatography (silica, 35 → 50% EtOAc in petroleum ether) to give enone **15** (270 mg, 19%) and triol **16** (912 mg, 93% based on 81% conversion): R_f = 0.24 (silica, 50% EtOAc in hexanes); IR (thin film) ν_{max} 3414, 2957, 2881, 1727, 1664, 1370 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.23 (d, J = 9.5 Hz, 1H, 10-H), 4.89 (d, J = 9.5 Hz, 1H, 5-H), 4.63 (d, J = 9.5 Hz, 1H, 20-H), 4.56 (d, J = 9.5 Hz, 1H, 20-H), 4.32 (dd, J = 11.0, 7.0 Hz, 1H, 7-H), 4.28 (d, J = 2.5 Hz, 1H, 10-OH), 3.89 (dd, J = 6.5, 4.0 Hz, 1H, 2-H), 3.57 (d, J = 6.5 Hz, 1H, 3-H), 2.78 (d, J = 19.5 Hz, 1H, 14-H), 2.58 (d, 4.0 Hz, 1H, 2-OH), 2.52 (d, J = 19.5 Hz, 1H, 14-H), 2.49–2.42 (m, 1H, 6-H), 2.03 (s, 3H, Me), 1.92–1.84 (m, 1H, 6-H), 1.68 (s, 3H, Me), 1.21 (s, 3H, Me), 1.04 (s, 3H, Me), 0.90 (t, J = 8.0 Hz, 9H, $Si(CH_2CH_3)_3$), 0.60–0.40 (band, 6H, $Si(CH_2CH_3)_3$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 208.9, 198.5, 170.1, 156.7, 138.8, 83.8, 81.2, 77.6, 75.7, 72.8, 72.5, 58.8, 45.8, 43.1, 42.8, 37.3, 32.7, 21.6, 17.5, 13.6, 9.7, 6.7, 5.1; FAB HRMS (NBA/NaI) *m/e* 575.2648, M + Na^+ calcd for $C_{28}H_{44}O_9Si$ 575.2652.

Carbonate 2. A solution of diol **16** (60.0 mg, 0.109 mmol) in THF (2 mL) was treated with carbonyldiimidazole (110.0 mg, 0.678 mmol) and stirred at 40 °C for 0.5 h. The reaction mixture was concentrated and redissolved in THF (5 mL). TLC analysis confirmed total consumption of starting material. Aqueous HCl (1 N, 5 mL) was added, and the resulting solution was stirred for 15 min at 25 °C. Et_2O (25 mL) was added, and the organic layer was separated, washed with aqueous $NaHCO_3$ (10 mL) and brine (10 mL), dried ($MgSO_4$), and concentrated to give carbonate **2** (58 mg, 93%) as a white foam: R_f = 0.50 (silica, 35% EtOAc in hexanes); IR (thin film) ν_{max} 3438, 2957, 2882, 1820, 1731, 1685, 1370, 1236 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 5.27 (d, J = 2.5 Hz, 1H, 10-H), 4.89 (d, J = 9.0 Hz, 1H, 5-H), 4.60 (d, J = 9.0 Hz, 1H, 20-H), 4.45 (d, J = 9.0 Hz, 1H, 20-H), 4.43 (d, J = 6.0 Hz, 1H, 2-H), 4.33 (dd, J = 10.0, 7.5 Hz, 1H, 7-H), 4.28 (d, J = 2.5 Hz, 1H, 10-OH), 3.54 (d, J = 6.0 Hz, 1H, 3-H), 2.88 (d, J = 20.0 Hz, 1H, 14-H), 2.75 (d, J = 20.0 Hz, 1H, 14-H), 2.54–2.47 (m, 1H, 6-H), 2.08 (s, 3H, Me), 2.06 (s, 3H, Me), 1.92–1.84 (m, 1H, 6-H), 1.77 (s, 3H, Me), 1.31 (s, 3H, Me), 1.15 (s, 3H, Me), 0.88 (t, J = 8.5 Hz, 9H, $Si(CH_2CH_3)_3$), 0.55–0.45 (band, 6H, $Si(CH_2CH_3)_3$); ^{13}C NMR (125 MHz, $CDCl_3$) δ 208.4, 195.5, 170.5, 154.0, 152.0, 141.2, 88.4, 83.9, 79.8, 79.0, 76.7, 75.7, 71.9, 60.3, 43.0, 41.6, 39.8, 37.7, 31.6, 21.5, 17.8, 14.4, 9.7, 6.6, 5.0; FAB HRMS (NBA) *m/e* 579.2652, M + H^+ calcd for $C_{29}H_{42}O_{10}Si$ 579.2626.

Alcohol 21a. A solution of carbonate **2** (10 mg, 0.0173 mmol) in THF (1 mL) at −78 °C was treated with *n*-BuLi (0.087 mL of a 1.6 M solution in hexanes, 0.139 mmol) and stirred for 1.0 h. The reaction mixture was poured into a mixture of Et_2O (10 mL) and aqueous NH_4Cl (5 mL), the organic layer was separated, and the aqueous layer was extracted with Et_2O (2 × 5 mL). The combined organic layer was washed with brine (5 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 35 → 50% EtOAc in hexanes) to give **21a** (7.9 mg, 72%) as an amorphous solid: R_f = 0.36 (silica, 35% EtOAc in petroleum ether); IR (film) ν_{max} 3437, 2962, 2865, 1726, 1671, 1367, 1239, 1105 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 5.36 (d, J = 6.5 Hz, 1H, 2-H), 5.26 (d, J = 2.5 Hz, 1 H, 10-H), 4.89 (br d, J = 8.0 Hz, 1H, 5-H), 4.47 (d, J = 8.0 Hz, 1H, 20-H), 4.32 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.26 (d, J = 2.5 Hz, 1H, 10-OH), 4.15 (d, J = 8.0 Hz, 1H, 20-H), 3.81 (d, J = 6.5 Hz, 1H, 3-H), 2.73 (d, J = 20.0 Hz, 1H, 14-H), 2.57 (d, J = 20.0 Hz, 1H, 14-H), 2.49−2.41 (m, 1H, 6-H), 2.38−2.23 (m, 2H, OCCH_2(CH$_2$)$_2$CH$_3$), 2.06 (s, 3H, Me), 2.04 (s, 3H, Me), 1.90−1.82 (m, 1H, 6-H), 1.67 (s, 1H, OH), 1.64 (s, 3H, Me), 1.68−1.52 (m, 2H, OCCH$_2$CH_2CH$_2$CH$_3$), 1.41−1.30 (m, 2H, OC(CH$_2$)$_2$CH_2CH$_3$), 1.19 (s, 3H, Me), 1.07 (s, 3H, Me), 0.94−0.86 (band, 12H, CH$_3$ of Bu, OSi(CH$_2$CH_3)$_3$), 0.58−0.45 (band, 6H, OSi(CH_2CH$_3$)$_3$); FAB HRMS (NBA) *m/e* 637.3421, M + H$^+$ calcd for $C_{33}H_{52}O_{10}Si$ 637.3408.

Alcohol 21c. A solution of carbonate **2** (46 mg, 0.0795 mmol) in THF (3 mL) at −78 °C was treated with 2-furyllithium (4 mL of a 0.47 M suspension in Et_2O, 1.88 mmol, prepared from furan and *n*-BuLi[26]) and stirred for 10 min. The reaction mixture was poured into a mixture of CH_2Cl_2 (15 mL) and aqueous NH_4Cl (20 mL), the organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 10 mL). The combined organic layer was washed with brine (10 mL), dried ($MgSO_4$), and concentrated to give **21c**, which was taken into the next step without further purification: R_f = 0.38 (silica, 20% EtOAc in petroleum ether); IR (film) ν_{max} 3442, 2956, 2882, 1727, 1672, 1468, 1300, 1240, 1110, 1007, 733 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.66−7.64 (m, 1H, furan), 7.24 (br d, J = 3.5 Hz, 1H, furan), 6.58 (dd, J = 3.5, 1.5 Hz, 1H, furan), 5.55 (d, J = 6.5 Hz, 1H, 2-H), 5.31 (d, J = 2.0 Hz, 1H, 10-H), 4.92 (br d, J = 9.0 Hz, 1H, 5-H), 4.43 (d, J = 8.5 Hz, 1H, 20-H), 4.37 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.32 (d, J = 2.0 Hz, 1H, 10-OH), 4.18 (d, J = 8.5 Hz, 1H, 20-H), 3.93 (d, J = 6.5 Hz, 1H, 3-H), 2.88 (d, J = 20.0 Hz, 1H, 14-H), 2.63 (d, J = 20.0 Hz, 1H, 14-H), 2.55−2.37 (m, 1H, 6-H), 2.15 (s, 3H, Me), 2.09 (s, 3H, Me), 1.93−1.87 (m, 1H, 6-H), 1.81 (s, 1H, OH), 1.71 (s, 3H, Me), 1.23 (s, 3H, Me), 1.15 (s, 3H, Me), 0.93 (t, J = 8.0 Hz, 9H, OSi(CH$_2$CH_3)$_3$), 0.62−0.42 (band, 6H, OSi(CH_2CH$_3$)$_3$); FAB HRMS (NBA/NaI) *m/e* 669.2717, M + Na$^+$ calcd for $C_{33}H_{46}O_{11}Si$ 669.2707.

Alcohol 21d. A solution of carbonate **2** (50.0 mg, 0.0864 mmol) in THF (5 mL) at −78 °C was treated with 2-thienyllithium (1.30 mL of a 1.0 M solution in THF, 1.30 mmol) and stirred for 0.5 h. The reaction mixture was poured into a mixture of Et_2O (10 mL) and aqueous NH_4Cl (5 mL), the organic layer was separated, and the aqueous layer was extracted with Et_2O (2 × 10 mL). The combined organic layer was washed with brine (10 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 10 → 35% EtOAc in hexanes) to give **2** (16.5 mg, 33%) and **21d** (36.8 mg, 96% based on 67% conversion) as amorphous solids: R_f = 0.56 (silica, 50% EtOAc in hexanes); IR (film) ν_{max} 3403, 2945, 2881, 1717, 1669, 1520, 1413, 1360, 1248, 1078; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.84 (dd, J = 3.5, 1.0 Hz, 1H, thiophene), 7.64 (d, J = 1.0, 5.0 Hz, 1H, thiophene), 7.14 (dd, J = 5.0, 3.5 Hz, 1H, thiophene), 5.53 (br d, J = 6.5 Hz, 1H, 2-H), 5.29 (d, J = 2.5 Hz, 1H, 10-H), 4.90 (br d, J = 7.5 Hz, 1H, 5-H), 4.44 (d, J = 8.5 Hz, 1H, 20-H), 4.35 (dd, J = 10.5 Hz, 6.5 Hz, 1H, 7-H), 4.29 (d, J = 2.5 Hz, 1H, 10-OH), 4.19 (d, J = 8.5 Hz, 1H, 20-H), 3.90 (d, J = 6.5 Hz, 1H, 3-H), 2.89 (d, J = 19.5 Hz, 1H, 14-H), 2.62 (d, J = 19.5 Hz, 1H, 14-H), 2.49−2.43 (m, 1H, 6-H), 2.15 (s, 3H, Me), 2.07 (s, 3H, Me), 1.92−1.84 (m, 1H, 6-H), 1.73 (s, 1H, OH), 1.71 (s, 3H, Me), 1.21 (s, 3H, Me), 1.13 (s, 3H, Me), 0.91 (t, J = 8.0 Hz, 9H, OSi(CH$_2$CH_3)$_3$), 0.56−0.49 (band, 6H, OSi(CH_2CH$_3$)$_3$); FAB HRMS (NBA) *m/e* 663.2655, M + H$^+$ calcd for $C_{33}H_{46}O_{10}SSi$ 663.2659.

Alcohols 21e and 22e. A solution of carbonate **2** (107.9 mg, 0.186 mmol) in THF (6.2 mL) at −78 °C was treated with 3-thienyllithium (2.76 mL of a 0.41 M solution in Et_2O−THF−hexanes (4.5:1:2), 1.13 mmol, prepared from 3-bromothiophene and *n*-BuLi[28]) and stirred for 1.5 h. The reaction mixture was poured into a mixture of CH_2Cl_2 (15 mL) and aqueous NH_4Cl (20 mL), the organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 10 mL). The combined organic layer was washed with brine (10 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 20 → 30% EtOAc in hexanes) to give **2** (16.9 mg, 16%), **21e** (87.0 mg, 83% based on 84% conversion), and **22e** (9.7 mg, 10% based on 84% conversion) as amorphous solids.

Alcohol 21e: R_f = 0.74 (silica, 50% EtOAc in hexanes), 0.41 (silica, 10% EtOAc in benzene, three elutions); IR (thin film) ν_{max} 3442, 3110, 2956, 2882, 1725, 1672, 1410, 1368, 1244, 1198, 1101, 988, 825, 744 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.18 (dd, J = 3.0, 1.2 Hz, 1H, thiophene), 7.54 (dd, J = 5.0, 1.2 Hz, 1H, thiophene), 7.37 (dd, J = 5.0, 3.0 Hz, 1H, thiophene), 5.56 (dd, J = 6.5, 1.0 Hz, 1H, 2-H), 5.31 (d, J = 2.5 Hz, 1H, 10-H), 4.92 (dd, J = 7.5, 2.0 Hz, 1H, 5-H), 4.40−4.34 (m, 2H, 20-H, 7-H), 4.31 (d, J = 2.5 Hz, 1H, 10-OH), 4.15 (d, J = 8.5 Hz, 1H, 20-H), 3.93 (d, J = 6.5 Hz, 1H, 3-H), 2.88 (d, J = 20 Hz, 1H, 14-H), 2.63 (dd, J = 20.0, 1.0 Hz, 1H, 14-H), 2.47 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.18 (s, 3H, Me), 2.10 (s, 3H, Me), 1.89 (ddd, J = 14.5, 10.5, 2.0 Hz, 1H, 6-H), 1.81 (br s, 1H, OH), 1.72 (s, 3H, Me), 1.23 (s, 3H, Me), 1.15 (s, 3H, Me), 0.93 (t, J = 8.0 Hz, 9H, OSi(CH$_2$CH_3)$_3$), 0.62−0.48 (band, 6H, Si(CH_2CH$_3$)$_3$); FAB HRMS (NBA/CsI) *m/e* 795.1640, M + Cs$^+$ calcd for $C_{33}H_{46}O_{10}SSi$ 795.1635.

Alcohol 22e: R_f = 0.54 (silica, 50% EtOAc in hexanes); IR (thin film) ν_{max} 3437, 3108, 2955, 2880, 1709, 1674, 1605, 1520, 1410, 1360, 1258, 1194, 1103, 1004, 829, 744 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.15 (dd, J = 3.0, 1.0 Hz, 1H, thiophene), 7.49 (dd, J = 5.0, 1.0 Hz, 1H, thiophene), 7.35 (dd, J = 5.0, 3.0 Hz, 1H, thiophene), 5.59 (d, J = 6.0 Hz, 1H, 2-H), 5.27 (d, J = 2.5 Hz, 1H, 10-H), 4.73 (dd, J = 9.5, 3.5 Hz, 1H, 5-H), 4.40 (d, J = 8.5 Hz, 1H, 20-H), 4.32 (d, J = 2.5 Hz, 1H, 10-OH), 4.15 (d, J = 8.5 Hz, 1H, 20-H), 3.92 (dd, J = 11.5, 6.0 Hz, 1H, 7-H), 3.44 (d, J = 19.5 Hz, 1H, 14-H), 3.30 (d, J = 6.0 Hz, 1H, 3-H), 2.91 (br s, 1H, OH), 2.61 (d, J = 19.5 Hz, 1H, 14-H), 2.38 (ddd, J = 14.5, 9.5, 6.0 Hz, 1H, 6-H), 2.09 (s, 3H, Me), 1.99 (ddd, J = 14.5, 11.5, 3.5 Hz, 1H, 6-H), 1.81 (br s, 1H, OH), 1.65 (s, 3H, Me), 1.24 (s, 3H, Me), 1.16 (s, 3H, Me), 0.91 (t, J = 8.0 Hz, 9H, OSi(CH$_2$CH_3)$_3$), 0.60−0.46 (band, 6H, OSi(CH_2CH$_3$)$_3$); FAB HRMS (NBA/CsI) *m/e* 753.1530, M + Cs$^+$ calcd for $C_{31}H_{44}O_9SSi$ 753.1530.

Acetate 26e. A solution of alcohol **21e** (68.4 mg, 0.103 mmol) and 4-(dimethylamino)pyridine (DMAP, 37.8 mg, 0.309 mmol) in CH_2Cl_2 (4.4 mL) at 25 °C was treated with Ac_2O (0.370 mL, 3.92 mmol) and stirred for 2 h. The reaction mixture was diluted with CH_2Cl_2 (5 mL), treated with aqueous $NaHCO_3$ (7 mL), and stirred vigorously for 25 min. The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 10 mL). The combined organic layer was washed with brine (5 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 30% EtOAc in hexanes) to give **26e** (66.0 mg, 91%) as an amorphous solid: R_f = 0.48 (silica, 10% EtOAc in benzene, three elutions); IR (film) ν_{max} 3518, 2956, 2881, 1727, 1676, 1520, 1460, 1371, 1236, 1098, 985, 824, 744 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.19 (dd, J = 3.0, 1.1 Hz, 1H, thiophene), 7.55 (dd, J = 5.0, 1.1 Hz, 1H, thiophene), 7.38 (dd, J = 5.0, 3.0 Hz, 1H, thiophene), 6.58 (s, 1H, 10-H), 5.61 (dd, J = 6.5, 0.7 Hz, 1H, 2-H), 4.92 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.47 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.38 (d, J = 8.5 Hz, 1H, 20-H), 4.14 (d, J = 8.5 Hz, 1H, 20-H), 3.88 (d, J = 6.5 Hz, 1H, 3-H), 2.89 (d, J = 20 Hz, 1H, 14-H), 2.64 (br d, J = 20 Hz, 1H, 14-H), 2.54 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.23 (s, 3H, Me), 2.18 (s, 3H, Me), 2.17 (s, 3H, Me), 1.87 (ddd, J = 14.5, 10.5, 2.0, 1H, 6-H), 1.85 (s, 1H, OH), 1.66 (s, 3H, Me), 1.26 (s, 3H, Me), 1.19 (s, 3H, Me), 0.92 (t, J = 8.0 Hz, 9H, OSi(CH$_2$CH_3)$_3$), 0.65−0.54 (band, 6H, OSi(CH_2CH$_3$)$_3$); FAB HRMS (NBA/CsI) *m/e* 837.1760, M + Cs$^+$ calcd for $C_{35}H_{48}O_{11}SSi$ 837.1741.

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Alcohol 27e. A solution of enone **26e** (57.3 mg, 0.0813 mmol) in MeOH–THF (5:1, 4.1 mL) at 0 °C was treated with $NaBH_4$ (69.1 mg, 1.83 mmol, added by portions) and stirred for 2.5 h. The reaction mixture was diluted with CH_2Cl_2 (10 mL), treated with aqueous NH_4Cl (5 mL), and stirred for 10 min. The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 10 mL). The combined organic layer was washed with brine (5 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 30% EtOAc in hexanes) to give **26e** (6.8 mg, 12%) and **27e** (45.2 mg, 89% based on 88% conversion) as amorphous solids: $R_f = 0.48$ (silica, 50% EtOAc in hexanes); IR (film) ν_{max} 3520, 2953, 2881, 1719, 1520, 1370, 1238, 1100, 979, 823, 746 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.20 (dd, J = 3.0, 1.0 Hz, 1H, thiophene), 7.57 (dd, J = 5.0, 1.0 Hz, 1H, thiophene), 7.35 (dd, J = 5.0, 3.0 Hz, 1H, thiophene), 6.45 (s, 1H, 10-H), 5.54 (d, J = 7.0 Hz, 1H, 2-H), 4.96 (br d, J = 8.5 Hz, 1H, 5-H), 4.82 (br dd, J = 12.0, 8.0 Hz, 1H, 13-H), 4.48 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.36 (d, J = 8.5 Hz, 1H, 20-H), 4.15 (d, J = 8.5 Hz, 1H, 20-H), 3.85 (d, J = 7.0 Hz, 1H, 3-H), 2.53 (ddd, J = 14.5, 9.5, 6.5, 1H, 6-H), 2.27 (s, 3H, Me), 2.28–2.21 (m, 2H, 14-CH_2), 2.18 (s, 6H, Me, Me), 2.03 (s, 1H, OH), 1.87 (ddd, J = 14.5, 10.5, 2.0 Hz, 1H, 6-H), 1.67 (s, 3H, Me), 1.65 (s, 1H, OH), 1.18 (s, 3H, Me), 1.04 (s, 3H, Me), 0.92 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.64–0.50 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 839.1908, M + Cs^+ calcd for $C_{35}H_{50}O_{11}SSi$ 839.1897.

Di-TES Taxoid 29e. To a solution of alcohol **27e** (19.5 mg, 0.0276 mmol, previously azeotroped twice with benzene) and β-lactam **28** (27.5 mg, 0.0721 mmol, previously azeotroped twice with benzene) in THF (1.4 mL) at 0 °C was added $NaN(SiMe_3)_2$ (0.066 mL of a 1.0 M solution in THF, 0.066 mmol) dropwise. The resulting solution was stirred for 0.5 h and poured into a mixture of CH_2Cl_2 (10 mL) and aqueous NH_4Cl (5 mL). The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 5 mL). The combined organic layer was washed with brine (5 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 20 → 30% EtOAc in hexanes) to give **27e** (1.1 mg, 6%) and **29e** (17.3 mg, 61% based on 94% conversion) as white solids: $R_f = 0.86$ (silica, 50% EtOAc in hexanes); IR (film) ν_{max} 3519, 3437, 2953, 2879, 1726, 1666, 1515, 1483, 1369, 1240, 1100, 979, 825, 746 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.32 (dd, J = 3.0, 1.2 Hz, 1H, thiophene), 7.76–7.73 (m, 2H), 7.60 (dd, J = 5.0, 1.2 Hz, 1H, thiophene), 7.52–7.29 (band, 9H), 7.10 (d, J = 9.0 Hz, 1H, NH), 6.44 (s, 1H, 10-H), 6.26 (br t, J = 9.0 Hz, 1H, 13-H), 5.72 (dd, J = 9.0, 2.0 Hz, 1H, 3′-H), 5.61 (d, J = 7.0 Hz, 1H, 2-H), 4.95 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.70 (d, J = 2.0 Hz, 1H, 2′-H), 4.48 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.37 (d, J = 8.5 Hz, 1H, 20-H), 4.23 (d, J = 8.5 Hz, 1H, 20-H), 3.81 (d, J = 7.0 Hz, 1H, 3-H), 2.56–2.49 (m, 1H, 6-H), 2.54 (s, 3H, Me), 2.35 (dd, J = 15.5, 9.0 Hz, 1H, 14-H), 2.17 (s, 3H, Me), 2.07 (dd, J = 15.5, 9.0 Hz, 1H, 14-H), 2.03 (d, J = 1.0 Hz, 3H, 18-Me), 1.94–1.87 (m, 1H, 6-H), 1.69 (s, 3H, Me), 1.68 (s, 1H, OH), 1.20 (s, 3H, Me), 1.18 (s, 3H, Me), 0.93 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.81 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.63–0.53 (band, 6H, $OSi(CH_2CH_3)_3$), 0.52–0.36 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 1220.3675, M + Cs^+ calcd for $C_{57}H_{77}O_{14}SSi_2N$ 1220.3658.

Taxoid 30e. A solution of silyl ether **29e** (17.3 mg, 0.0159 mmol) in THF (0.6 mL) at 25 °C was treated with HF·pyridine (0.150 mL) and stirred for 2 h. The reaction mixture was poured into a mixture of EtOAc (10 mL) and aqueous $NaHCO_3$ (5 mL), and the resulting mixture was stirred for 10 min. The organic layer was separated, and the aqueous layer was extracted with EtOAc (2 × 10 mL). The combined organic layer was washed with brine (5 mL), dried ($MgSO_4$), concentrated, and purified by preparative TLC (silica, 25% EtOAc in petroleum ether) to give **30e** (7.7 mg, 56%) as a colorless film: R_f = 0.11 (silica, 50% EtOAc in hexanes); IR (film) ν_{max} 3496, 3434, 2940, 1723, 1648, 1519, 1370, 1243, 1071, 975 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.32 (dd, J = 3.0, 1.0 Hz, 1H, thiophene), 7.75–7.72 (m, 2H), 7.60 (dd, J = 5.0, 1.0 Hz, 1H, thiophene), 7.53–7.33 (band, 9H), 6.95 (d, J = 9.0 Hz, 1H, NH), 6.28–6.23 (m, 2H, 10-H, 13-H), 5.81 (dd, J = 9.0, 2.0 Hz, 1H, 3′-H), 5.58 (d, J = 7.0 Hz, 1H, 2-H), 4.95 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.80 (dd, J = 4.5, 2.0 Hz, 1H, 2′-H), 4.41 (br t, J = 7.5 Hz, 1H, 7-H), 4.36 (d, J = 8.5 Hz, 1H, 20-H), 4.22 (d, J = 8.5 Hz, 1H, 20-H), 3.78 (d, J = 7.0 Hz, 1H, 3-H), 3.49 (d, J = 4.5 Hz, 1H, 2′-OH), 2.55 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.45 (br s, 1H, OH), 2.40 (s, 3H, Me), 2.34 (dd, J = 15.5, 9.0 Hz, 1H, 14-H), 2.25 (dd, J = 15.5, 9.0 Hz, 1H, 14-H), 2.24 (s, 3H, Me), 1.89 (ddd, J = 14.5, 11.0, 2.0 Hz, 1H, 6-H), 1.81 (d, J = 2.0 Hz, 3H, 18-Me), 1.74 (br s, 1H, OH), 1.67 (s, 3H, Me), 1.24 (s, 3H, Me), 1.13 (s, 3H, Me); FAB HRMS (NBA/CsI) *m/e* 992.1940, M + Cs^+ calcd for $C_{45}H_{49}O_{14}NS$ 992.1928.

MPA Taxoid 31e. A solution of taxoid **30e** (4.3 mg, 0.005 mmol) and Et_3N (0.0033 mL, 0.0237 mmol) in CH_2Cl_2 (0.2 mL) at 25 °C was treated with 2-fluoro-1-methylpyridinium *p*-toluenesulfonate (2.1 mg, 0.0075 mmol) and stirred for 35 min. The reaction mixture was directly purified by HPLC (Vydak RP-18, 22.5 × 3 mm, A → B 0.5 h linear; A, 20% MeOH in 20 mM NH_4OAc; B, 100% MeOH; 9 mL/min, t_R = 26.12 min) to give **30e** (0.8 mg, 19%) and **31e** (4.1 mg, 100% based on 81% conversion) as a colorless film: ^1H NMR (500 MHz, $CDCl_3$) δ 10.5 (d, J = 7.5 Hz, 1H), 8.44 (ddd, J = 9.0, 7.5, 2.0 Hz, 1H), 8.33–8.29 (m, 2H), 8.15 (dd, J = 3.0, 1.0 Hz, 1H, thiophene), 8.12 (br d, J = 6.0 Hz, 1H), 7.84 (br d, J = 8.5 Hz, 1H), 7.74–7.69 (m, 2H), 7.53 (dd, J = 5.0, 1.0 Hz, 1H, thiophene), 7.48–7.34 (band, 7H), 7.16–7.12 (m, 1H), 6.53–6.43 (m, 1H, 2′-H), 6.21 (s, 1H, 10-H), 6.03 (dd, J = 10.5, 8.0 Hz, 1H, 3′-H), 5.82 (br t, J = 9.0 Hz, 1H, 13-H), 5.44 (d, J = 7.0 Hz, 1H, 2-H), 4.90 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.33 (dd, J = 11.0, 6.5 Hz, 1H, 7-H), 4.30 (d, J = 8.0 Hz, 1H, 20-H), 4.15 (d, J = 8.0 Hz, 1H, 20-H), 4.08 (s, 3H, N^+Me), 3.68 (d, J = 7.0 Hz, 1H, 3-H), 2.58–2.49 (m, 1H, 6-H), 2.52 (s, 3H, OAc), 2.21 (s, 3H, OAc), 2.04 (s, 3H, OAc), 2.02 (br s, 2H, OH, OH), 1.88 (ddd, J = 14.5, 11.5, 2.0 Hz, 1H, 6-H), 1.78 (br s, 3H, 18-Me), 1.64 (s, 3H, Me), 1.61 (dd, J = 16.0, 7.0 Hz, 1H, 14-H), 1.18 (dd, J = 16.0, 9.0 Hz, 1H, 14-H), 1.13 (s, 3H, Me), 1.08 (s, 3H, Me).

Alcohols 21f, 22f, and 16. A solution of carbonate **2** (62.6 mg, 0.108 mmol) in THF (5.4 mL) at −78 °C was treated with 2-lithiopyridine (1.15 mL of a 0.44 M solution in Et_2O–pentane (1:1), 0.506 mmol, prepared from 2-bromopyridine and *t*-BuLi[29]) and stirred for 1.3 h. The reaction mixture was poured into a mixture of EtOAc (10 mL) and aqueous NH_4Cl (5 mL), the organic layer was separated, and the aqueous layer was extracted with EtOAc (2 × 10 mL). The combined organic layer was washed with brine (5 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 70 → 100% EtOAc in petroleum ether) to give **16** (16.3 mg, 27%), **21f** (28.0 mg, 39%), and **22f** (8.4 mg, 13%) as amorphous solids.

Alcohol 21f: $R_f = 0.60$ (silica, EtOAc); ^1H NMR (500 MHz, $CDCl_3$) δ 8.77 (ddd, J = 4.5, 1.7, 1.0 Hz, 1H, pyridine), 8.05 (br d, J = 7.5 Hz, 1H, pyridine), 7.89 (ddd, J = 7.5, 7.5, 1.7 Hz, 1H, pyridine), 7.53 (ddd, J = 7.5, 4.5, 1.0 Hz, 1H, pyridine), 5.61 (dd, J = 6.5, 1.0 Hz, 1H, 2-H), 5.33 (d, J = 2.5 Hz, 1H, 10-H), 4.92 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.39 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.36 (d, J = 9.0 Hz, 1H, 20-H), 4.33 (d, J = 2.5 Hz, 1H, 10-OH), 4.28 (d, J = 9.0 Hz, 1H, 20-H), 3.96 (d, J = 6.5 Hz, 1H, 3-H), 2.98 (d, J = 20.0 Hz, 1H, 14-H), 2.71 (dd, J = 20.0, 1.0 Hz, 1H, 14-H), 2.50 (s, 1H, OH), 2.48 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.15 (s, 3H, Me), 2.11 (s, 3H, Me), 1.90 (ddd, J = 14.5, 10.5, 2.0 Hz, 1H, 6-H), 1.76 (s, 3H, Me), 1.24 (s, 3H, Me), 1.16 (s, 3H, Me), 0.93 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.63–0.47 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 790.2060, M + Cs^+ calcd for $C_{34}H_{47}O_{10}NSi$ 790.2024.

Alcohol 22f: $R_f = 0.45$ (silica, EtOAc); IR (film) ν_{max} 3435, 2954, 2879, 1732, 1674, 1589, 1362, 1305, 1241, 1116, 998, 829, 741 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.73 (br d, J = 4.5 Hz, 1H, pyridine), 8.15 (br d, J = 7.5 Hz, 1H, pyridine), 7.90 (ddd, J = 7.5, 7.5, 1.7 Hz, 1H, pyridine), 7.56 (ddd, J = 7.5, 4.5, 1.0 Hz, 1H, pyridine), 5.53 (dd, J = 7.5, 1.0, 1H, 2-H), 5.30 (d, J = 2.5 Hz, 1H, 10-H), 4.84 (dd, J = 9.5, 3.0 Hz, 1H, 5-H), 4.81 (br s, 1H, OH), 4.31 (d, J = 2.5 Hz, 1H, 10-OH), 4.25 (s, 2H, 20-CH_2), 3.97 (dd, J = 11.5, 6.5 Hz, 1H, 7-H), 3.31 (d, J = 19.5 Hz, 1H, 14-H), 3.23 (d, J = 7.5 Hz, 1H, 3-H), 2.57 (br d, J = 19.5 Hz, 1H, 14-H), 2.43 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.11 (s, 3H, Me), 1.95 (ddd, J = 14.5, 11.5, 3.0 Hz, 1H, 6-H), 1.92 (br s, 1H, OH), 1.70 (s, 3H, Me), 1.24 (s, 3H, Me), 1.17 (s, 3H, Me), 0.91 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.60–0.46 (band, 6H, $OSi(CH_2CH_3)_3$).

Alcohol 21g. To a solution of 3-lithiopyridine (1.15 mmol) in THF (7 mL), prepared from 3-bromopyridine and *n*-BuLi,[30] at −100 °C,

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(30) Parham, W. E.; Piccirilli, R. M. *J. Org. Chem.* **1977**, *42*, 257–260.

was added a solution of carbonate **2** (133.1 mg, 0.230 mmol) in THF (2 mL) *via* cannula. The resulting solution was stirred for 1 h, allowed to warm to −78 °C, stirred for 1 h, and poured into a mixture of EtOAc (10 mL) and aqueous NH_4Cl (10 mL). The organic layer was separated, and the aqueous layer was extracted with EtOAc (2 × 10 mL). The combined organic layer was washed with brine (10 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 70 → 95% EtOAc in petroleum ether) to give **2** (64.8 mg, 49%) and **21g** (43.9 mg, 57% based on 51% conversion) as amorphous solids: R_f = 0.56 (silica, EtOAc); IR (film) ν_{max} 3435, 2956, 2882, 1731, 1671, 1592, 1366, 1280, 1240, 1109, 991, 824, 739 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 9.24 (br s, 1H, pyridine), 8.81 (d, J = 1.0, 4.5 Hz, 1H, pyridine), 8.30 (ddd, J = 8.0, 2.0, 2.0 Hz, 1H, pyridine), 7.44 (dd, J = 8.0, 4.5 Hz, 1H, pyridine), 5.66 (d, J = 6.5 Hz, 1H, 2-H), 5.32 (s, 1H, 10-H), 4.92 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.38 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.32 (br s, 1H, OH), 4.30 (d, J = 8.5 Hz, 1H, 20-H), 4.13 (d, J = 8.5 Hz, 1H, 20-H), 3.96 (d, J = 6.5 Hz, 1H, 3-H), 2.92 (d, J = 19.5 Hz, 1H, 14-H), 2.66 (d, J = 19.5 Hz, 1H, 14-H), 2.48 (ddd, J = 15.5, 9.5, 6.5 Hz, 1H, 6-H), 2.18 (s, 3H, Me), 2.10 (s, 3H, Me), 2.03 (s, 1H, OH), 1.89 (ddd, J = 14.5, 10.5, 2.0 Hz, 1H, 6-H), 1.72 (s, 3H, Me), 1.23 (s, 3H, Me), 1.16 (s, 3H, Me), 0.92 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.62−0.48 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 790.2030, M + Cs^+ calcd for $C_{34}H_{47}O_{10}NSi$ 790.2024.

Alcohol 21h. A solution of carbonate **2** (150 mg, 0.259 mmol) in THF (20 mL) at −78 °C was treated with 4-lithio-*N*,*N*-dimethylaniline (6.5 mL of a 0.39 M solution in Et_2O−pentane (3:1), 2.54 mmol, prepared from 4-bromo-*N*,*N*-dimethylaniline and *t*-BuLi[31]) and stirred for 15 min. The reaction mixture was poured into a mixture of CH_2Cl_2 (35 mL) and aqueous NH_4Cl (20 mL), the organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 20 mL). The combined organic layer was washed with brine (20 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 10 → 35% EtOAc in petroleum ether) to give **21h** (55.0 mg, 30%) as an amorphous solid: R_f = 0.26 (silica, 35% EtOAc in hexanes); IR (film) ν_{max} 3414, 2924, 1706, 1669, 1605, 1530, 1094; ^1H NMR (500 MHz, $CDCl_3$) δ 7.90 (d, J = 9.0 Hz, 2H, Ar), 6.64 (d, J = 9.0 Hz, 2H, Ar), 5.60 (br d, J = 7.0 Hz, 1H, 2-H), 5.29 (d, J = 2.5 Hz, 1H, 10-H), 4.89 (br d, J = 9.5 Hz, 1H, 5-H), 4.37 (d, J = 8.5 Hz, 1H, 20-H), 4.36 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.31 (d, J = 2.5 Hz, 1H, 10-OH), 4.13 (br d, J = 8.5 Hz, 1H, 20-H), 3.90 (d, J = 7.0 Hz, 1H, 3-H), 3.05 (s, 6H, NMe_2), 2.93 (s, 1H, OH), 2.90 (d, J = 20.0 Hz, 1H, 14-H), 2.61 (br d, J = 20.0 Hz, 1H, 14-H), 2.49−2.40 (m, 1H, 6-H), 2.16 (s, 3H, Me), 2.08 (s, 3H, Me), 1.90−1.83 (m, 1H, 6-H), 1.69 (s, 3H, Me), 1.20 (s, 3H, Me), 1.13 (s, 3H, Me), 0.90 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.56−0.49 (band, 6H, $OSi(CH_2CH_3)_3$; FAB HRMS (NBA/NaI) *m/e* 722.3354, M + Na^+ calcd for $C_{37}H_{53}O_{10}NSi$ 722.3336.

Alcohol 21i. A solution of carbonate **2** (47 mg, 0.0812 mmol) in THF (2 mL) at −78 °C was treated with 1-lithionaphthalene (6.3 mL of a 0.32 M solution in Et_2O, 2.03 mmol, prepared from 1-bromonaphthalene and *t*-BuLi[32]) and stirred for 5 min. The reaction mixture was poured into a mixture of CH_2Cl_2 (15 mL) and aqueous NH_4Cl (20 mL), the organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 10 mL). The combined organic layer was washed with brine (10 mL), dried ($MgSO_4$), and concentrated to give alcohol **21i**, which was taken into the next step without further purification: R_f = 0.27 (20% EtOAc in petroleum ether); IR (film) ν_{max} 3442, 2954, 2882, 1724, 1671, 1461, 1362, 1279, 1228, 1195, 1092, 987, 826, 736 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.66 (s, 1H, naphthalene), 8.07 (dd, J = 9.0, 2.0 Hz, 1H, naphthalene), 7.97−7.89 (m, 3H, naphthalene), 7.68−7.57 (m, 2H, naphthalene), 5.71 (br d, J = 6.5 Hz, 1H, 2-H), 5.35 (d, J = 2.5 Hz, 1H, 10-H), 4.94 (br d, J = 8.0 Hz, 1H, 5-H), 4.41 (dd, J = 11.0, 7.0 Hz, 1H, 7-H), 4.37 (d, J = 8.5 Hz, 1H, 20-H), 4.35 (d, J = 2.0 Hz, 1H, 10-OH), 4.18 (d, J = 8.5 Hz, 1H, 20-H), 4.00 (d, J = 6.5 Hz, 1H, 3-H), 3.02 (d, J = 19.5 Hz, 1H, 14-H), 2.69 (d, J = 19.5 Hz, 1H, 14-H), 2.54−2.45 (m, 1H, 6-H), 2.27 (s, 3H, Me), 2.13 (s, 3H, Me), 1.94−1.87 (m, 1H, 6-H), 1.86 (s, 1H, OH), 1.75 (s, 3H, Me), 1.25 (s, 3H, Me), 1.20 (s, 3H, Me), 0.94 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.63−0.49 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA) *m/e* 707.3270, M + H^+ calcd for $C_{39}H_{50}O_{10}Si$ 707.3252.

Alcohols 21b and 22b. A solution of carbonate **2** (111.3 mg, 0.192 mmol) in THF (2 mL) at −78 °C was treated with vinyllithium (3.7 mL of a 0.52 M solution in Et_2O, 1.92 mmol, prepared from tetravinyltin and *n*-BuLi[33]) and stirred for 2.25 h. The reaction mixture was poured into a mixture of CH_2Cl_2 (20 mL) and aqueous NH_4Cl (10 mL), the organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (3 × 10 mL). The combined organic layer was washed with brine (15 mL), dried ($MgSO_4$), concentrated, and purified by flash chromatography (silica, 30 → 50% EtOAc in petroleum ether) to give **21b** (60.0 mg, 52%) and **22b** (25.7 mg, 24%) as white foams.

Alcohol 21b: R_f = 0.52 (silica, 50% EtOAc in hexanes); IR (film) ν_{max} 3442, 2956, 2882, 1727, 1672, 1407, 1368, 1243, 1182, 1110, 1050, 986, 826, 736 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.51 (dd, J = 17.0, 1.0 Hz, 1H, vinyl H), 6.13 (dd, J = 17.0, 10.5 Hz, 1H, vinyl H), 6.00 (dd, J = 10.5, 1.0 Hz, 1H, vinyl H), 5.45 (br d, J = 6.5 Hz, 1H, 2-H), 5.30 (d, J = 2.5 Hz, 1H, 10-H), 4.91 (br d, J = 9.5 Hz, 1H, 5-H), 4.44 (d, J = 8.5 Hz, 1H, 20-H), 4.35 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.30 (d, J = 2.5 Hz, 1H, 10-OH), 4.14 (d, J = 8.5 Hz, 1H, 20-H), 3.88 (d, J = 6.5 Hz, 1H, 3-H), 2.79 (d, J = 20.0 Hz, 1H, 14-H), 2.61 (d, J = 20.0 Hz, 1H, 14-H), 2.48 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.09 (s, 3H, Me), 2.08 (s, 3H, Me), 1.89 (ddd, J = 14.5, 10.5, 2.0 Hz, 1H, 6-H), 1.72 (s, 1H, OH), 1.68 (s, 3H, Me), 1.22 (s, 3H, Me), 1.12 (s, 3H, Me), 0.92 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.62−0.46 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 739.1925, M + Cs^+ calcd for $C_{31}H_{46}O_{10}Si$ 739.1915.

Alcohol 22b: R_f = 0.24 (silica, 50% EtOAc in hexanes); IR (film) ν_{max} 3439, 2955, 2881, 1711, 1671, 1409, 1365, 1188, 1115, 980, 833, 735 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.48 (br d, J = 17.0 Hz, 1H, vinyl H), 6.10 (dd, J = 17.0, 10.5 Hz, 1H, vinyl H), 5.97 (br d, J = 10.5 Hz, 1H, vinyl H), 5.47 (br d, J = 6.0 Hz, 1H, 2-H), 5.25 (d, J = 2.5 Hz, 1H, 10-H), 4.75 (dd, J = 9.5, 3.5 Hz, 1H, 5-H), 4.38 (d, J = 8.5 Hz, 1H, 20-H), 4.30 (d, J = 2.5 Hz, 1H, 10-OH), 4.24 (d, J = 8.5 Hz, 1H, 20-H), 3.90 (dd, J = 11.5, 6.0 Hz, 1H, 7-H), 3.28 (d, J = 19.5 Hz, 1H, 14-H), 3.24 (d, J = 6.0 Hz, 1H, 3-H), 3.06 (br s, 1H, OH), 2.58 (d, J = 19.5 Hz, 1H, 14-H), 2.38 (ddd, J = 14.5, 9.5, 6.0 Hz, 1H, 6-H), 2.07 (s, 3H, Me), 1.98 (ddd, J = 14.5, 11.5, 3.5 Hz, 1H, 6-H), 1.87 (s, 1H, OH), 1.61 (s, 3H, Me), 1.23 (s, 3H, Me), 1.13 (s, 3H, Me), 0.90 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.59−0.45 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 697.1802, M + Cs^+ calcd for $C_{29}H_{44}O_9Si$ 697.1809.

Thioether 21m. A solution of vinyl ester **21b** (55.6 mg, 0.0916 mmol) and 4-(dimethylamino)pyridine (DMAP, 1.8 mg, 0.0147 mmol) in CH_2Cl_2 (4.3 mL) at 25 °C was treated with PhSH (0.030 mL, 0.292 mmol) and stirred for 1.5 h. The reaction mixture was concentrated and purified by flash chromatography (silica, 30% EtOAc in petroleum ether) to give **21m** (58.1, 88%) as a white solid: R_f = 0.37 (silica, 30% EtOAc in hexanes), 0.34 (10% EtOAc in PhH, two elutions); IR (film) ν_{max} 3441, 3057, 2956, 2883, 1732, 1672, 1600, 1367, 1238, 1111, 988, 825, 739 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.39−7.24 (band, 5H), 5.44 (d, J = 6.5 Hz, 1H, 2-H), 5.28 (d, J = 2.5 Hz, 1H, 10-H), 4.90 (dd, J = 9.5, 2.0 Hz, 1H, 5-H), 4.38 (d, J = 8.0 Hz, 1H, 20-H), 4.33 (dd, J = 10.5, 6.5 Hz, 1H, 7-H), 4.29 (d, J = 2.5 Hz, 1H, 10-OH), 4.18 (d, J = 8.0 Hz, 1H, 20-H), 3.83 (d, J = 6.5 Hz, 1H, 3-H), 3.24−3.13 (m, 2H, CH_2SPh), 2.76 (d, J = 19.5 Hz, 1H, 14-H), 2.72−2.58 (m, 3H, 14-H, CH_2CH_2SPh), 2.47 (ddd, J = 14.5, 9.5, 6.5 Hz, 1H, 6-H), 2.39 (s, 1H, OH), 2.07 (s, 3H, Me), 2.05 (s, 3H, Me), 1.89 (ddd, J = 14.5, 10.5, 2.0 Hz, 1H, 6-H), 1.68 (s, 3H, Me), 1.23 (s, 3H, Me), 1.12 (s, 3H, Me), 0.92 (t, J = 8.0 Hz, 9H, $OSi(CH_2CH_3)_3$), 0.61−0.47 (band, 6H, $OSi(CH_2CH_3)_3$); FAB HRMS (NBA/CsI) *m/e* 849.2085, M + Cs^+ calcd for $C_{37}H_{52}O_{10}SSi$ 849.2105.

Cytotoxicity Measurements. Cells (10^4/well) were plated on 96 well plates with the following controls: no cells and toxic control (1 × 10^{-3} M sodium dodecyl sulfate (SDS)). The drug was diluted in ethanol and added directly to the wells. Plates were incubated at 37 °C under 5% carbon dioxide in sterile air, in a humidified incubator for 72 h. A solution (50 μL) of 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-5-[(phenylamino)carbonyl]-2*H*-tetrazolium hydroxide, XTT, 1 mg mL^{-1}) in phosphate-buffered saline (PBS, 100 mM) was added to each well. In the presence of viable cells, this colorless clear solution

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is enzymatically transformed to give a pink coloration. The plates were read at 450 nm using a microplate reader (Molecular Devices Thermomax).

Tubulin Polymerization Assays. Experiments were performed in 96 well plates at 37 °C following the standard protocol. In each case, 1.0 mM GTP was used to promote the initial polymerization ot tubulin. Negative control: tubulin (1.0 mg mL^{-1}) alone, $CaCl_2$ (0.25 mM) added after 15 min. Positive Taxol control: tubulin (1.0 mg mL^{-1}) with Taxol (10^{-6} M), $CaCl_2$ (0.25 mM) added after 15 min. 2′-MPA-3-thiophene: tubulin (1.0 mg mL^{-1}) with 2′-MPA-3-thiophene (10^{-6} M), $CaCl_2$ (0.25 mM) added after 15 min. 3-Thiophene: tubulin (1.0 mg mL^{-1}) with 3-thiophene (10^{-6} M), $CaCl_2$ (0.25 mM) added after 15 min. 2-Thiophene: tubulin (1.0 mg mL^{-1}) with 2-thiophene (10^{-6} M), $CaCl_2$ (0.25 mM) added after 15 min. Turbidity was measured as optical density at 340 nm using a microplate reader (Molecular Devices Thermomax).

***In Vivo* Efficacy.** The tumor model was generated from an ATT PC-3 prostate (and an OVCAR-3 in the case of the 2′-MPA-3-thiophene) carcinoma cell line that was maintained under standard proliferation conditions (37 °C, 5% carbon dioxide in sterile air). Each cohort comprised eight randomly chosen, female athymic nude mice, for a population size of 48. Hemocytometer-counted cells suspended in Hanks medium (Gibco, Grand Island, NY) were implanted subcutaneously (10^6 cells in 0.4 mL per mouse). Solid tumor growth was measured on each third day, and tumor volume was determined using the equation (length) × $(\text{width})^2(0.5)$. Equimolar amounts of the test compounds (1.0 μM) were injected intraperitoneally on days 1, 3, 5, 7, 9, and 11 using the following media: control, 5% dextrose in water (D5W); Taxol suspended in Cremaphor/D5W (5/95, 18.0 mg/kg of animal weight); 2′-MPA-3-thiophene dissolved in D5W (23.9 mg/(kg of animal weight); 3-thiophene suspended in Cremaphor/D5W (5/95, 18.0 mg/(kg of animal weight); 2-thiophene suspended in Cremaphor/D5W (5/95, 18.0 mg/(kg of animal weight). The procedures used for the maintenance of tumors and the experimental details were according to protocols set forth by the Developmental Therapeutics Program, National Cancer Institute (NCI). Inferences about efficacy were drawn by ANOVA followed by protected *t*-test.

Acknowledgment. We thank E. Bombardelli for a generous gift of 10-deacetylbaccatin III and Drs. D. H. Huang and G. Siuzdak for NMR and mass spectroscopic assistance, respectively. We also thank C.-H. Chang for TEM studies, S. Mauch for *in vivo* animal studies, and R. Merlock for development and execution of cytotoxicity and tubulin assays. This work was financially supported by the NIH, The Scripps Research Institute, fellowships from Rhône-Poulenc Rorer (P.G.N.), the Office of Naval Research (R.K.G.), NSERC (J.R.), and grants from Merck Sharp & Dohme, Pfizer, Inc., Schering Plough, and Glaxo, Inc.

Supplementary Material Available: Procedures for syntheses of compounds **4B−6B**, **8a−d**, **9a,b**, **10−12**, **26c,d**, **26f−i**, **27c,d**, **27f−i**, **29c,d**, **29f−i**, **30c,d**, **30f−i**, **26m**, **27m**, **29m**, **30m**, and **21j−l** (32 pages). This material is contained in many libraries on microfiche, immediately follows this article in the microfilm version of the journal, can be ordered from the ACS, and can be downloaded from the Internet; see any current masthead page for ordering information and Internet access instructions.

JA943574A