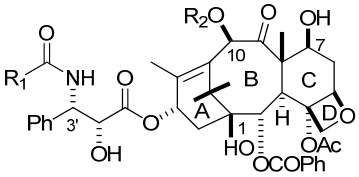
Chapter 2. Introduction to paclitaxel and docetaxel

2.1 Discovery and history



	R_1	R_2
paclitaxel	Ph	Ac
docetaxel	*t*-BuO	H

Figure 2.1 Paclitaxel and docetaxel

Paclitaxel and docetaxel (Figure 2.1) are both microtubule-interactive mitotic inhibitors that are widely used in cancer chemotherapy. Paclitaxel was isolated from the bark of the Pacific Yew tree, *Taxus brevifolia* by Wall and Wani at the Research Triangle Institute in 1967.[1] Later, in 1971 Wall and Wani published its chemical structure as well as some biological test results.[2] Paclitaxel was originally recognized as a cytotoxic and antileukemic agent with excellent activity, but it had poor solubility in water and was only available in low yield from its natural source (in 0.02% yield from Pacific Yew bark), which lowered people's interest in developing it as a drug. But the situation changed after further biological evaluations at NCI revealed its activity against various human solid tumor xenografts in nude mice in 1977, and shortly after that, Horwitz and coworkers discovered its interesting mechanism of action of paclitaxel in 1979.[3] They found that paclitaxel bound to β-tubulin, promoted tubulin polymerization and stabilized

microtubules from disassembly. This was an unknown mechanism at that time, and is considered as a key discovery in the development history of paclitaxel. The clinical trials began at 1984, and paclitaxel was eventually licensed to Bristol-Myers Squibb and finally approved for treatment of ovarian cancer by FDA in 1992. Paclitaxel is now used to treat patients with lung, ovarian, breast cancer, head and neck cancer, as well as advanced forms of Kaposi's sarcoma. Its annual sales peaked in 2000, reaching 1.6 billion dollars.

Docetaxel is a semisynthetic analog of paclitaxel and was discovered in the extensive research conducted to solve the scarcity issue of paclitaxel.[4] It can be synthesized by the esterification of 10-deacetyl baccatin III, which is extracted from the renewable and readily available European yew tree. Docetaxel differs from paclitaxel at two positions in the structure. Firstly, an *N*-Boc group replaced the *N*-benzoyl group in paclitaxel at the C3′ position on the side chain; in addition, docetaxel has a free hydroxyl group at the C10 position instead of the acetate ester in paclitaxel, which results in better solubility in water than paclitaxel.[5] Docetaxel has the same mechanism of action as paclitaxel involving promoting tubulin assembly, stabilizing microtubules in the cell and arresting the cell cycle, ultimately leading to apoptosis. It is approximately twice as potent as paclitaxel as a cytotoxic antimicrotubule agent.[4] Docetaxel is currently a well established anti-mitotic chemotherapy medication used for the treatment of breast, ovarian, prostate and non-small cell lung cancer, as well as locally advanced or metastatic breast or non small-cell lung cancer and metastatic castration resistant prostate cancer. It

was marketed worldwide by Sanofi-Aventis under the name Taxotere in 2004. The annual sales are approximately 2.5 billion dollars in 2007.

Paclitaxel and docetaxel are two of the most successful anticancer drugs in the past two decades, and the discovery of their unique mechanism of action opened a door to the development of other antimitotic drugs. This chapter will briefly introduce the chemistry and biology of paclitaxel and docetaxel.

2.2 Bioactivity

Since Horwitz and coworkers discovered the ability of paclitaxel to bind to β-tubulin, promote tubulin assembly and stabilize microtubules in 1979,[3] its bioactivity and mechanism of action have been extensively studied.

2.2.1 Interaction with β-tubulin and microtubules

As mentioned in Chapter 1, the assembly and disassembly of micrutubules in cells plays a crucial role in cell mitosis. Paclitaxel and docetaxel display antiproliferative activity through their interactions with β-tubulin and microtubules,[6] which suppress microtubule dynamics, disrupt the mitotic spindle and eventually result in cell death by apoptosis.[3]

The binding of paclitaxel or docetaxel with tubulin promotes the polymerization of

tubulin and stabilizes the microtubules, which suppresses the microtubule dynamics. Interestingly, paclitaxel and docetaxel bind to the tubulin on microtubules more tightly than to soluble tubulin itself.[7] The binding site is located on β-tubulin on the inner surface of the microtubules, which was confirmed by an electron diffraction structure of tubulin complexed with paclitaxel.[8] The binding of paclitaxel with tubulin introduces conformational changes of tubulin, which facilitate assembly and increase the affinity between neighbouring tubulin molecules.[9] Paclitaxel is able to bind to tubulin subunits on microtubules with a 1:1 stoichiometric ratio, so the increase of tubulin polymerization requires a high concentration of paclitaxel. However, the binding of a small amount of paclitaxel was found to efficiently suppress microtubule dynamics without increasing tubulin polymerization.[10]

Paclitaxel also decreases the critical concentration of tubulin required for polymerization and accelerates tubulin assembly.[3,11] The paclitaxel-induced microtubules are quite different from the normal ones in structure. The average diameter of paclitaxel-induced microtubules is 22 nm rather than 24 nm, and paclitaxel-induced microtubules consist with 12 protofilaments, instead of the normal 13 protofilaments.[12] The stabilities of paclitaxel-induced microtubules are maintained even under conditions that cause disassembly of normal microtubules. Meanwhile, paclitaxel was found to interact with microtubule-organizing centers and introduce their disarrangement, which changes the morphology of microtubule spindles in the cell.[10]

2.2.2 Drug resistance

Cancer cells can always find a way to resist drug effects, and this proved to be true for paclitaxel and docetaxel, as with other chemotherapies. The resistance to paclitaxel is generally developed through over-expression of P-glycoprotein and the mutation of the paclitaxel binding site on tubulin. P-glycoprotein is a transport protein which can pump hydrophobic substances out of the cell and maintain low concentrations of these substances inside the cell. Paclitaxel is a substrate of P-glycoprotein, so cancer cells can develop resistance through lowering the intracellular concentration of paclitaxel by P-glycoprotein mediated efflux.[13] On the other hand, upon treatment with paclitaxel, cancer cells can survive through mutation of the paclitaxel-binding site on tubulin, which results in the elimination of the drug target for paclitaxel inside the cancer cell.[14]

2.2.3 Side effects

Paclitaxel and docetaxel are not able to distinguish cancer cells from normal ones, so they are cytotoxic to all the cells. In fact, they can kill cancer cells more than normal cells, which results from the faster proliferative rate of cancer cells. The cytotoxicity of paclitaxel and docetaxel to normal cells leads to their side effects, which include hair loss, decrease in white blood cells, numbness of the fingers and toes, bone marrow suppression, and neutropenia (loss of neutrophils).[15] The decrease in white blood cells generally results in the susceptibility of the body to infections. These side effects make patients suffer a lot from pain and inconvenient life. So medicinal chemists have been trying to

reduce the side effects of paclitaxel and docetaxel by modifying the drug structure and introducing targeted delivery, and the investigations are still in progress.

2.2.4 Docetaxel for treatment of HRPC

Although docetaxel is similar to paclitaxel in structure, as well as in mechanism of action, it possesses some other interesting pharmaceutical profiles which paclitaxel does not have, among which it is the most promising that upon treatment with docetaxel patients with hormone refractory prostate cancer (HRPC) obtained significant survival and palliative benefits in two randomized trials. This result led to further clinical trials to investigate the efficiency of docetaxel against HRPC either by docetaxel itself or in combination with other drugs. Currently, docetaxel has become the best available drug for HRPC, and docetaxel-based therapies have become the standard of care for patients with HRPC.[16]

2.3 Structure-activity relationships (SAR)

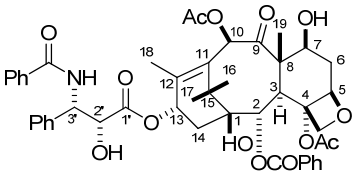


Figure 2.2 Structure of paclitaxel

Although paclitaxel achieved great success as an anticancer drug, it is an extremely

complicated structure, which delayed the development of improved analogs. Meanwhile, paclitaxel was found to be much less active against multidrug-resistant cancer cell lines and showed cytotoxicity to normal cells, resulting in side effects. Therefore, modifications to improve its activity and to reduce its side effects were necessary for its further development. These goals required that its structure-activity relationships (SAR) must be well understood. In the past three decades, extensive studies were carried out on the SAR of paclitaxel, with the goal of developing analogs with simple structures, better activities and fewer side effects. The results of the SAR studies is summarized in this section.

2.3.1 Main body: tetracyclic ring system

The unusual and distorted tetracyclic main body of paclitaxel was proved to be crucial for paclitaxel to maintain optimum conformation and drug-protein interaction. However, it was found that not all functional groups in the main body of paclitaxel contribute to the bioactivity. In general, the northern hemisphere can tolerate structural modification better than the southern hemisphere. The details are discussed below.

2.3.1.1 Northern hemisphere

Hydroxylation of the C6 position dramatically reduced the bioactivity by 30 fold,[17] whereas modifications at the C7 position such as acylation and dehydroxylation did not change the activity significantly.[18] In addition, it was reported that the conversion of the

C7 hydroxyl group to a keto group significantly decreased the activity of paclitaxel because it facilitated the opening of the oxetane ring at the C4-C5 position.[19] The keto group at the C9 position can be reduced to a hydroxyl group without loss of bioactivity.[20] Deacetylation at the C10 position preserved the activity of paclitaxel.[18a,21] Additionally, acylation of the C7 and C10 positions or the single acetylation of the C10 position of docetaxel preserved the activity.[21] The rearrangement around the double bond at the C11-C12 positions decreased the antiproliferative activity moderately,[22] and the formation of the C7-C8-C19 cyclopropane ring did not drastically affect the binding of paclitaxel to the microtubule binding site.[21,23]

2.3.1.2 Southern hemisphere

In comparison with the northern hemisphere, the southern hemisphere is far more essential for paclitaxel to maintain its antimicrotubule activity. This region is believed to play a more important role in maintaining the optimum conformation of paclitaxel, and the modifications in the southern hemisphere affect the bioactivity of paclitaxel dramatically, as discussed below.

Dehydroxylation at C1 decreases the activity slightly, indicating that the C1 hydroxyl group is helpful but not essential for the bioactivity of paclitaxel.[24] The C2 benzoyloxy group was proved to be crucial to maintain optimum conformation, and the aromatic substitutions affect the activity of paclitaxel dramatically.[25] Interestingly, the analogs with *meta* position substituted by certain functional groups or with the benzoyloxy group

replaced by certain alkenyl groups gave improved bioactivities.[26] In addition, the stereochemistries at the C2 and C13 positions are both essential for the activity of paclitaxel.[27] The removal of the acetate group at the C4 position decreases the activity significantly, but replacement by the methyl carbonate group improves the activity.[28] The oxetane ring at the C4-C5 position is required for activity, and opening of this ring decreases the activity significantly. The sulfetane analog, with oxygen replaced by sulfur, gave poor activity, but the taxol analog with the oxetane ring replaced with a cyclopropane ring was still active. In addition, hydroxylation at the C14 position does not affect the activity of paclitaxel.[18g,29]

2.3.2 Side chain at the C13 position

Although the side chain at the C13 position of paclitaxel is flexible in comparison to the tetracyclic skeleton, it is crucial for paclitaxel to maintain its bioactive conformation and tubulin-binding activity. Its removal leads to dramatic reduction or total loss of the activity, and major modifications on the side chain affect the activity of paclitaxel significantly in either positive or negative ways. An important example is docetaxel, which has a side chain with the *N*-benzoyl group of paclitaxel replaced by an *N*-Boc group at the C3′ position, and shows better tubulin-binding activity and cytotoxicity than paclitaxel, indicating that well-manipulated modifications on the side chain can improve the activity of paclitaxel.

The stereochemistries at the C2′ and C3′ positions have been investigated, and

analogs with the naturally occurring stereochemistry always had better bioactivity than others with different stereochemistries.[18a] The free C2′ hydroxyl group or a hydrolysable ester is required for the activity of paclitaxel or docetaxel.[18a,30] The insertion of a methylene group between the C1′ and C2′ positions reduces the activity of paclitaxel dramatically.[31] The phenyl group at the C3′ position is replacable by large alkyl groups, alkenyl groups or substituted phenyl groups without affecting the activity.[32] In some cases the replacement even improves the activity, but replacement with small alkyl groups such as the methyl group results in a remarkable loss of activity.[18a] Importantly, the *N*-acyl group at the C3′ position is crucial for maintaining correct conformation and good bioactivity of paclitaxel, but the modifications are still allowed.[18a] Interestingly, some of the acyl analogs gave improved activity.[18a]

Figure 2.3 summarizes the SAR information discussed in this section.

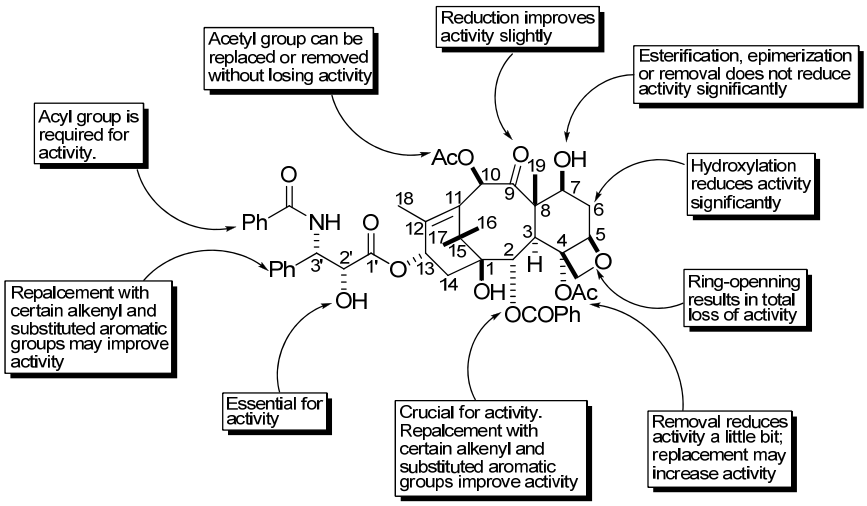


Figure 2.3 SAR of paclitaxel

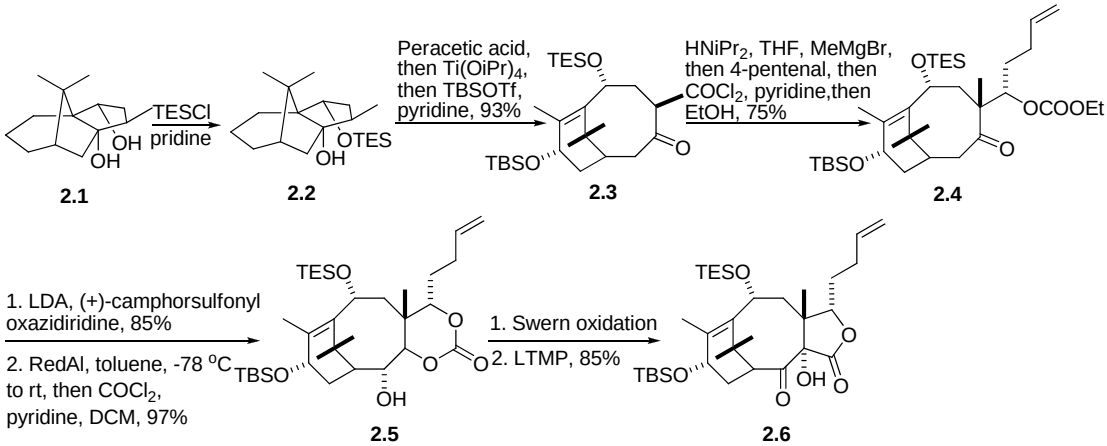
2.4 Synthesis

As shown in the first part of this chapter, paclitaxel is an extremely complex structure, consisting of an unusual and distorted tetracyclic skeleton with specific stereochemistries at different positions. Its total synthesis proved to be one of the greatest challenges in synthetic chemistry and of great value and importance to the development of new organic synthetic methods. Meanwhile, the scarcity of paclitaxel from its natural source prompted scientists to find a better way to obtain enough material in good quality and yield for further studies. Therefore, the synthesis of paclitaxel attracted the attention of synthetic chemists all over the world, and some of the successful approaches are reviewed and discussed below.

2.4.1 Total synthesis

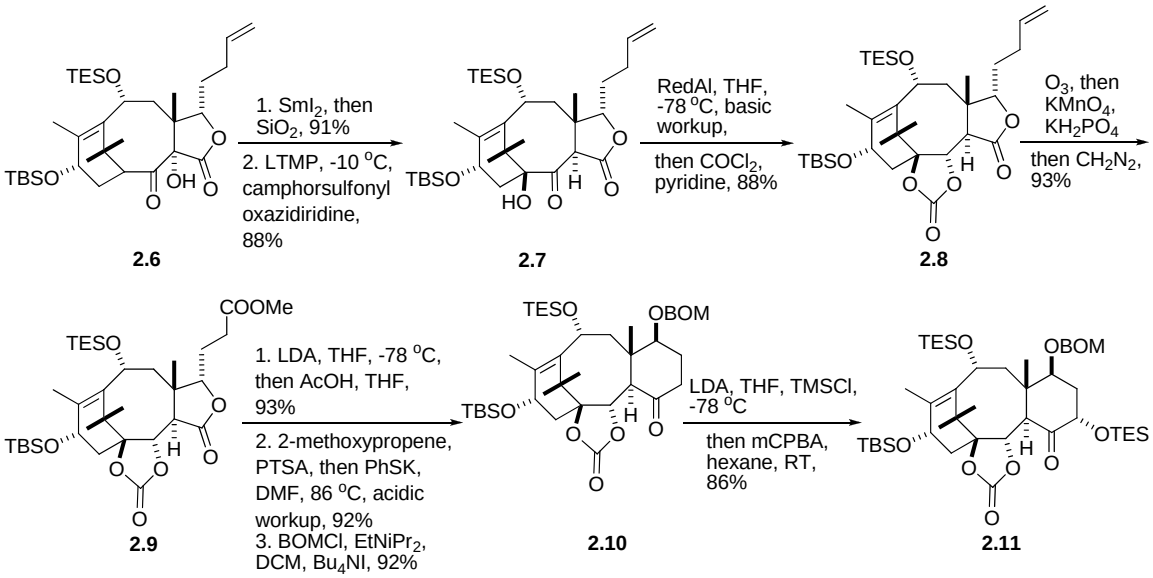
2.4.1.1 The Holton synthesis

In 1994, Holton and coworkers described their synthesis of paclitaxel for the first time in two papers.[33] The Holton synthesis began with the intermediate **2.1** readily available from camphor, which was silylated to give TES ether **2.2**. Then compound **2.2** was converted to ketone **2.3** in 93% yield, which subsequently underwent aldol condensation to give ketone **2.4** in 75% yield. Hydroxylation, reduction and cyclization of **2.4** afforded carbonate **2.5** in 82% yield, which was then converted to hydroxy lactone **2.6** via Swern oxidation and Chan rearrangement (Scheme 2.1).



Scheme 2.1 Synthesis of fragment **2.6**

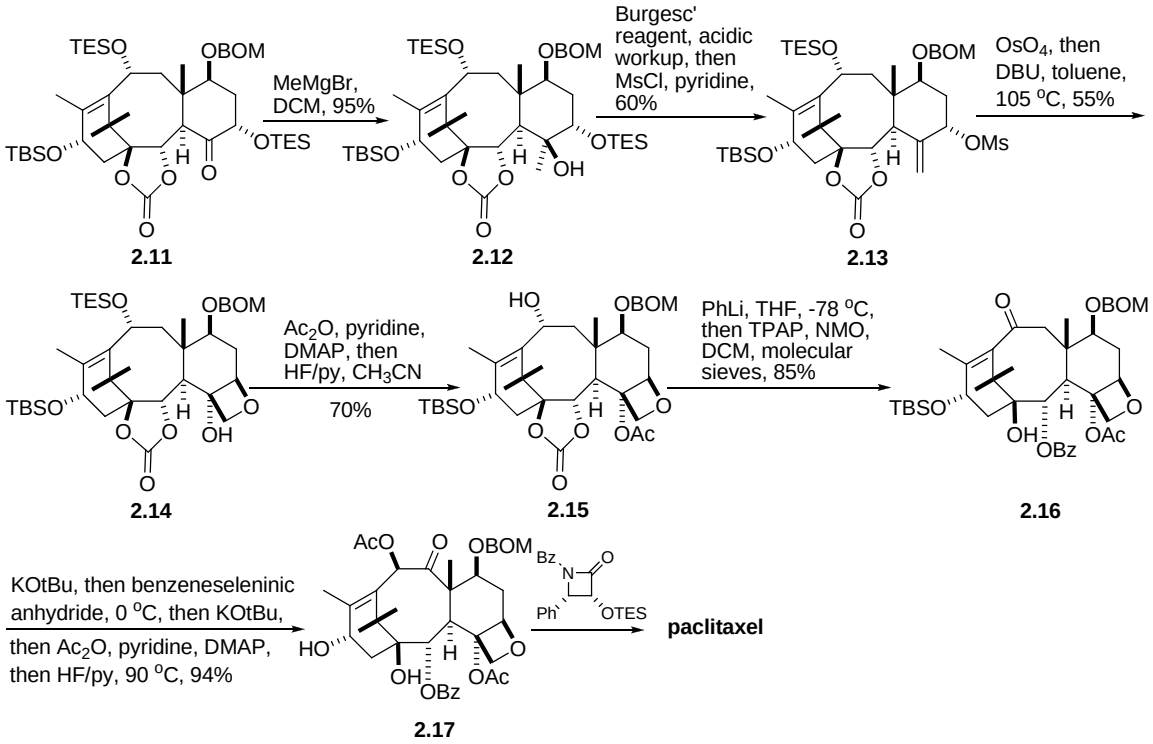
After samarium diiodide reduction, tautomerization and hydroxylation, lactone **2.6** was transformed to hydroxy ketone **2.7**, which was treated with phosgene to give lactone carbonate **2.8** quantitatively (Scheme 2.2).



Scheme 2.2 Synthesis of fragment **2.11**

Subsequently, **2.8** underwent ozonolysis, oxidation and esterification to provide

methyl ester **2.9**, which was converted to its MOP ether via Dieckman cyclisation and MOP protection. Decarbomethoxylation followed by BOM protection converted **2.9** to **2.10**, which was converted to ketone **2.11** via enolation and oxidation. Upon treatment with the Grignard reagent, ketone **2.11** was converted to tertiary alcohol **2.12**, which underwent elimination with Burgess' reagent and Ms protection to afford alkene **2.13**. Osmylation followed by cyclisation provided oxetanol **2.14**, which was converted to alcohol **2.15** via acetylation and TES deprotection.



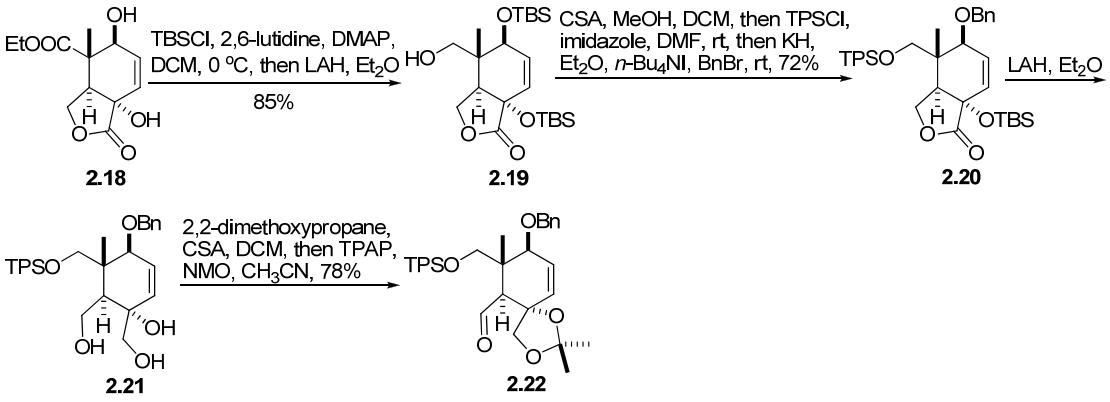
Scheme 2.3 Synthesis of paclitaxel

Treatment of **2.15** with phenyllithium and TPAP oxidation produced unsaturated ketone **2.16**, which was converted to 7-BOM-13-TBS baccatin III in two steps in high yield. Subsequently, TBS deprotection followed by coupling with β-lactam and global

deprotection finally furnished paclitaxel in 93% yield (Scheme 2.3). The Holton synthesis is the first successful synthetic approach of paclitaxel and the overall yield is about 4.5% from **2.1**.

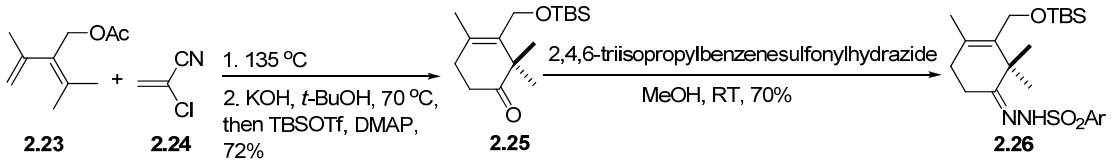
2.4.1.2 The Nicolaou synthesis

In 1994, Nicolaou and co-workers published their total synthesis of paclitaxel in *Nature*.[34] In 1995 they published two follow-up papers in *JACS* describing their synthesis in detail.[35]



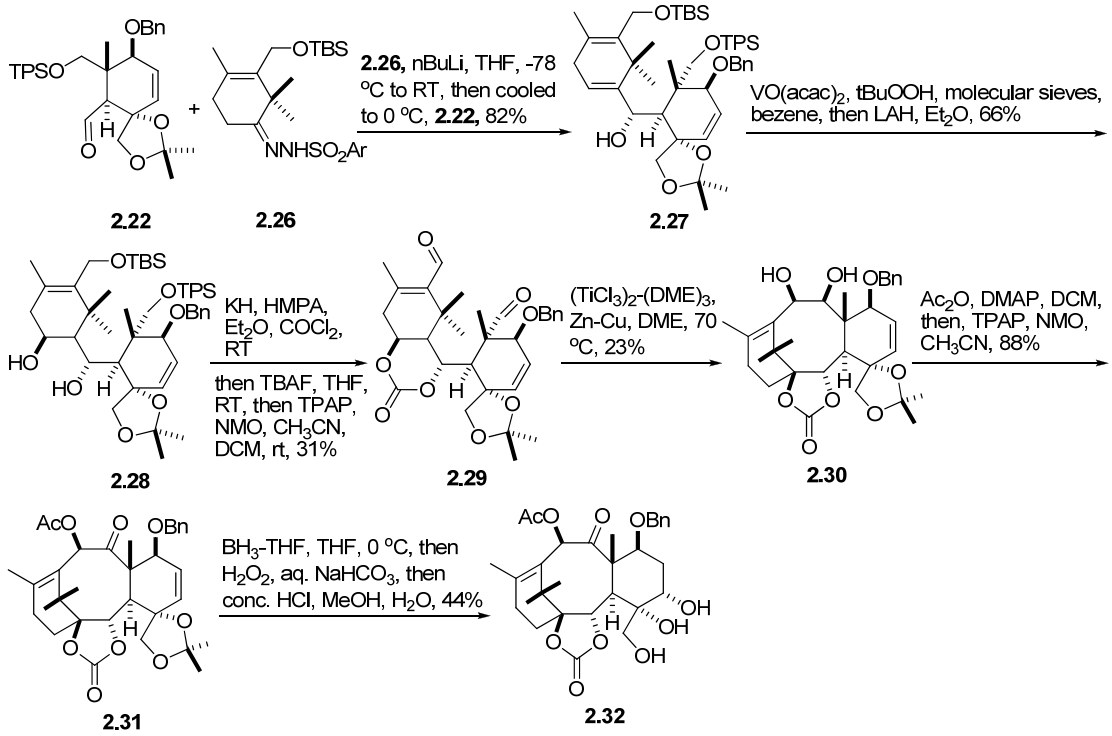
Scheme 2.4 Synthesis of fragment **2.22**

The Nicolaou synthesis started from a previously reported intermediate **2.18**, which can be readily obtained from commercially available materials. After 7 steps, intermediate **2.18** was converted to ring C precursor **2.22** in 38% overall yield (Scheme 2.4). The synthesis of ring A precursor **2.26** began with two commercially available materials **2.23** and **2.24** in overall 51% yield.



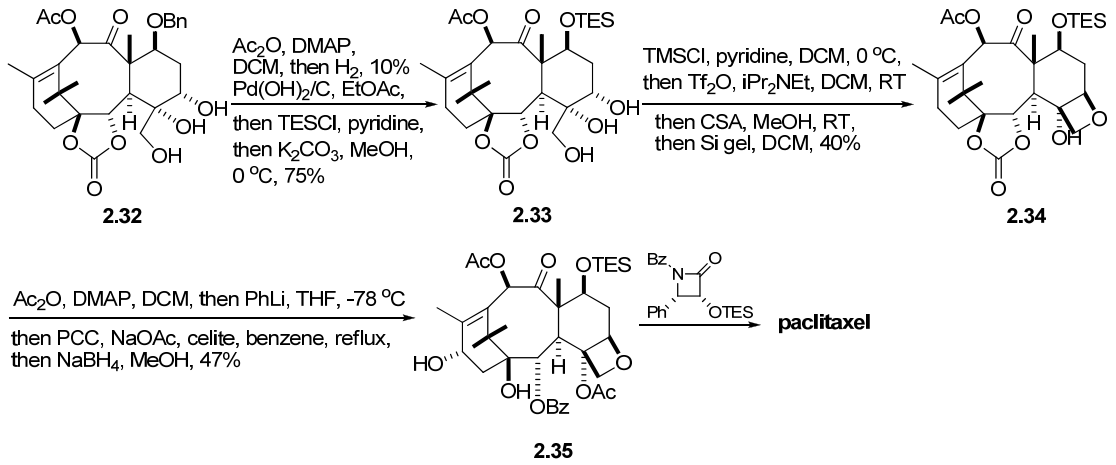
Scheme 2.5 Synthesis of fragment **2.26**

Subsequently, the two precursors were coupled by Shapiro reaction and McMurry coupling to form the ABC ring skeleton in overall 12% yield. The following oxidation of C9 hydroxyl group to a keto group gave intermediate **2.31** in 88% yield, to which the oxetane ring was subsequently installed to establish the ABCD ring skeleton.



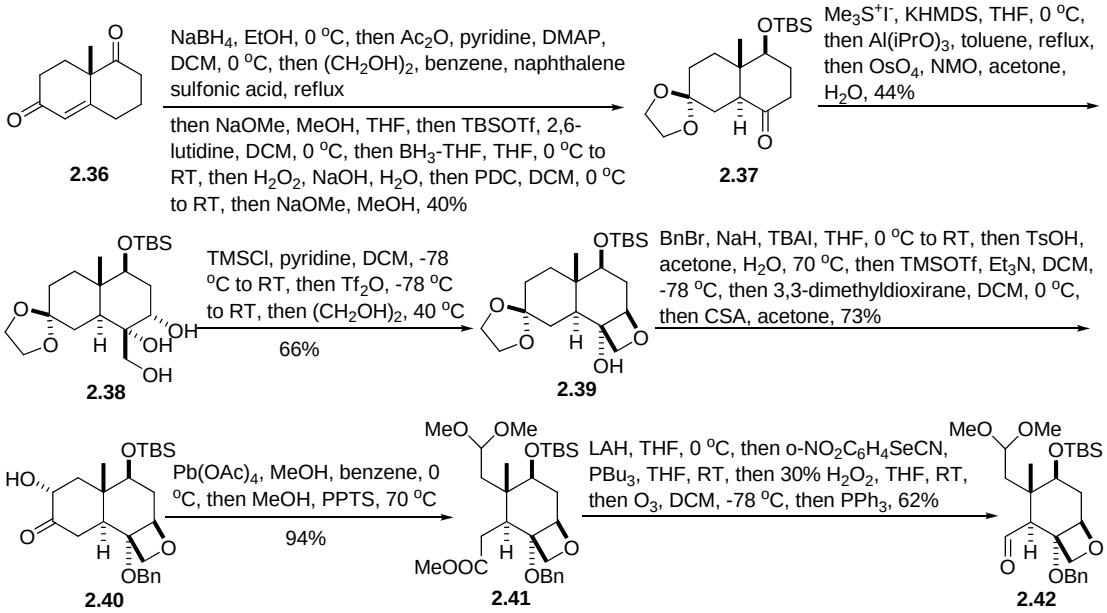
Scheme 2.6 Synthesis of fragment **2.32**

After careful addition of various substituents around ring B and C, intermediate **2.34** was converted to protected baccatin III **2.35** in 47% yield.



Scheme 2.7 Synthesis of paclitaxel

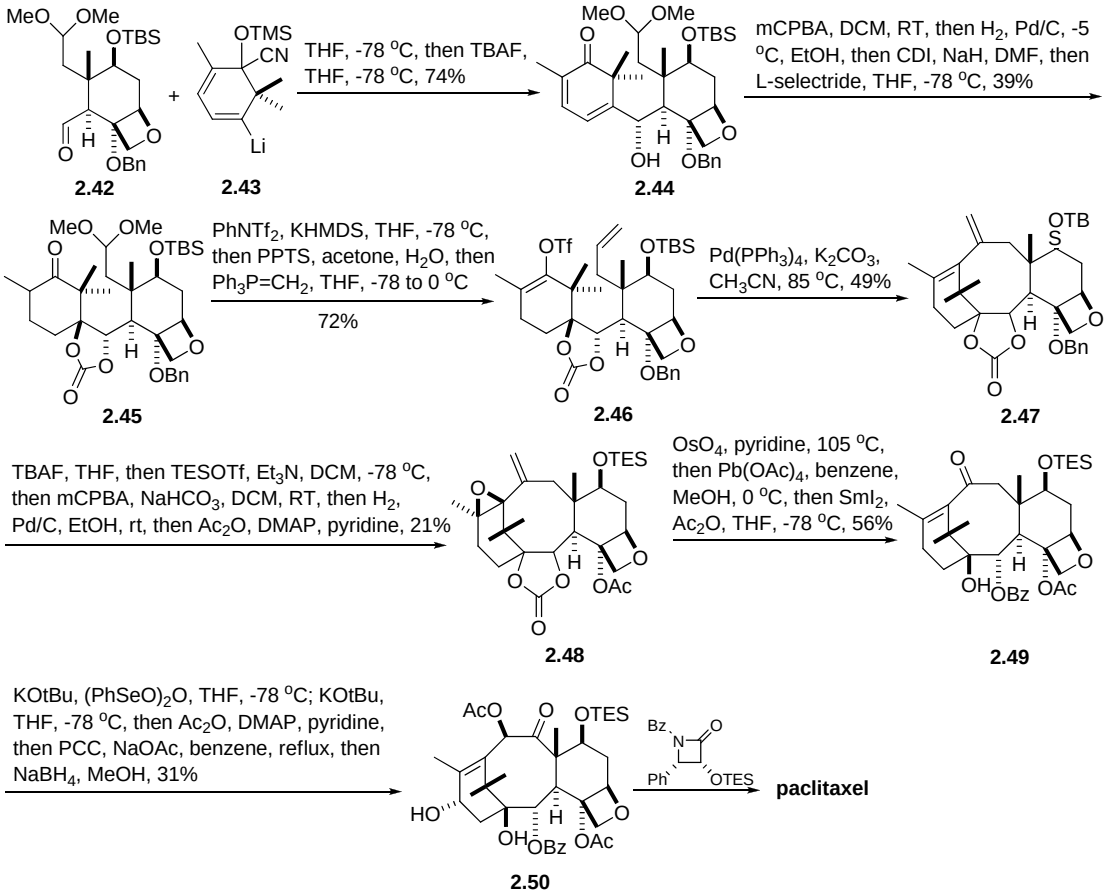
Finally, baccatin III was coupled with β-lactam to attach the side chain to the C13 position, and the following global deprotection completed the Nicolaou synthesis in about 0.1% overall yield from **2.18**.



Scheme 2.8 Synthesis of fragment **2.42**

2.4.1.3 The Danishefsky synthesis

Danishefsky and colleagues published their synthetic approach of paclitaxel in 1996.[36] Their synthetic strategy is based on the retrosynthetic analysis shown in Figure 2.4. In this approach, the C ring and D rings are formed first and then coupled with the A ring, and finally the B ring is established via a ring-closing Heck reaction.



Scheme 2.9 Synthesis of paclitaxel

The Danishefsky synthesis began with the Wieland-Miescher ketone **2.36**, which was converted to **2.38** in 17% yield over ten steps. The D ring (oxetane ring) was formed by

treatment of **2.38** with ethylene glycol at reflux in 69%. Compound **2.39** was then converted to the CD ring precursor **2.43** in 43% yield over ten steps, which was subsequently coupled with the A ring precursor **2.44** giving rise to intermediate **2.45** after ketone deprotection in 74% yield.

After the conversion of **2.45** to **2.48** in 28% yield over six steps, the B ring was formed from **2.48** via a Heck reaction to give rise to **2.49** in 49% yield. The following elaboration of **2.49** provided protected baccatin III **2.50** in 36% yield over thirteen steps. Finally, after coupling with β-lactam followed by global deprotection, **2.15** was converted to paclitaxel in overall 0.18% yield from **3.36**.

Besides the three synthesis mentioned above, other successful syntheses of paclitaxel include the Wender synthesis,[37] the Kuwajima synthesis,[38] and the Mukaiyama synthesis.[39] Their retrosynthetic strategies are summarized in Figure 2.4.

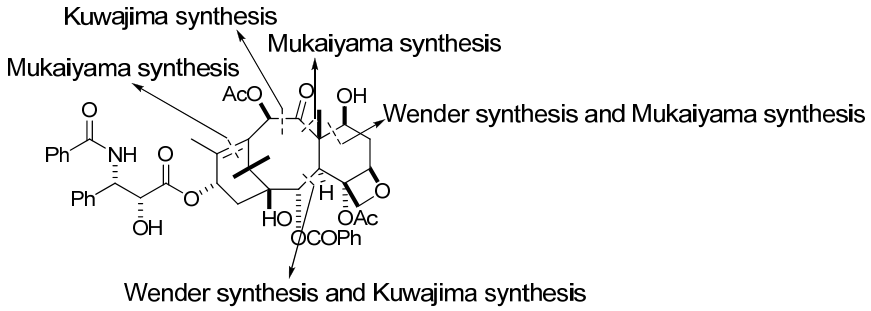


Figure 2.4 Retrosynthesis of paclitaxel

2.4.2 Semisynthesis

Although some successful total synthetic approaches of paclitaxel were developed over the past twenty years, none of them can provide a satisfactory solution to paclitaxel's supply issue due to their low yields and overall length. The supply issue was initially addressed by a semisynthetic method developed by Potier in 1988.[40] The semisynthesis starts with 10-deacetylbaccatin III (10-DAB) which can be obtained from the leaves of the European Yew, *Taxus baccata*, in good yield (1g/kg). In comparison with the slow growing Pacific Yew, the European Yew grows easily and rapidly, and more importantly, it can be harvested without killing the plant, which makes the European Yew an excellent raw material source. Up to date, several semisynthsis of paclitaxel or docetaxel based on Potier's strategy have been reported and will be discussed in this section.

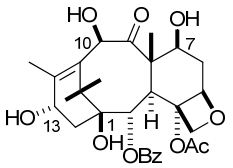


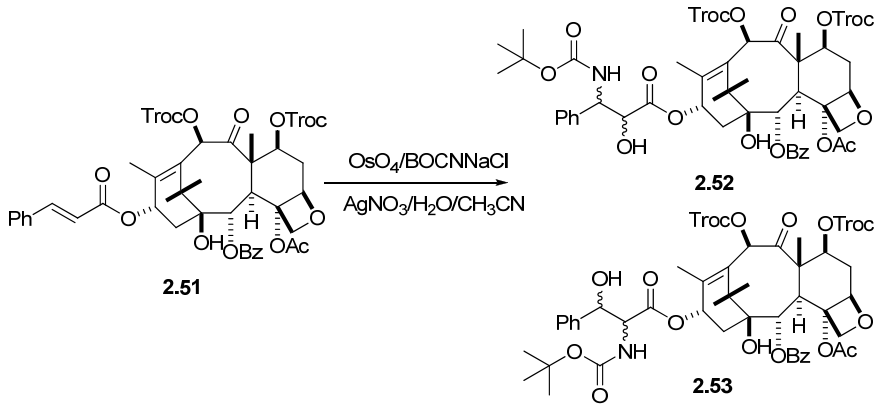
Figure 2.5 10-deacetyl baccatin III (10-DAB)

As shown above, 10-DAB has four hydroxyl groups, and it was found that in general the reactivity sequence of these four groups is C7 > C10 > C13 > C1. Since the side chain needs to be installed selectively at the C13 position, the more reactive hydroxyl groups at the C7 and C10 positions must be protected before introducing the side chain. The most

commonly used protecting strategy for the synthesis of paclitaxel or docetaxel is that the C7 hydroxyl is protected as the TES ether to give 7-TES-10-deacetyl baccatin III, after which the C10 hydroxyl is acetylated. Docetaxel has a free hydroxyl group at the C10 position, thus the strategy for synthesis of docetaxel is that both of the C7 and C10 are protected with the trichloroethoxycarbonyl (Troc) group and later the Troc groups are removed simultaneously at the final stage to give free hydroxyl groups at the C7 and C10 positions.

2.4.2.1 Cinnamate method

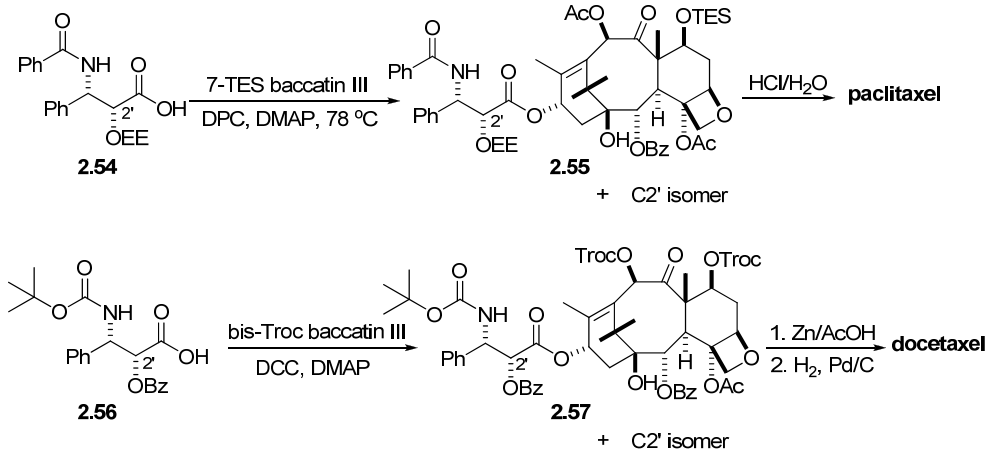
The cinnamate method was used early to set up the two chirality centers at the C2′ and C3′ positions via a Sharpless oxyamination (Scheme 2.10).[4] However, this method gave the desired product in low yield and poor stereoselectivity.[41] The attempt to improve yield and stereoselectivity through the addition of chiral tertiary bases did improve the overall yield, but failed to increase the selectivity. People came to realize that the pre-setup chirality centers on the side chain precursor were necessary for a successful semisynthesis. However, this synthesis was the route by which the semisynthetic analog docetaxel was discovered.



Scheme 2.10 The cinnamate approach to docetaxel

2.4.2.2 The acyclic side chain precursors

The first acyclic side chain precursor with correct stereochemistry was reported by Greene and Potier. This precursor with its C2′ hydroxyl protected as the ethoxyethyl (EE) ether was coupled to the C13 position of 7-TES baccatin III with dipyridylcarbonate (DPC) and dimethylaminopyridine (DMAP) in toluene to give protected paclitaxel in 80% yield (Scheme 2.11). This was subsequently treated with 0.5 M HCl in ethanol at 0 ˚C to remove the EE and TES protecting groups and produce paclitaxel in 89% yield.



Scheme 2.11 The acyclic side chain precursors

Later on, a similar side chain precursor with C2′ protected with a benzoyl group was used to synthesize docetaxel. The coupling of the side chain and Troc protected 10-DAB was achieved by treatment with dicyclohexylcarbodiimide (DCC) and DMAP to give protected docetaxel, which was deprotected by treatment with Zn/AcOH followed by hydrogenation to give docetaxel in 61% yield (Scheme 2.11). The drawback of these two methods includes easy C2′ isomerization during the coupling and the need for excess coupling agents to achieve the coupling and avoid the formation of byproducts.[42]

2.4.2.3 Oxazolidines

The coupling of the side chain and baccatin III is difficult due to the steric hindrance at either C13 position of baccatin III or the carboxyl group of the side chain. To minimize this steric hindrance, a side chain precursor with the C2′ hydroxyl and C3′ amine protected by a single protecting group to form an oxazolidine ring was developed to reduce the steric congestion of the carboxyl group of the side chain, as shown in Figure 2.6.[43]

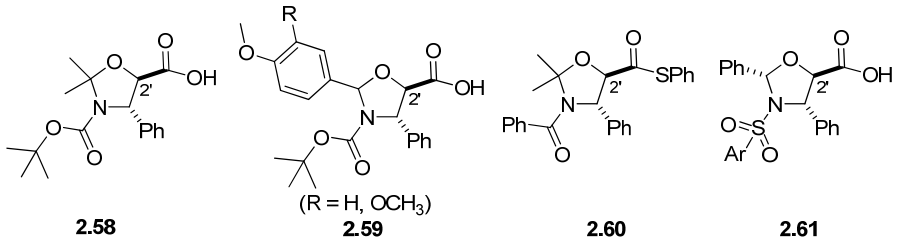
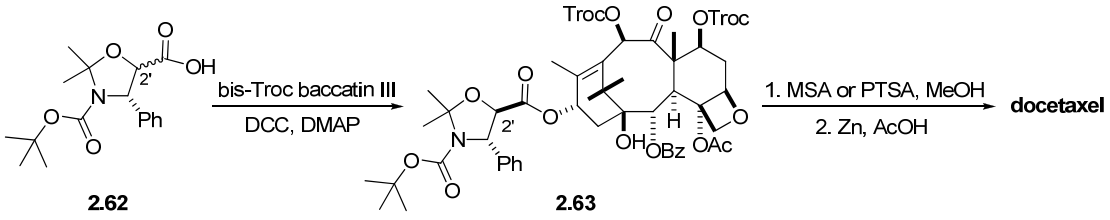


Figure 2.6 The oxazolidine side chain precursors

Another advantage of this strategy is that the *trans* relationship of the phenyl and

carboxyl groups reduces the risk of C2′ isomerization during the coupling. Interestingly, the C2′ stereochemistry was found to be unnecessary to establish the correct C2′ chirality on the side chain. Both (2′*S*,3′*S*)-oxazolidine and (2′*R*,3′*S*)-oxazolidine gave the same desired product in similar yields.

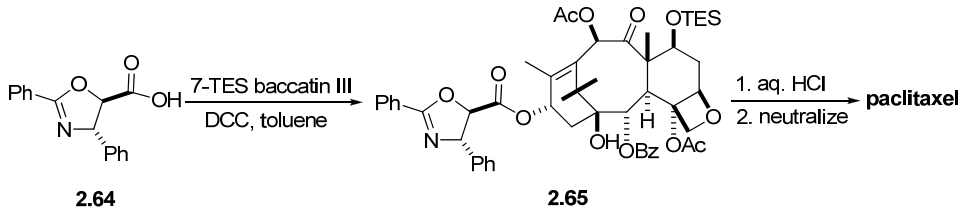


Scheme 2.12 The oxazolidine approach

2.4.2.4 Oxazolines

This approach was reported by the Bristol-Myers Squibb group, Kingston and Gennari.[44] They utilized oxazolines as the side chain precursor (Scheme 2.13).

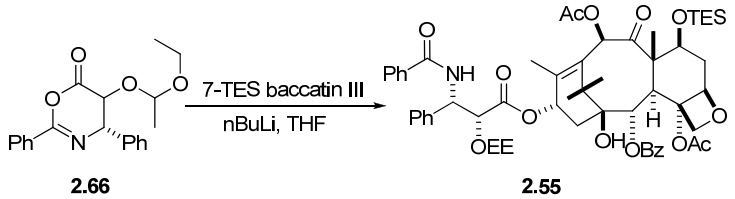
One of the advantages of this method is its atom-economy since the protecting group in the oxazoline is also part of the structure of paclitaxel. On the other hand, the planar five-membered ring precursor minimizes the steric hindrance of the carboxyl group and thus improves the efficiency of the coupling of side chain and baccatin III. However, it is still not the most satisfactory method since it generally requires excessive amount of side chain precursor which must be removed by chromatography after the coupling, and the major byproduct is hard to separate.



Scheme 2.13 The oxazoline approach to paclitaxel

2.4.2.5 Oxazinones

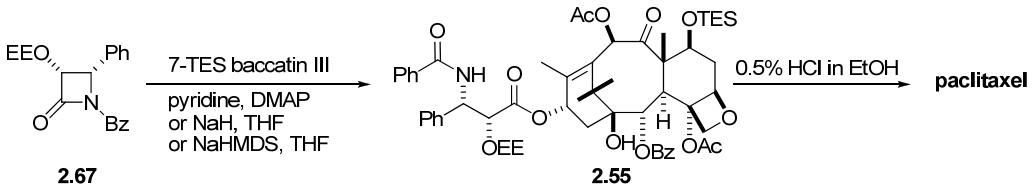
As shown in Scheme 2.14, an oxazonone was used as an activated ester coupling with baccatin III to prepare paclitaxel. This method was reported by Holton and Swindell.[45]



Scheme 2.14 The oxazinone approach

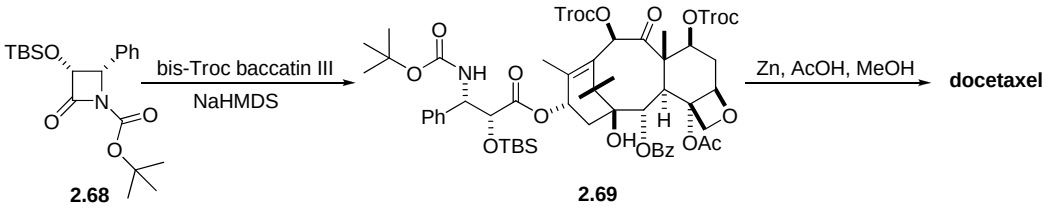
2.4.2.6 β-lactam

Holton and co-workers first reported the use of a β-lactam as the side chain precursor. In their original method, five equivalents of the β-lactam precursor was coupled with 7-TES baccatin III by DMAP in pyridine, giving protected paclitaxel in 90% yield. This method is not suitable for docetaxel synthesis because the *tert*-butoxyl carbamate is not as activating as the benzoyl group for DMAP-mediated coupling reaction.



Scheme 2.15 The EE protected β-lactam precursor

Later, Ojima reported the use of NaH or NaHMDS as the base for connection of β-lactam to protected baccatin III, and this allows the synthesis of docetaxel through this strategy.[46] A revised method as shown below used a TBS protected β-lactam as the side chain precursor, instead of the EE protected one. This avoided the production of diastereomers formed by the EE protected β-lactam and allowed the synthesis of docetaxel. The β-lactam method has become the most widely used method due to the easy synthesis of the β-lactam and the good yield in coupling the β-lactam with protected baccatin III.



Scheme 2.16 The TBS protected β-lactam precursor

2.5 Summary

As two of the most successful anticancer drugs, paclitaxel and docetaxel have been extensively studied during the past three decades. The discovery of their mechanism of action gave us a new drug target in the fight against cancer and prompted scientists to search for better microtubule-interactive agents as anticancer drugs. However, neither paclitaxel nor docetaxel is a perfect drug due to their high toxicity and side effects. Therefore, futher investigations are still needed to improve drug potency and reduce the side effects.

References

1. Wall, M. E.; Wani, M. C., Recent progress in plant anti-tumor agents. *153rd National Meeting of the American Chemical Society*, Miami Beach, FL, **1967**.
2. Wani, M. C.; Taylor, H. L.; Wall, M. E.; Coggon, P.; McPhail, A. T., Plant antitumor agents. VI. The isolation and structure of taxol, a novel antileukemic and antitumor agent from *taxus brevifolia*. *J. Am. Chem. Soc.* **1971**, *93*, 2325-2327.
3. Schiff, P. B.; Fant, J.; Horwitz, S. B., Promotion of microtubule assembly in vitro by taxol. *Nature* **1979**, *277*, 665-667.
4. Kant, J.; O'Keeffe, W. S.; Chen, S.-H.; Farina, V.; Fairchild, C.; Johnston, K.; Kadow, J. F.; Long, B. H.; Vyas, D., A chemoselective approach to functionalize the C-10

position of 10-deacetylbaccatin iii. Synthesis and biological properties of novel C-10 taxol analogues. *Tetrahedron Lett.* **1994**, *35*, 5543-5546.

5. Clarke, S. J.; Rivory, L. P., Clinical pharmacokinetics of docetaxel. *Clin. Pharmacokinet.* **1999**, *36*, 99-114.
6. Rao, S.; Horwitz, S. B.; Ringel, I., Direct photoaffinity labeling of tubulin with taxol. *J. Natl. Cancer Inst.* **1992**, *84*, 785-788.
7. Nicolaou, K. C.; Dai, W.-M.; Guy, R. K., Chemistry and biology of taxol. *Angew. Chem. Int. Ed. Engl.* **1994**, *33*, 15-44.
8. (a) Schiff, P. B.; Horwitz, S. B., Taxol stabilizes microtubules in mouse fibroblast cells. *Proc. Natl. Acad. Sci.* **1980**, *77*, 1561-1565; (b) Nogales, E.; Whittaker, M.; Milligan, R. A.; Downing, K. H., High-resolution model of the microtubule. *Cell* **1999**, *96*, 79-88.
9. Amos, L. A.; Löwe, J., How taxol stabilises microtubule structure. *Chem. & Biol.* **1999**, *6*, 65-69.
10. Derry, W. B.; Wilson, L.; Jordan, M. A., Substoichiometric binding of taxol suppresses microtubule dynamics. *Biochem.* **1995**, *34*, 2203-2211.
11. Parness, J.; Horwitz, S. B., Taxol binds to polymerized tubulin in vitro *J. Cell Biol.* **1981** *91*, 479-487
12. Andreu, J. M.; Bordas, J.; Diaz, J. F.; Garcia de Ancos, J.; Gil, R.; Medran, F. J.; Nogales, E.; Pantos, E.; Towns-Andrews, E., Low resolution structure of microtubules in solution. Synchrotron X-ray scattering and electron microscopy of

taxol-induced microtubules assembled from purified tubulin in comparison with glycerol and map-induced microtubules. *J. Mol. Biol.* **1992**, *226*, 169-184.

13. (a) Hsu, S. I.; Lothstein, I.; Horwitz, S. B., Dierential overex-pression of three mdr gene family members in multidrug-resistant J774.2 mouse cells. Evidence that distinct P-glycoprotein precursors are encoded by unique mdr genes. *J. Cell Biochem.* **1989**, *264*, 12053-12062; (b) Roy, S. N.; Horwitz, S. B., A phosphoglycoprotein associated with taxol resistance in J774.2 cells. *Cancer Res.* **1985**, *45*, 3856-3863; (c) Lothstein, L.; Hsu, S. I.; Horwitz, S. B.; Greenberger, L. M., Alternate overexpression of two P-glycoprotein genes is associated with changes in multidrug resistance in a J7742 cell line. *J. Biol. Chem.* **1990**, *265*, 1821-1831.
14. Schibler, M. J.; Cabral, F., Taxol-dependent mutants of chinese hamster ovary cells with alterations in alpha- and beta-tubulin. *J. Cell. Biol.* **1986**, *102*, 1531-1552.
15. (a) Donehower, R. C.; Rowinsky, E. K.; Grochow, L. B.; Longnecker, S. M.; Ettinger, D. S., Phase I trial of taxol in patients with advanced cancer. *Cancer Treatment Reports* **1987**, *71*, 1171-1177; (b) Rowinsky, E. K.; Burke, P. J.; Karp, J. E.; Tucker, R. W.; Ettinger, D. S.; Donehower, R. C., Phase I and pharmacodynamic study of taxol in refractory acute leukemias *Cancer Res* **1989** *49*, 4640-4647 ; (c) Grem, J. L.; Tutsch, K. D.; Simon, K. J.; Alberti, D. B.; Willson, J. K. V.; Tormey, D. C.; Swaminathan, S.; Trump, D. L., Phase I study of taxol administered as a short IV infusion daily for 5 days. *Cancer Treatment Reports* **1987**, *71*, 1179-1184.
16. (a) Gulley, J.; Dahut, W. L., Chemotherapy for prostate cancer: finally an advance!

Am. J. Therap. **2004**, *11*, 288-294; (b) Wolff, J. M., Significance of docetaxel in the chemotherapy of hormone-refractory prostate cancer. *Onkologie* **2003**, *26*, 37-40; (c) Khan, M. A.; Carducci, M. A.; Partin, A. W., The evolving role of docetaxel in the management of androgen independent prostate cancer. *J. Urology* **2003**, *170*, 1709-1716; (d) Beer, T. M.; El-Geneidi, M.; Eilers, K. M., Docetaxel (Taxotere) in the treatment of prostate cancer. *Exp. Rev. Anticancer Ther.* **2003**, *3*, 261-268; (e) Petrylak, D. P., Chemotherapy for the treatment of hormone-refractory prostate cancer. *Eur. Urology Supp.* **2002**, *1*, 15-23; (f) Tannock, L. F.; de Wit, R.; Berry, W. R.; Horti, J.; Pluzanska, A.; Kim, N.; Oudard, S.; Theodore, C.; James, N. D.; Turesson, I.; Rosenthal, M. A.; Eisenberger, M. A., Docetaxel plus prednisone or mitoxantrone plus prednisone for advanced prostate cancer. *N. Engl. J. Med.* **2004**, *351*, 1502-1512; (g) Petrylak, D. P.; Tangen, C. M.; Hussain, M. H. A.; Lara, P. N., Jr.; Jones, J. A.; Taplin, M. E.; Burch, P. A.; Berry, D.; Moinpour, C.; Kohli, M.; Benson, M. C.; Small, E. J.; Raghavan, D.; Crawford, E. D., Docetaxel and estramustine compared with mitoxantrone and prednisone for advanced refractory prostate cancer. *N. Engl. J. Med.* **2004**, *351*, 1513-1520.

17. Harris, J. W.; Katki, A.; Anderson, L. W.; Chmurny, G. N.; Paukstelis, J. V.; Collins, J. M., Isolation, structural determination, and biological activity of 6a-hydroxytaxol, the principal human metabolite of taxol. *J. Med. Chem.* **1994**, *37*, 706-709.

18. (a) Gueritte-Voegelein, F.; Guenard, D.; Lavelle, F.; Le Goff, M.-T.; Mangatal, L.; Potier, P., Relationships between the structure of taxol analogues and their antimitotic

activity. *J. Med. Chem.* **1991**, *34*, 992-998; (b) Lataste, H.; Senilh, V.; Wright, M.; Guenard, D.; Potier, P., Relationships between the structures of taxol and baccatine III derivatives and their in vitro action on the disassembly of mammalian brain and physarum amoebal microtubules. *Proc. Natl. Acad. Sci.* **1984**, *81*, 4090-4094; (c) Mathew, A. E.; Mejillano, M. R.; Nath, J. P.; Himes, R. H.; Stella, V. J., Synthesis and evaluation of some water-soluble prodrugs and derivatives of taxol with antitumor activity. *J. Med. Chem.* **1992**, *35*, 145-151; (d) Kingston, D. G. I.; Samaranayake, G.; Ivey, C. A., The chemistry of taxol, a clinically useful anticancer agent. *J. Nat. Prod.* **1990**, *53*, 1-12; (e) Senilh, V.; Blechert, S.; Colin, M.; Guenard, D.; Picot, F.; Potier, P.; Varenne, P., Mise en evidence de nouveaux analogues du taxol extraits de taxus baccata. *J. Nat. Prod.* **1984**, *47*, 131-137; (f) Chaudhary, A. G.; Rimoldi, J. M.; Kingston, D. G. I., Modified taxols. 10. Preparation of 7-deoxytaxol, a highly bioactive taxol derivative, and interconversions of taxol and 7-*epi*-taxol. *J. Org. Chem.* **1993**, *58*, 3798-3799; (g) Chen, S.-H.; Huang, S.; Kant, J.; Fairchild, C.; Wei, J.; Farina, V., Synthesis of 7-deoxy- and 7,10-dideoxytaxol via radical intermediates. *J. Org. Chem.* **1993**, *58*, 5028-5029.

19. Magri, N. F.; Kingston, D. G. I., Modified taxols. 2. Oxidation products of taxol. *J. Org. Chem.* **1986**, *51*, 797-802.

20. (a) Klein, L. L., Synthesis of 9-dihydrotaxol: A novel bioactive taxane. *Tetrahedron Lett.* **1993**, *34*, 2047-2050; (b) Georg, G. I.; Cheruvallath, Z. S.; Vander Velde, D. G.; Himes, R. H., Stereoselective synthesis of 9b-hydroxytaxanes via reduction with

samarium diiodide. *Tetrahedron Lett.* **1995**, *36*, 1783-1786.

21. Chen, S.-H.; Huang, S.; Wei, J.; Farina, V., Serendipitous synthesis of a cyclopropane-containing taxol analog via anchimeric participation of an unactivated angular methyl group. *J. Org. Chem.* **1993**, *58*, 4520-4521.
22. Chen, S.-H.; Fairchild, C.; Mamber, S. W.; Farina, V., Taxol structure-activity relationships: synthesis and biological evaluation of 10-deoxytaxol. *J. Org. Chem.* **1993**, *58*, 2927-2928.
23. Margraff, R.; Bezard, D.; Bourzat, J. D.; Commercon, A., Synthesis of 19-hydroxy docetaxel from a novel baccatin. *Bioorg. Med. Chem. Lett.* **1994**, *4*, 233-236.
24. Kingston, D. G. I.; Chordia, M. D.; Jagtap, P. G., Synthesis and biological evaluation of 1-deoxypaclitaxel analogues. *J. Org. Chem.* **1999**, *64*, 1814-1822.
25. Chen, S.-H.; Wei, J.-M.; Farina, V., Taxol structure-activity relationships: synthesis and biological evaluation of 2-deoxytaxol. *Tetrahedron Lett.* **1993**, *34*, 3205-3206.
26. Chaudhary, A. G.; Gharpure, M. M.; Rimoldi, J. M.; Chordia, M. D.; Gunatilaka, A. A. L.; Kingston, D. G. I.; Grover, S.; Lin, C. M.; Hamel, E., Unexpectedly facile hydrolysis of the 2-benzoate group of taxol and synthesis of analogs with increased activities. *J. Am. Chem. Soc.* **1994**, *116*, 4097-4098.
27. (a) Chordia, M. D.; Kingston, D. G. I., Synthesis and biological evaluation of 2-*epi*-paclitaxel. *J. Org. Chem.* **1996**, *61*, 799-801; (b) Hoemann, M. Z.; Vander Velde, D.; Aube, J.; Georg, G. I.; Jayasinghe, L. R., Synthesis of 13-*epi*-taxol via a transannular delivery of a borohydride reagent. *J. Org. Chem.* **1995**, *60*, 2918-2921.

28. (a) Datta, A.; Jayasinghe, L. R.; Georg, G. I., 4-deacetyltaxol and 10-acetyl-4-deacetyltaxotere: synthesis and biological evaluation. *J. Med. Chem.* **1994**, *37*, 4258-4260; (b) Neidigh, K. A.; Gharpure, M. M.; Rimoldi, J. M.; Kingston, D. G. I.; Jiang, Y. Q.; Hamel, E., Synthesis and biological evaluation of 4-deacetylpaclitaxel. *Tetrahedron Lett.* **1994**, *35*, 6839-6842; (c) Chen, S.-H.; Kadow, J. F.; Farina, V.; Fairchild, C. R.; Johnston, K. A., First syntheses of novel paclitaxel (taxol) analogs modified at the C4-position. *J. Org. Chem.* **1994**, *59*, 6156-6158; (d) Georg, G. I.; Ali, S. M.; Boge, T. C.; Datta, A.; Falborg, L.; Himes, R. H., Selective C-2 and C-4 deacylation and acylation of taxol: the first synthesis of a C-4 substituted taxol analogue. *Tetrahedron Lett.* **1994**, *35*, 8931-8934; (e) Chen, S.-H.; Fairchild, C.; Long, B. H., Synthesis and biological evaluation of novel C-4 aziridine bearing paclitaxel (taxol) analogs. *J. Med. Chem.* **1995**, *38*, 2263-2267; (f) Chen, S.-H.; Wei, J.-M.; Long, B. H.; Fairchild, C. A.; Carboni, J.; Mamber, S. W.; Rose, W. C.; Johnston, K.; Casazza, A. M.; Kadow, J. F.; Farina, V.; Vyas, D.; Doyle, T. W., Novel C-4 paclitaxel (taxol) analogs: Potent antitumor agaents. *Bioorg. & Med. Chem. Lett.* **1995**, *5*, 2741-2746.

29. Samaranayake, G.; Magri, N. F.; Jitrangsri, C.; Kingston, D. G. I., Modified taxols. 5. Reaction of taxol with electrophilic reagents, and preparation of a rearranged taxol derivative with tubulin assembly activity. *J. Org. Chem.* **1991**, *56*, 5114-5119.

30. (a) Kant, J.; Huang, S.; Wong, H.; Fairchild, C.; Vyas, D.; V., F., Studies toward structure-activity relationships of taxol: synthesis and cytotoxicity of taxol analogues

with C-2' modified phenylisoserine side chains. *Bioorg. Med. Chem. Lett.* **1993**, *3*, 2471-2474; (b) Magri, N. F.; Kingston, D. G. I., Modified taxols. 4. Synthesis and biological activity of taxols modified in the side chain. *J. Nat. Prod.* **1988**, *51*, 298-306.

31. Jayasinghe, L. R.; Datta, A.; Ali, S. M.; Zygmunt, J.; Vander Velde, D. G.; Georg, G. I., Structure-activity studies of antitumor taxanes: synthesis of novel C-13 side chain homologated taxol and taxotere analogs. *J. Med. Chem.* **1994**, *37*, 2981-2984.
32. Kingston, D. G. I., History and chemistry. In *Paclitaxel in cancer treatment*, McGuire, W. P.; Rowinsky, E. K., Eds. Marcel Dekker, Inc.: New York, Basel, Hong Kong, **1995**; Vol. 8, pp 1-33.
33. (a) 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., First total synthesis of taxol. 1. Functionalization of the B ring. *J. Am. Chem. Soc.* **1994**, *116*, 1597-1598; (b) 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., First total synthesis of taxol. 2. Completion of the C and D rings. *J. Am. Chem. Soc.* **1994**, *116*, 1599-1600.
34. Nicolaou, K. C.; Yang, Z.; Liu, J. J.; Ueno, H.; Nantermet, P. G.; Guy, R. K.; Claiborne, C. F.; Renaud, J.; Couladouros, E. A.; Paulvannan, K.; Sorensen, E. J., Total synthesis of taxol. *Nature* **1994**, *367*, 630-634.

35. (a) Nicolaou, K. C.; Nantermet, P. G.; Ueno, H.; Guy, R. K.; Couladouros, E. A.; Sorensen, E. J., Total synthesis of taxol. 1. Retrosynthesis, degradation, and reconstitution. *J. Am. Chem. Soc.* **1995**, *117*, 624-633; (b) Nicolaou, K. C.; Liu, J.-J.; Yang, Z.; Ueno, H.; Sorensen, E. J.; Claiborne, C. F.; Guy, R. K.; Hwang, C.-K.; Nakada, M.; Nantermet, P. G., Total synthesis of taxol. 2. Construction of A and C ring intermediates and initial attempts to construct the ABC ring system. *J. Am. Chem. Soc.* **199**5, *117*, 634-644.

36. Danishefsky, S. J.; Masters, J. J.; Young, W. B.; Link, J. T.; Snyder, L. B.; Magee, T. V.; Jung, D. K.; Isaacs, R. C. A.; Bornmann, W. G.; Alaimo, C. A.; Coburn, C. A.; Di Grandi, M. J., Total synthesis of baccatin III and taxol. *J. Am. Chem. Soc.* **1996**, *118*, 2843-2859.

37. Wender, P. A.; Badham, N. F.; Conway, S. P.; Floreancig, P. E.; Glass, T. E.; Houze, J. B.; Krauss, N. E.; Lee, D.; Marquess, D. G.; McGrane, P. L.; Meng, W.; Natchus, M. G.; Shuker, A. J.; Sutton, J. C.; Taylor, R. E., The pinene path to taxanes. 6. A concise stereocontrolled synthesis of taxol. *J. Am. Chem. Soc.* **1997**, *119*, 2757-2758.

38. Kuwajima, I.; Kusama, H., Synthetic studies on taxoids: enantioselective total synthesis of (+)-taxusin and (-)-taxol. *Synlett* **2000**, 1385-1401.

39. Mukaiyama, T.; Shiina, I.; Iwadare, H.; Saitoh, M.; Nishimura, T.; Ohkawa, N.; Sakoh, H.; Nishimura, K.; Tani, Y.-I.; Hasegawa, M.; Yamada, K.; Saitoh, K., Asymmetric total synthesis of taxol. *Chem. Eur. J.* **1999**, *5*, 121-161.

40. Denis, J. N.; Greene, A. E.; Guenard, D.; Gueritte-Voegelein, L. M.; Potier, P., A

highly efficient, practical approach to natural taxol. *J. Am. Chem. Soc.* **1988**, *110*, 5917-5919.

41. Mangatal, L.; Adeline, M.-T.; Guenard, D.; Gueritte-Voegelein, F.; Potier, P., Application of the vicinal oxyamination reaction with asymmetric induction to the hemisynthesis of taxol and analogues *Tetrahedron* **1989**, *45*, 4177-4190
42. Kanawawa, A. M.; Denis, J. N.; Greene, A. E., Highly stereocontrolled and efficient preparation of the protected esterification-ready docetaxel (Taxotere) side chain. *J. Org. Chem* **1994**, *59*, 1238-1240.
43. Commercon, A.; Bezard, D.; Bernard, F.; Bourzat, J. D., Improved protection and esterification of a precursor of the taxotere and taxol side chains. *Tetrahedron Lett.* **1992**, *33*, 5185-5188.
44. (a) Kingston, D. G. I.; Chaudhary, A. G.; Gunatilaka, A. A. L.; Middleton, M. L., Synthesis of taxol from baccatin III via an oxazoline intermediate. *Tetrahedron Lett.* **1994**, *35*, 4483-4484; (b) Gennari, C.; Carcano, M.; Donghi, M.; Mongelli, N.; Vanotti, E.; Vulpetti, A., Taxol semisynthesis: a highly enantio- and diastereoselective synthesis of the side chain and a new method for ester formation at C-13 using thioesters. *J. Org. Chem.* **1997**, *62*, 4746-4755.
45. Swindell, C. S.; Krauss, N. E.; Horwitz, S. B.; Ringel, I., Biologically active taxol analogues with deleted A-ring side chain substituents and variable C-2' configurations. *J. Med. Chem.* **1991**, *34*, 1176-1184.
46. (a) Ojima, I.; Habus, I.; Zhao, M.; Zucco, M.; Park, Y. H.; Sun, C. M.; Brigaud, T.,

New and efficient approaches to the semisynthesis of taxol and its C-13 side chain analogs by means of β-lactam synthon method. *Tetrahedron* **1992**, *48*, 6985-7012; (b) Ojima, I.; Zucco, M.; Duclos, O.; Kuduk, S. D.; Sun, C. M.; Park, Y. H., N-acyl-3-hydroxy-β-lactams as key intermediates for Taxotere and its analogs. *Bioorg. Med. Chem. Lett.* **1993**, *3*, 2479-2482.