ASYMMETRIC APPROACHES TO THE TETRACYCLINE ANTIBIOTICS

Reported by Hahdi H. Perfect October 3, 2005

INTRODUCTION

The tetracycline class of antibiotics comprise a distinct family of substituted hydronaphthacene compounds produced by strains of *Streptomyces* bacteria. They were first discovered in 1948 by Duggar and coworkers,[1] with the isolation of chlorotetracycline (**1)**. Two years later, Finlay and coworkers reported the isolation of terramycin (**2)** produced by *Streptomyces rimosus.*[1] In 1953, Woodward and coworkers were able to deduce the structures of **1** and **2** via IR, UV, and chemical degradation studies.[2] The tetracycline antibiotics are characterized structurally by a highly functionalized A-ring that is cis-fused to the B-ring, a C-ring containing a tetrasubstituted stereogenic center and a D ring that is aromatic. These natural products have been subject of a great deal of synthetic work, but this review will focus primarily on asymmetric syntheses.

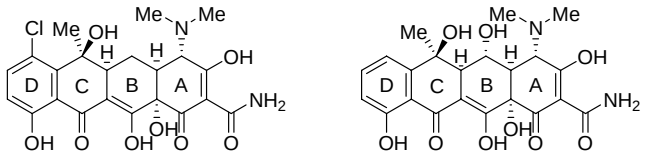

Figure 1. The Structures of Chlorotetracycline **1** and Terramycin **2.**

BIOLOGICAL ACTIVITY

Members of the tetracycline family have long been known to inhibit protein synthesis in bacteria by binding to the 30S subunit of the bacterial ribosome.[1,3,4,5] From a structure-activity perspective, a tetracycline molecule has two distinct regions: the upper half, termed the non-keto-enol system, can be modified to increase biological activity; the lower half, however, needs to remain intact for biological activity to be preserved (Figure 1).[1] Tetracyclines are selectively toxic towards bacteria because of structural differences between ribosomal RNA from bacteria vs. eukaryotic cells, and high drug concentrations that develop in susceptible bacterial cells.[4] This selectivity gives them low toxicity towards eukaryotic cells, an attractive feature for antibiotics. Recently, details of the binding of a tetracycline to the 30S subunit was elucidated by Boderson and coworkers using X-ray crystallography.[5] As expected from structure-activity studies, the majority of the interactions with the 30S subunit are through hydrogen bonding with the keto-enol system on the lower half of the skeleton. Because of their potent activity, tetracyclines have been widely used in the medical and veterinary fields.[3] Extensive use

of these antibiotics, however, has lead to the appearance of resistant strains of bacteria, a problem that is being addressed by the development of new derivatives. The most promising derivatives are the glycyclines,[6-10] which contain a glycyl moiety at the C-9 postion. As of 2004, tigecycline **3** is in Phase 3 clinical trials for urinary tract, abdominal, and skin infections, and appears to have minimal side effects.[8]

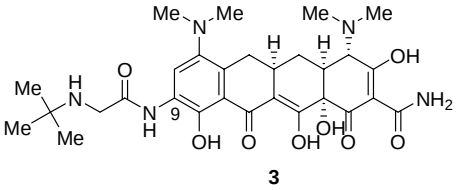

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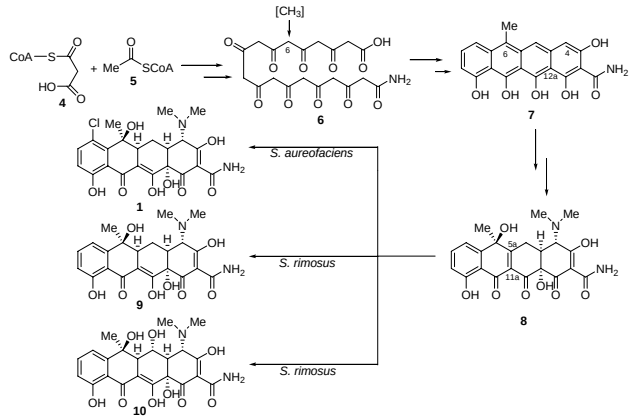
Figure 2. Structure of Tigecycline

BIOSYNTHETIC PATHWAY

The biosynthesis of tetracyclines was first studied by McCormick and coworkers, and was further elaborated by the Hostalek group.[1] The synthesis of a polyketide chain is carried out by sequential addition of acetyl-CoA units **5** to malonyl-CoA **4** (Scheme 1). The resulting polyketide **6** undergoes a series of condensation reactions catalyzed by ketoreductases and aromatases to produce the tetracycline nucleus. The exact details of these processes remain unknown. Polyol **7** is the first isolable and distinct naphthacene ring in this pathway. The C-4 position is then oxygenated via a hydroxylase, followed by other oxidation reactions that install a hydroxyl group at the C-12a position and a ketone at the C-4 position. Amination is then carried out at the C-4 position, followed by methylation of the amine. Oxidation of the C-6 position installs a hydroxyl group and places a double bond between C-5a-C-11a, as shown in enone **8**.

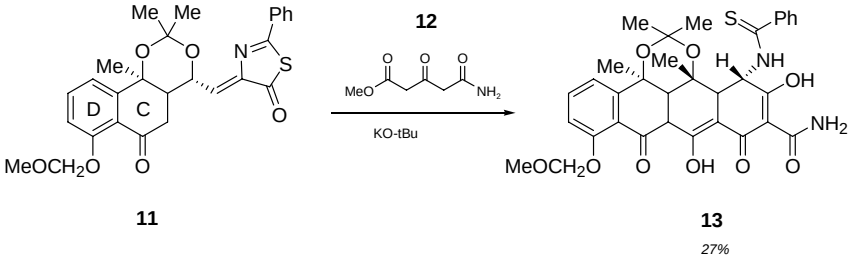
Scheme 1. The Biosynthesis of Chlorotetracycline 1, Tetracycline 9, Terramycin 10.
Enone **8** is then transformed by various bacteria into chlorotetracycline **1**, tetracycline **9**, and oxytetracycline **10**.



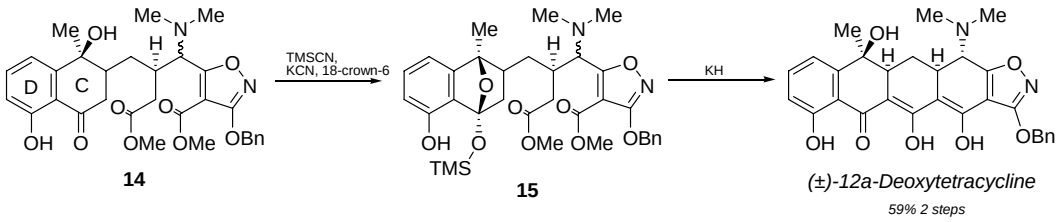
RACEMIC SYNTHESES OF TETRACYCLINE DERIVATIVES

The first synthesis of a tetracycline antibiotic was reported by Woodward and coworkers in 1968 with the synthesis of (±)-6-demethyl-6-deoxytetracycline.[11] Although their route suffered from low yields (25 steps, ~0.002% overall), this synthesis is of historical significance because it was the first preparation of a member of the tetracycline family. The Muxfeldt group[12] achieved the total synthesis of **(±)-**terramycin, in 1979, utilizing a different approach that started from a CD ring precursor and built out to the A ring. The key step involved generation of thiazolone intermediate **11**, which was then condensed with glutamate derivative **12**. This low-yielding but elegant condensation created 3 new C-C bonds in one step (Scheme 3), with most of the positions having the appropriate substitution.[13] In 1996, a landmark synthesis carried out by Stork and coworkers[14] produced (±)-12a-deoxytetracycline (17 steps, 22% overall yield). The key step in this synthesis involved a double Claisen cyclization to generate the A and B rings of the tetracycline nucleus (Scheme 4). This involves generating ketal **15** from the C ring hydroxyl group in **14**, followed by deprotonation at the α position. Hydroxylation at the C-12a position could not be achieved. Stork did develop a method to protect the vinylogous carbamide functionality of the tetracyclines as an isoxazole, a strategy that would be utilized again.[15]

Scheme 3. Key Step in Muxfeldt's synthesis of (±)-Terramycin



Scheme 4. Key Step in Stork's Synthesis of (±)-12a-Deoxytetracycline

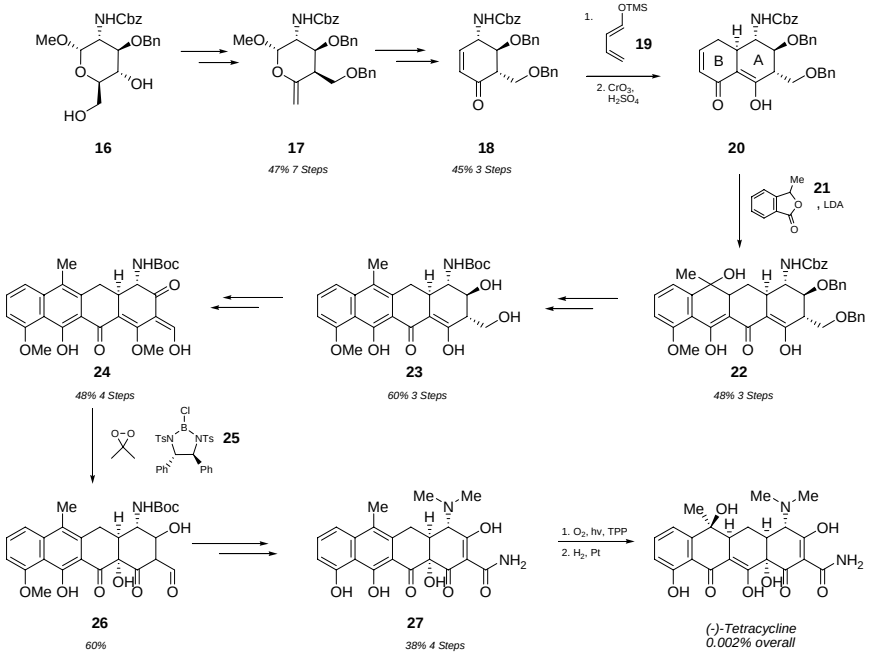


ASYMMETRIC TOTAL SYNTHESES OF TETRACYLINE ANTIBIOTICS

(-)-Tetracycline, Tatsuta 2000

Despite significant synthetic efforts, an asymmetric total synthesis of a tetracycline was not achieved until 2000 by Tatsuta and coworkers (Scheme 5).[16] They took a novel approach: rather than beginning with a D- or DC-ring precursor, Tatsuta and coworkers started from an A-ring precursor. Thus, a D-glucosamine derivative **16**[17] was converted to **17** in 47% yield over 7 steps. This exocyclic enol ether (**17)** was converted to thermodynamically stable cyclohexanone **18** in 45% yield over 3 steps through a Ferrier rearrangement. The electron poor alkene within **18** underwent a regio-selective Diels-Alder cycloaddition with diene **19 (**in the presence of a radical inhibitor) to construct the AB ring system. The unstable cycloadduct was converted to α,β -unsaturated enone **20** via acidic oxidation. This intermediate was treated with isobenzofuran derivative **21** to generate the tetracycline skeleton **22** in 48% yield over 3 steps via a tandem Michael-Dieckmann reaction.[16,18] The enol **22** was converted to **24** by protecting group manipulations and oxidations. Epoxidation of **24** by dimethyldioxirane and a chiral cyclic borane **25**[19] selectively afforded the aldehyde **26** as a single product. Using a protocol established by Scott and coworkers,[19] Tatsuta was able to access (-)-tetracycline using a photoxidation of **27** followed by hydrogenation. This constitutes the first total synthesis of (-)-tetracycline in 34 steps from D-glucosamine, with an overall yield of 0.002%. Notably, this strategy addressed the stereochemistry at C-4.

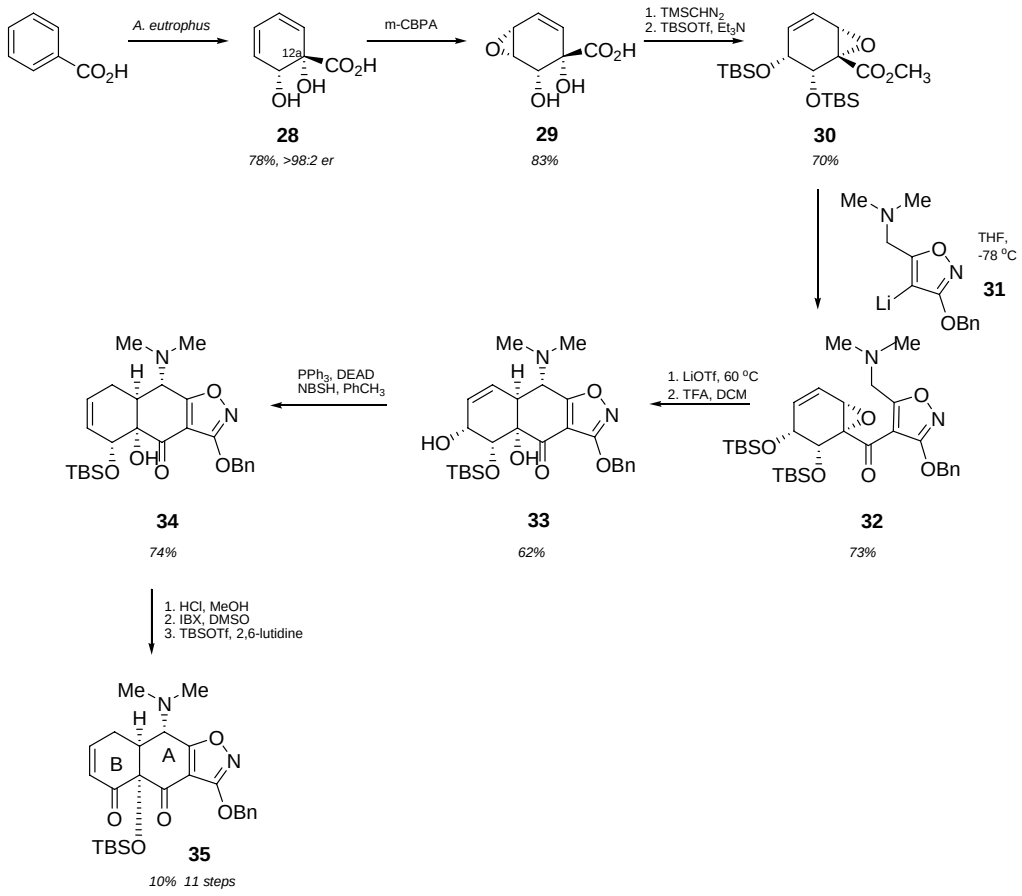
Scheme 5. Tatsuta's synthesis of (-)-Tetracycline



6-Deoxytetracycline and Synthetic Derivatives, Myers 2005

Myers and coworkers[21] recently reported the synthesis of 6-deoxytetracycline and structurally diverse analogs. 6-Deoxytetracycline was chosen because it has been shown to exhibit the same, if not greater, potentency in various strains of bacteria than tetracycline itself.[21,22] Furthermore, the compound is more resistant towards degradation. Historically, methods to introduce the C-12a hydroxyl group have been very inefficient, as shown by Tatsuta and coworkers.[16] Thus, the Myers group chose to install the correct C-12a stereochemistry at an early stage by building up the AB ring system. With this system intact, the D-ring was then coupled to the AB-ring system, stereospecifically generating the C-ring in the process. This approach allows for rapid access to analogs.

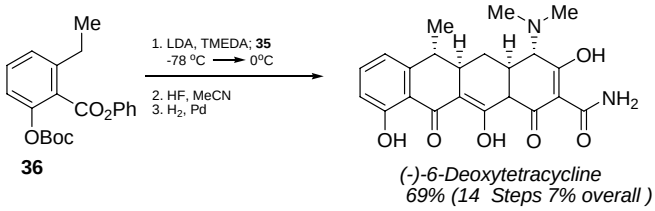
Scheme 6. Synthesis of AB Ring Precursor



Starting with benzoic acid (Scheme 6), whole cell microbial dihydroxylation was carried out to give the diol **28** in >98:2 er. The diol was subjected to hydroxyl-directed epoxidation by *m*-chloroperbenzoic acid (*m*-CPBA), followed by esterifcation by trimethylsilyldiazomethane. The

resulting epoxide was isomerized and bis-silyl protected by *tert*-butyldimethylsilyltriflate (TBSOTf) to give ester **30** in 45% yield over 3 steps.[23] Using a strategy developed by Stork and coworkers,[14,15] the vinylogous carbamic acid moiety within the tetracyclines was masked as a 5-benzyloxyisoxazole. The organolitihium reagent **31**, prepared in 4 steps,[24,25] was added to ester **30** to provide ketone **32**. Construction of the AB ring system was accomplished through the treatment of this ketone with lithium triflate at 60 ^{o}C and subsequent TBS deprotection to give the tricycle **33** with the required cis-fused ring geometry in 62% yield. Double bond migration and concurrent reduction were carried out with triphenylphosphine, diethyl azodicarboxylate, and *o*-nitrobenzylsulfonylhydrazine in 74% yield, giving allylic silyl ether **34**. Deprotection of the TBS group followed by oxidation with IBX and protection of the tertiary alcohol with TBSOTf afforded the α,β-unsaturated enone **35** in 66% yield. Following this efficient AB ring construction, the only remaining hurdle was to couple the AB- and D-rings. The coupling strategy, outlined in Scheme 7, called for the tandem Michael-Diekmann reaction[18] that was previously used by Tatsuta[16] in the total synthesis of (-)-tetracycline. Consequently, the anion of the substituted benzene A-ring unit **36** was generated *in situ* at -78 ^{o}C, followed by addition of CD-ring unit **35** to provide (-)-6-deoxytetracycline in 69%. This impressive reaction produces only one of the four possible diastereomeric products.

Scheme 7. Assembly of AB and D rings yielding (-)-6-Deoxytetracycline



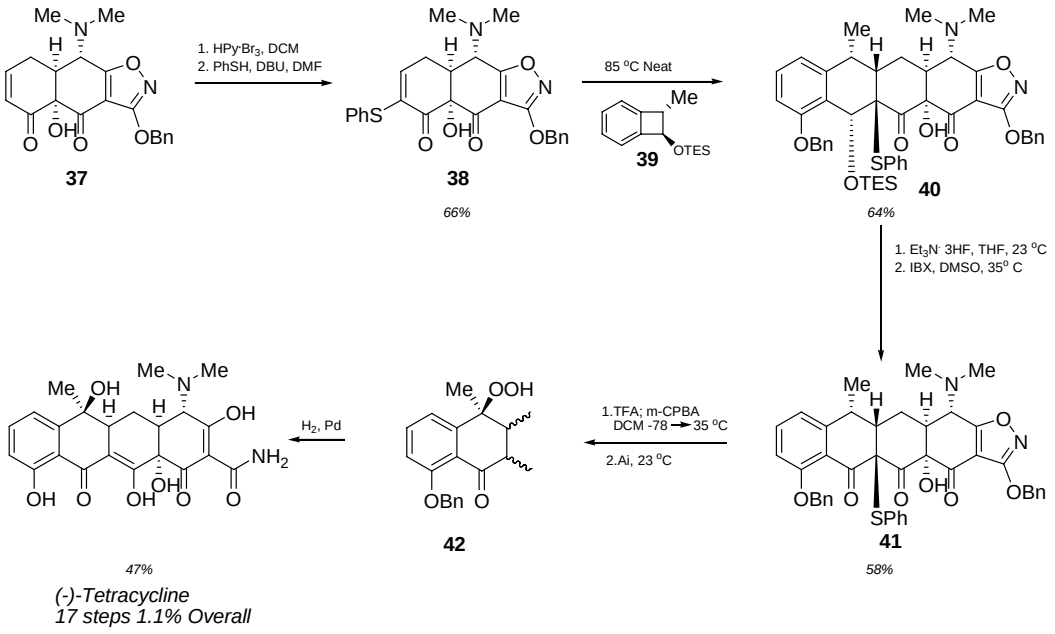
Myers and coworkers went on to make several derivatives of (-)-6-deoxytetracycline. Unfortunately, the coupling reaction had to be optimized for each case, a minor drawback to this otherwise efficient, convergent strategy. Compared to previous attempts to prepare members of this family, Myers' strategy towards 6-deoxytetraycline is revolutionary, and with this convergent approach, derivatives that might be more potent can now be readily accessed.

(-)-Tetracycline, Myers 2005

After the development of their convergent approach toward the tetracycline family, Myers and coworkers decided to apply a similar strategy towards (-)-tetracycline[26] **(**Scheme 8**)**. Starting from the

same AB-ring precursor **37**, a phenylthio substituent was added to the α position of the enone. A neat solution of enone **38** was then heated in the presence of benzocyclobutene **39** to effect an endo-selective Diels-Alder cycloaddition, giving the tetracycline nucleus **40**.[27] Subsequent deprotection of the silyl ether in **40** and oxidation furnished **41**. This triketone was oxidized by *m*-CPBA in the presence of trifluoroacetic acid, to give an intermediate sulfoxide, which then eliminated to give the protected anhydrotetracycline. Isolation of the resulting product proved difficult, however, because it instantly became oxidized in the presence of air to give the peroxide stereoselectively. In practice, the product of the *m*-CPBA oxidation was simply dissolved in chloroform and stirred open to air, and the crude peroxide **42** that formed was then hydrogenated to give (-)-tetracycline in 42% from **41**.

Scheme 8. Myers' Synthesis of (-)-Tetracycline



The beauty of Myers' synthesis, when compared to Tatsuta's, is the remarkably quick access it provides to natural tetracycline from simple starting materials.

CONCLUSION

The tetracyclines represent an important family of antibiotics, and they pose very difficult synthetic challenges. Even though bacteria that are resistant to tetracyclines are appearing, the low mammalian toxicity of the members of this class still provides an impetus to continue the search for synthetically viable derivatives. (-)-Tetracycline has been accessed in a long, linear sequence that used a tandem

Michael-Dieckmann reaction to generate the complete tetracycline skeleton from an AB-ring precursor. A more recent, alternative strategy was based on a highly convergent approach that involved coupling the AB-ring with a D-ring precursor, in the process generating the C-ring stereoselectively. Rapid access to these synthetic derivatives using the latter strategy now provides a robust and flexible means for preparing new antibiotics.

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