Two-Phase Synthesis of (−)-Taxuyunnanine D
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Supporting Information

ABSTRACT: The first successful effort to replicate the beginning of the Taxol oxidase phase in the laboratory is reported, culminating in the total synthesis of taxuyunnanine D, itself a natural product. Through a combination of computational modeling, reagent screening, and oxidation sequence analysis, the first three of eight C−H oxidations (at the allylic sites corresponding to C-5, C-10, and C-13) required to reach Taxol from taxadiene were accomplished. This work lays a foundation for an eventual total synthesis of Taxol capable of delivering not only the natural product but also analogs inaccessible via bioengineering.

Only eight C−H oxygenation events separate the minimally oxidized hydrocarbon taxadiene (1) from Taxol (2). Known by enzymologists as the oxidase phase, the approximate order of oxidation is proposed to occur at C-5, C-10/C-13, C-9, C-7/C-2, and then C-1/C-4/C-20 (Figure 1A). Divergent oxidation pathways and acylation patterns lead to hundreds of members in the taxane family. Intriguingly, the first three of eight oxygenation events involve the formal activation of allylic C−H bonds. In an ongoing effort to replicate the two phases of Taxol biosynthesis in the laboratory, we focused on uncovering the innate reactivity of 1 so as to achieve a controlled oxidation of these positions (C-5, C-10, and C-13). In order to obtain material to launch these investigations, an artificial cyclase phase for taxanes was devised, culminating in the first enantioselective, scalable total synthesis of (+)-taxadiene (1). In fact, decagram-scale reproduction of that route was independently accomplished by Albany Molecular Research Inc. to provide “taxadieneone,” the precursor for 1. We present a systematic approach to mimicking the early stages of the Taxol oxidase phase and may form the basis of an eventual scalable total synthesis of 2.

Extensive empirical studies into the fundamental reactivity of taxadiene (1), computational modeling, and reagent development were all enlisted to achieve the first total synthesis of (−)-taxuyunnanine D (3) in only five steps from 1.

As a doubly unsaturated and strained hydrocarbon, taxadiene (1) is “spring loaded” for oxidation. Thus, numerous possible oxidative pathways are conceivable. Presumably due to the relative ease by which allylic C−H bonds can be broken, C-5, C-10, and C-13 appear to be oxidized first in the biosynthetic pathway. The Croteau/Williams team’s enzymatic studies demonstrated that C-5 of 1 would likely be oxidized first in both the biosynthesis and in the laboratory, and this might be rationalized by relative steric hindrance around the C-11(12) and C-4(5) olefins (Figure 1B). After C-5 oxidation, the question of whether C-10 or C-13 would be more easily oxidized is complicated by potentially competitive C-18 oxidation.

Figure 1. Planning stages for taxane oxidase phase. (A) Retrosynthetic rationale for the oxidase phase toward (−)-taxuyunnanine D (3). (B) Reasoning for the synthetic order of allylic oxidation: C-5 (steric hindrance, based on MM2-minimized model of 1), C-13 (most stable radical), and then C-10 (C-13 ketone stabilizes C-10 radical). ∆∆G values are relative energies of indicated allylic radical species in kcal/mol (see Supporting Information, SI).

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oxidation. The relative C−H bond strength for radical oxidation reactions can be estimated by comparing the stability of the radical species resulting from homolytic C−H bond cleavage. DFT calculations (using Gaussian099 and UB3LYP/6-31+G(d,p)9 level of theory) modeling such radical species indicate a radical centered on C-13 of 4 (ΔG for C-13 hydrogen atom abstraction, relative to that on C-10 and C-18, is 0 kcal/mol) would be more stable than that centered on C-10 or C-18 (ΔΔG = 10.6 and 6.4 kcal/mol, respectively). This might be rationalized by the additional strain of converting the sp3-hybridized C-10 methylene to an sp2-hybridized methine, in the already strained eight-membered ring. Such strain might even be enough to outweigh any preference for a secondary allylic radical over a primary one. After C-13 oxidation, there are only two probable carbon atoms subject to allylic oxidation, and DFT calculations suggest that a C-13 ketone might help drive selectivity to C-10 oxidation if this substrate were to undergo a radical-based oxidation (compare 5 and 6). With this basic reasoning in mind, extensive empirical investigations began.

Over the course of hundreds of experiments, five different mechanism-based categories of allylic oxidation10 were evaluated for each of the consecutive targeted oxidations (Table 1, see also SI for an extended summary): (1) π-allyl metal complex oxidation, (2) stoichiometric transition-metal-mediated single-electron oxidation, (3) radical halogenation, (4) transition-metal-catalyzed hydrogen abstraction, and (5) pericyclic oxidation. The screening strategy thus required that each category of oxidant be screened for each allylic C−H bond.11 In accord with initial predictions, the C-5 position of 1 (prepared as described in ref 4) was exclusively oxidized first using Pd-catalyzed allylic acetoxylation under conditions pioneered by Åkermark and Bäckvall.12 All other attempts at C-5 oxidation were unsuccessful, leading to either decomposition or indecipherable product mixtures. With a solution to the C-5 oxidation in hand, this product was then evaluated under the five oxidant categories mentioned above. With halogenation conditions, the electron-rich C-11(12) olefin reacted preferentially to allylic positions. Pericyclic oxidations such as SeO2 and 1O2 allylic oxidation reactions were selective for C-18 oxidation, and catalytic metal-mediated oxidation only gave degradation. A chromium-based oxidant was identified to selectively oxidize C-13 (vide infra). Although transition-metal oxidants such as Cr(VI) oxides are known to effect C-13 oxidation,13 this has never been reported on such minimally oxidized taxanes. Finally, the product containing both C-5 and C-13 oxidation was extensively screened across the same panel in order to functionalize C-10. These extensive trials demonstrated the singularity of each of these allylic positions and laid

| category | conditions | 1 [C-5] | 4 [C-13] | 5 [C-10] |
|----------|------------|---------|----------|----------|
| 1        | Pd(OAc)2, BQ | yes     | no       | no       |
| 2        | Cr3 or CrV | no      | yes      | no       |
| 3        | NBS, (BzO)2 | no      | no       | yes      |
| 4        | M cat., ROOR’ | no      | no       | no       |
| 5        | SeO2 or O2  | no      | no       | no       |

Panel was also employed on substrate 6 with no success. See ref 14. 
C-18 was cleanly oxidized.

Scheme 1. Synthesis of (−)-Taxuyunnanine D (3) from (+)-Taxadiene (1)4

(a) Pd(OAc)2, p-benzoquinone (BQ), anisole, acetic acid, 50°C; (b) Cr(V) reagent 9, MnO2, 15-crown-5, trifluorotoluene, 80°C; (c) NBS, benzoyl peroxide, CCl4, reflux; then AgOTf, Et3SiOH, toluene, 0°C; (d) DIBAL, toluene, −78°C; then MeOH, 0°C; then Ac2O, DMAP, Et3N; (e) IBX, DMSO, 80°C.
Scheme 2. Cr(V) Complex 9 Continued Studiesa

A. Cr(VI) oxidation of 4 with conventional reagents

\[ \begin{array}{c}
4 \quad \text{PCC or} \quad \text{CrO}_3 \cdot \text{DMP} \\
\quad \text{OAc} \\
\quad \text{NaOAc, DCM, 0°C} \\
\quad \text{7, 1:1:1} \end{array} \]

B. A rarely employed Cr(V)-complex

\[ \begin{array}{c}
\text{[Cr(VI)]} \quad \text{[Cr(V)]} \\
\text{16} \quad \text{[ref 17d]} \\
\text{[Na}_2\text{CrO}_7 \text{]} \\
\text{[acetone]} \\
\text{[NaO]} \\
\text{[O]} \end{array} \]

C. Proposed mechanism for observed Cr(V) allylic oxidation

\[ \begin{array}{c}
\text{4} \quad \text{1. SET} \quad 2. \text{-H}^+ \\
\text{5} \quad \text{13} \end{array} \]

D. Divergent reactivity of Cr(VI) and Cr(V)

\[ \begin{array}{c}
\text{18} \quad \text{aPCC} \quad \text{17} \quad \text{bcomplex 9} \\
\text{[Cr(VI)]} \quad \text{[Cr(V)]} \quad \text{[Cr(V)]} \\
\text{19} \quad \text{(59%)} \quad \text{(44%)} \end{array} \]

a(A) Byproducts 7 and 8 from Cr(VI) oxidation. (B) Synthesis of 9. (C) Proposed mechanism for the formation of 10 with Cr(V) reagent 9. (D) Orthogonal reactivity of Cr(VI) and Cr(V) reagents. (a) PCC, NaOAc, DCM, 0°C; (b) Cr(V) reagent 9, MnO2, 15-crown-5, triethylsilanol, 80°C.

the groundwork to achieve the total synthesis of a taxane expressing oxygenation at only the C-5, C-10, and C-13 positions: taxuyunnanine D (3).

The synthesis, outlined in Scheme 1, commences with the C-5 oxidation discussed above (Step 1). A chemical counterpart to the enzymatic C-5 oxidation of \( \Delta^{15} \)-taxadiene is without precedent.14 Thus, palladium-catalyzed acetoxilation with \( \text{Pd(OAc)}_2 \), benzoquinone, and acetic acid afforded the C-5\( \alpha \)-acetate 4 in 35% yield with no other isomers observed. After extensive screening of solvents, additives, and co-oxidants, it was discovered that addition of four equivalents of anisole increased the yield from \( \sim 35\% \) to 49%. The origin of this improvement is currently under investigation, but it was qualitatively observed that less palladium black was produced when anisole is employed, implying that anisole may facilitate reoxidation of \( \text{Pd}(0) \) to \( \text{Pd}(II) \) in this case.

Step 2, the oxygenation of C-13, proved to be quite challenging. C-13 allylic oxidation is known using chromium(VI) oxide 3,5-dimethylpyrazole complex \( 13a \) (CrO\(_3\)·DMP) or pyridinium chlorochromate \( 13b \) (PCC), but both transforma-
Babler—Dauben oxidative rearrangement to give enone, but Cr(V) gave direct allylic oxidation product. Neither reaction showed any trace of the other’s product in their respective crude NMR spectrum (see SI). This implies that SET occurs faster than binding to the alcohol to effect oxidative rearrangement or binding to an alcohol is simply unproductive toward the Babler—Dauben reaction. This might be explained by a different oxidation potential required for SET, unavailable binding sites on complex, or incorrect geometry for allylic transposition if an alkoxide complex is formed. This reactivity with tertiary allylic alcohols appears to be unknown in the literature and should prove useful for such transformations.

The study of Taxol’s biosynthesis is still an area of immense interest from a bioengineering standpoint. The focus of our work is to understand and learn how the conversion of a minimally oxidized taxane to Taxol might be achieved entirely toward the Babler or binding to an alcohol is simply unproductive. This realization, however, required extensive groundwork to access higher oxidized taxane natural products and analogs currently inaccessible through biosynthesis.

ASSOCIATED CONTENT
1 Supporting Information
Experimental procedures, analytical data, and full ref 8. This material is available free of charge via the Internet at http://pubs.acs.org.

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