A New Semisynthesis of Paclitaxel from Baccatin III

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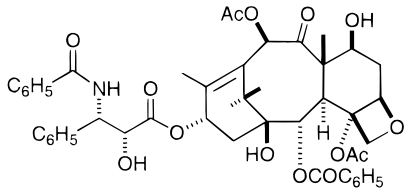
A new method for the semisynthesis of paclitaxel (Taxol) from baccatin III via a dioxo-oxathiazolidine intermediate is reported.

The important anticancer drug paclitaxel (Taxol) (**1**)[1] was



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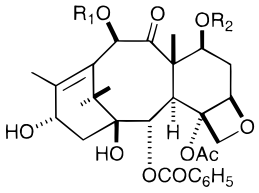
originally isolated in low yield from the bark of the Pacific Yew, *Taxus brevifolia* Nutt. (Taxaceae).[2] Because of its clinical importance, and because extraction on a large scale from yew bark was perceived as a threat to the environment, a great deal of effort has gone into finding alternate sources of this important substance. The major method used at the present time for the commercial preparation of paclitaxel is a semisynthetic route from a protected form of baccatin III (**2**)[3] which can readily be prepared from 10-deacetyl baccatin III (10-DAB, **3**), which itself can be

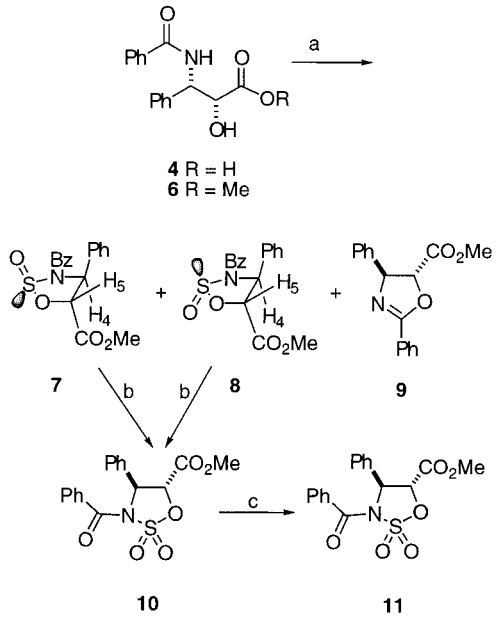


2 R_1 = Ac, R_2 = H
3 R_1 = R_2 = H
5 R_1 = Ac, R_2 = $OSiEt_3$

isolated in significant quantities (1 g/Kg) from the renewable needles of the European Yew, *Taxus baccata* L. (Taxaceae).[4] Since the yield of 10-DAB is about 10 times greater than the amount of paclitaxel that can be isolated from the bark of *T. brevifolia*, its conversion to paclitaxel is an attractive option.

A paclitaxel synthesis from 10-DAB requires the preparation of the *N*-benzoyl-*β*-phenylisoserine paclitaxel side chain (**4**),[5,6] its protection and coupling to a suitably protected baccatin III derivative, and deprotection to the final product. Several methods of coupling the paclitaxel side chain to protected baccatin III have also been developed,[5] but not all of them give high yields, and so we decided to develop a new protected form of the paclitaxel side chain. The more successful coupling methods have relied on the formation of some sort of heterocyclic derivative of **4** to "tie back" the α-OH and *β*-NHCOPh groups and thus reduce their steric bulk.[7] It has been reported that if this kind of linkage is not used, then acylation of baccatin III proceeds poorly and gives lower yields of paclitaxel, sometimes coupled with epimerization at the 2′ position.[8]

Scheme 1[a]



4 R = H
6 R = Me

7 **8** **9**

10 **11**

[a] Key: (a) $SOCl_2$, Et_3N, PhH, 0−5 °C; **7**, 68%; **8**, 14%; **9**, 0−3%; (b) $NaIO_4$, $RuCl_3$, CCl_4, CH_3CN, H_2O, 91%; (c) LiOH, H_2O, THF, 92%.

We selected an oxathiazolidine derivative of the paclitaxel side chain for investigation, based on reports that this ring system is stable to basic conditions but can be hydrolyzed under acidic conditions.[9] Treatment of the methyl ester of the paclitaxel side chain (**6**) with thionyl chloride in benzene gave a mixture of the two isomeric 2-oxo-1,2,3-oxathiazolidines **7** and **8** together with small amounts of oxazoline **9**, previously prepared as part of an alternative protected side chain derivative.[10] The use of solvents more polar than benzene gave higher yields or even exclusive formation of oxazoline **9**[11] (Scheme 1).

The isomeric nature of the two products **7** and **8** was proved by oxidation of both compounds with ruthenium chloride and periodate to the same 2,2-dioxo-1,2,3-oxathiazolidine derivative **10** (Scheme 1). The geometry of the isomeric products was assigned by NMR spectrometry. It is reported that in the case of a five-membered heterocyclic ring the sulfoxide bond (S═O) deshields the ring substituents that are cis to it.[9] The chemical shift of H_4 is further downfield when it is cis to the S═O bond, and further upfield when it is trans. The same trend applies to H_5 as well. Thus, the major product was assigned as **7**, and the secondary product as **8**. Interestingly, the coupling constant of the H_4 and H_5 protons of the major product (**7**) was found to be 8.8 Hz and that of the corresponding protons of the

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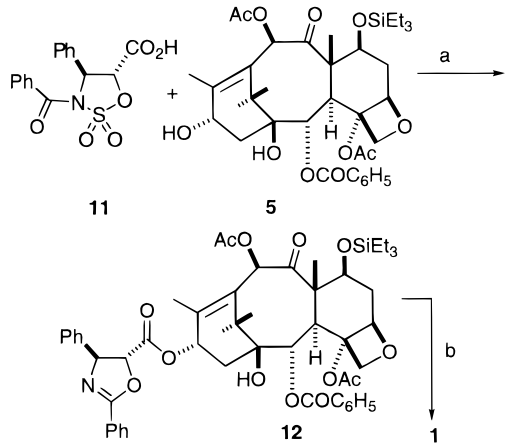
Scheme 2[a]



[a] Key: (a) DCC, DMAP, $PhCH_3$, 75 °C, 30%; (b) HCl, EtOH.

secondary product (**8**) was found to be 2.4 Hz. It is surprising that these values are so different, but it is assumed that the difference is due to subtle differences in the conformations of the five-membered rings in **7** and **8**.

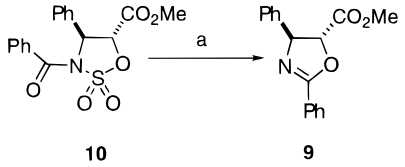
During these studies the major product (**7**) was found to be sparingly soluble in ethanol. Thus, it was possible to isolate **7** in pure form, without the need of further purification, by washing the crude product with ethanol after workup. The secondary product (**8**) was not very stable and was hydrolyzed to starting material even on a TLC plate.

The next step was the hydrolysis of the methyl ester **7** to the corresponding carboxylic acid. Although several hydrolysis conditions were tried, in all cases the sulfur linkage was broken before the methyl ester was hydrolyzed, contrary to literature reports that oxathiazolidines are stable under basic conditions.[9] We thus investigated the use of the 2,2-dioxo-1,2,3-oxathiazolidine derivative **10**, which could readily be prepared by oxidation of **7** or of a mixture of **7** and **8**. Compound **10** was readily hydrolyzed to the corresponding carboxylic acid **11** in the presence of lithium hydroxide, and the stage was set for the coupling of **11** with 7-(triethylsilyl)baccatin III (**5**) to give paclitaxel (Scheme 2).

Coupling of **11** with 7-(triethylsilyl)baccatin III (**5**) was carried out in the presence of dicyclohexylcarbodiimide (DCC) and 4-(dimethylamino)pyridine (DMAP). Surprisingly the product was not the expected dioxothiazolidine derivative, but instead the previously prepared oxazoline derivative **12**. The structure of **12** was assigned by comparison of its ^1H NMR spectrum with the corresponding spectrum of the literature compound.[10] The unexpected product **12** was then converted to paclitaxel (**1**) by the published procedure (Scheme 2).[10]

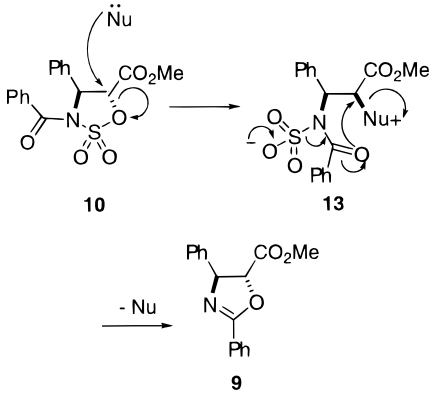
This surprising result prompted a short mechanistic study of the coupling reaction. Compound **10** was subjected to the coupling conditions in the absence of 7-(triethylsilyl)-baccatin III. Conversion of the 2,2-dioxo-1,2,3-oxathiazolidine derivative **10** to the *trans*-oxazoline **9** was observed (Scheme 3), indicating that conversion of **11** to its oxazoline analogue can occur in the absence of baccatin III. Presumably attack of a nucleophile on the C-2 position of **10** gives the intermediate **13** by an S_N2 reaction, resulting in inversion of configuration at C-2. This is then followed by intramolecular attack of the lone pair electrons of the carbonyl oxygen of the N-benzoyl group with concomitant elimination of sulfur trioxide to give the trans oxazoline **9** with overall retention of configuration (Scheme 4).

Scheme 3[a]

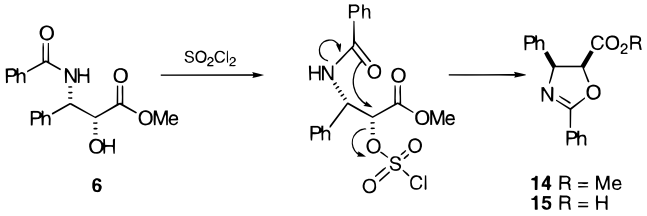


[a] Key: (a) DCC, DMAP, $PhCH_3$, 75 °C, 20%.

Scheme 4



Scheme 5



In an attempt to prepare the 2,2-dioxo-1,2,3-oxathiazolidine **10** in one step from the paclitaxel side chain methyl ester, ester **6** was treated with sulfuryl chloride and DMAP in CH_2Cl_2. Again, the results were surprising, since the *cis*-oxazoline **14** was the observed product (Scheme 5). The same product was also obtained when the reaction conditions were changed to sulfuryl chloride/pyridine or sulfuryl chloride/triethylamine/benzene. The structure of product **14** was assigned with the help of NMR and mass spectral data and by comparison with the *trans*-oxazoline **9**. In particular, the coupling constant of the H_4 and H_5 protons (J = 10.8 Hz) was consistent with that obtained by molecular modeling and Karplus correlation[12] for the cis isomer. Formation of **14** presumably occurs by esterification of the 2′-OH group with sulfuryl chloride, followed by backside attack of the amide carbonyl group with displacement of the chlorosulfate group. The *cis*-oxazoline **14** was then converted to its corresponding acid **15** in the presence of lithium hydroxide.

Coupling of the cis oxazoline **15** with 7-triethylsilylbaccatin III **5** gave the same *trans*-oxazoline ester **12** as did the *trans*-oxazoline **9**[10] or the dioxo-oxathiazolidine **10**. The 2′-carbon thus undergoes epimerization during coupling to give the more stable trans oxazoline product. *This finding means that a practical paclitaxel synthesis can be achieved from a mixture of oxazolines epimeric at the 2′-position, much as was previously reported for oxazolidine derivatives.*[13]

In summary, the chemistry described provides an alternate to the previously reported routes for the semisynthesis of paclitaxel. In addition, the transformation of the paclitaxel side chain to the *trans*-oxazoline **9** via the dioxo-oxathiazolidine derivative **10** and to the *cis*-oxazoline **14**

by treatment with sulfuryl chloride provides routes to both oxazoline diastereomers from the same starting material, and the epimerization of oxazoline **14** during coupling with 7-triethylsilylbaccatin III allows the use of either oxazoline isomer for paclitaxel synthesis.

Experimental Section

General Experimental Procedures. Chemicals were obtained from Aldrich Chemical Co. and were used without further purification, unless otherwise noted. Thionyl chloride was obtained from Acros Chemical Co. All anhydrous reactions were performed in oven-dried glassware under argon. Tetrahydrofuran (THF) was distilled over sodium/benzophenone, dichloromethane was distilled over calcium hydride, and toluene was distilled over sodium prior to use. All reactions were monitored by E. Merck analytical thin-layer chromatography (TLC) plates (silica gel 60 GF, aluminum back) and analyzed with 254 nm UV light and/or vanillin/sulfuric acid spray and/or iodine vapor. Silica gel for column chromatography was purchased from E. Merck (230−400 mesh). Preparative thin-layer chromatography (PTLC) plates (silica gel 60 GF) were purchased from Analtech. ^{1}H and ^{13}C NMR spectra were obtained in $CDCl_3$ or CD_3OD on Varian Unity 400 spectrometer (operating at 399.951 MHz for ^{1}H and 100.578 MHz for ^{13}C) and were assigned by comparison of chemical shifts and coupling constants with those of related compounds. Chemical shifts were reported as δ values relative to tetramethylsilane (TMS) as internal reference, and coupling constants were reported in Hz. FAB mass spectra were obtained at the Nebraska Center for Mass Spectrometry, University of Nebraska, and CI mass spectra were obtained in the Department of Chemistry, Virginia Polytechnic Institute and State University.

The phrase "worked-up in the usual way" refers to diluting the reaction mixture with excess organic solvent, washing with water and brine, drying over anhydrous sodium sulfate, and evaporating the solvent in vacuo unless otherwise noted. The methyl ester of the paclitaxel side chain and 7-(triethylsilyl)-baccatin III[14] were prepared following the procedures that are reported in the literature, and NMR data of these compounds were identical to those in the literature.

3-*N*-Benzoyl-4-phenyl-(4*S*,5*R*)-2-oxo-1,2,3-oxathiazolidine methyl esters (7 and 8) and (4*S*,5*R*)-2,4-Diphenyl-5-(methoxycarbonyl)-2-oxazoline (9). To a stirred solution of (2*S*,3*R*)-*N*-benzoyl-3-phenylisoserine methyl ester (100 mg, 0.33 mmol) in anhydrous benzene (4 mL) under argon was added triethylamine (5 equiv, 0.2 mL), and the mixture was stirred for 5 min at room temperature. The reaction mixture was then cooled to 3 °C where thionyl chloride (4 equiv, 1.336 mmol, 0.1 mL in 0.2 mL of benzene) diluted in benzene was introduced dropwise over 15 min. TLC immediately showed the formation of three new compounds, along with the starting material; one of the products was the major product (**7**), while one was of intermediate amount (**8**) and the third (**9**) was either minor or absent, depending on the exact conditions. After workup in the usual way with EtOAc and saturated aqueous sodium bicarbonate, EtOH was added to the crude product to yield the major ethanol-insoluble product **7** in pure form (68%). The rest of the crude product was subjected to PTLC purification (40% EtOAc/hexanes) to yield **8** (14%) and **9** (0−3%).

Major product (7): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.35−7.74 (m, 10H), 5.69 (d, 1H, J = 8.8 Hz), 5.66 (d, 1H, J = 8.8 Hz), 3.84 (s, 3H); ^{13}C NMR ($CDCl_3$, 400 MHz) δ 167.65, 166.69, 135.517, 134.045, 132.71, 129.05, 128.91, 128.76, 127.99, 86.14, 63.40, 53.34; HRFABMS m/z 345.0746 (calcd for $C_{17}H_{15}NO_5S$, 345.0671); LRFABMS $(M + H)^+$ m/z 346.

Intermediate product (8): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.35−7.67 (m, 10H), 6.224 (d, 1H, J = 2.4 Hz), 5.273 (d, 1H, J = 2.4 Hz), 3.89 (s, 3H).

Minor product (9): ^{1}H NMR, ^{13}C NMR and mass spectral data identical with literature data.[11] CIMS $(M + H)^+$ m/z 282.

3-*N*-Benzoyl-4-phenyl-(4*S*,5*R*)-2,2-dioxo-1,2,3-oxathiazolidine Methyl Ester (10). To a stirred solution of the oxathiazolidine (**7** or **8**) in $CH_3CN/CCl_4/H_2O$ (1:1:2) solvent system at room temperature was added an excess amount of $NaIO_4$ and a catalytic amount of $RuCl_3$. The reaction mixture was stirred for 45 min. and then filtered through sand/Celite/silica gel. After workup in the usual way with EtOAc, the crude product was isolated via PTLC (40% EtOAc/hexanes) in 91% yield. The same product was obtained from both **7** and **8**. ^{1}H NMR ($CDCl_3$, 400 MHz δ 7.38−7.83 (m, 10H), 6.03 (d, 1H, J = 8.4 Hz), 5.16 (d, 1H, J = 8.4 Hz), 3.87 (s, 3H); ^{13}C NMR ($CDCl_3$, 400 MHz) δ 166.49, 164.73, 135.07, 133.41, 132.47, 129.56, 129.28, 128.71, 128.46, 127.25, 78.38, 62.98, 53.71; HRFABMS m/z 361.0699 (calcd for $C_{17}H_{15}NO_6S$, 361.0620); LRFABMS $(M + H)^+$ m/z 362.0.

3-*N*-Benzoyl-4-phenyl-(4*S*,5*R*)-2,2-dioxo-1,2,3-oxathiazolidine Carboxylic Acid (11). To a stirred solution of 40 mg (0.11 mmol) starting material (**10**) in THF (2 mL) was added 500 μL of water, and the mixture was stirred for 5 min. LiOH (3 equiv, 8 mg, 0.33 mmol) was then introduced, and the reaction mixture was stirred for 45 min. When the TLC showed the disappearance of starting material, the reaction mixture was diluted with EtOAc, acidified with dilute HCl, and the usual workup procedure was performed. The crude product was applied on a PTLC plate (10% $MeOH/CH_2Cl_2$) in order to give the desired acid in 92% yield. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.38−7.79 (m, 10H), 7.14 (bs, 1H), 6.03 (d, J = 8.0 Hz), 5.18 (d, 1H, J = 8.0 Hz); ^{13}C NMR ($CDCl_3$, 400 MHz) δ 167.60, 166.79, 135.06, 133.49, 132.31, 129.62, 129.29, 128.75, 128.43, 127.31, 78.16, 62.89.

Acylation of 7-Triethylsilylbaccatin III with Acid (11) and Isolation of Paclitaxel Derivative 12. To a stirred emulsion of the acid (**11**) in anhydrous toluene was added 1 equiv of DMAP and 4 equiv of DCC, and the mixture stirred under argon for 5 min. 7-(Triethylsilyl)baccatin III (0.25 equiv) was then introduced, and the reaction mixture was warmed to 75 °C and stirred overnight. TLC showed the formation of a new product. The workup was performed in the usual way with EtOAc after the filtration of the reaction mixture through Celite. The product (**12**) was isolated via PTLC (40% EtOAc/hexane) in 30% yield. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.37−8.23 (m, 15H), 6.42 (s, 1H, H-10), 6.18 (t, 1H, H-13), 5.68 (d, 1H, J = 7.2 Hz, H-3′), 5.60 (d, 1H, J = 6.8 Hz, H-2), 4.93−4.96 (m, 2H, H-2′,5), 4.50 (dd, 1H, H-7), 4.29 (d, 1H, J = 8.4 Hz, H-20), 4.14 (d, 1H, J = 8.4 Hz, H-20), 3.84 (d, 1H, J = 6.8 Hz, H-3), 2.54 (m, 1H, H-6), 2.23−2.40 (m, 2H, H2−14), 2.16 (s, 3H, Me, 4-Ac), 2.08 (s, 3H, Me,10-Ac), 2.06 (t, 1H, H-6), 1.68 (s, 3H, Me-18), 1.58 (s, 3H, Me-19), 1.24 (s, 3H, Me-17), 1.19 (s, 3H, Me-16), 0.92 (t, 9H, 3 × Me, 7-TES), 0.56 (q, 6H, 3xCH_2, 7-TES); HRFABMS $(M + H)^+$ m/z 950.4166 (calcd for $C_{53}H_{64}NO_{13}Si$, 950.4147).

Conversion of 3-*N*-Benzoyl-4-phenyl-(4*S*,5*R*)-2,2-dioxo-1,2,3-oxathiazolidine Methyl Ester (10) to (4*S*,5*R*)-2,4-Diphenyl-5-(methoxycarbonyl)-2-oxazoline (9). To a stirred emulsion of 10 mg of compound **10** in anhydrous toluene was added 1 equiv of DMAP and 4 equiv of DCC, and the mixture was stirred under argon for 5 min. TLC showed the formation of a new product. The workup was performed in the usual way with EtOAc after the filtration of the reaction mixture through Celite. The product (**9**) was isolated via PTLC (40%EtOAc/hexane) in 20% yield. Spectral data were identical to those recorded.

(4*S*,5*S*)-2,4-Diphenyl-5-(methoxycarbonyl)-2-oxazoline (14). To a stirred solution of (2*S*,3*R*)-*N*-benzoyl-3-phenylisoserine methyl ester (25 mg, 0.08 mmol) in CH_2Cl_2 was added DMAP (7 equiv). The reaction mixture was then cooled to 0 °C, and sulfuryl chloride (3 equiv) was introduced dropwise. The product **14** was isolated in 65% yield after workup in the usual way and purification via PTLC. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.22−8.11 (m, 10H), 5.74 (d, 1H, J = 10.8 Hz), 5.38 (d, 1H, J = 10.8 Hz), 3.198 (s, 3H); ^{13}C NMR ($CDCl_3$, 400 MHz) δ 168.45, 164.73, 136.86, 131.92, 128.66, 128.41, 128.11, 128.06, 127.70, 126.69, 81.04, 73.45, 51.54; CIMS $(M + H)^+$ m/z 282.

(4*S*,5*S*)-2,4-Diphenyl-2-oxazolinecarboxylic Acid (15). To a stirred solution of ester **15** (40 mg, 0.11 mmol) in EtOH (2 mL) was added water (500 μL) and the mixture stirred for 5 min. LiOH (8 mg, 0.33 mmol, 3 equiv) was then introduced, and the reaction mixture was stirred for 45 min. When TLC showed the disappearance of starting material, the reaction mixture was diluted with EtOAc, acidified with dilute HCl, and the usual workup procedure was performed. The crude product was purified by PTLC (10% MeOH/CH_2Cl_2) to give the acid **15** in 92% yield. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.22−8.11 (m, 10H), 5.74 (d, 1H, J = 11.2 Hz) 5.38 (d, 1H, J = 11.2 Hz); CIMS $(M + H)^+$ m/z 268.

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References and Notes

(1) The name taxol was assigned to compound **1** by its discoverers.[2] Several years after this assignment of the name, the name Taxol was registered as a trademark by Bristol−Myers Squibb Pharmaceuticals for its clinical formulation of taxol, and the generic name paclitaxel was substituted for the compound of structure **1**. To avoid infringing on Bristol−Myers Squibb's trademark, and because the name Taxol no longer accurately describes the chemical substance **1**, the name paclitaxel will be used whenever the chemical substance taxol is intended.

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