

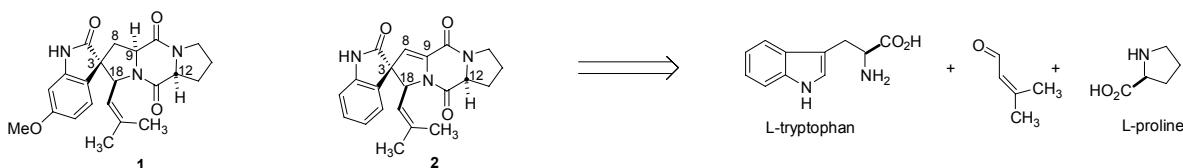
# THE TOTAL SYNTHESIS OF (-)-SPIROTRYPROSTATIN B

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## Introduction

The ability to inhibit the uncontrolled proliferation of cancer cells is a major focus in the discovery of chemotherapeutic agents that regulate Eukaryotic cell cycle progression.<sup>1</sup> Secondary metabolites of microbial origin have recently been found to be a promising source of novel cell cycle inhibitors. Screening of the fermentation broth of the fungus *Aspergillus fumigatus*, led to the discovery of a family of indole alkaloids with anti-mitotic activity.<sup>2-4</sup> Two members of this family, spirotryprostatins A and B (**1** and **2**), contain a unique spirooxindole ring system and inhibit the cell cycle in the G2/M phase with IC<sub>50</sub>'s of 197.5 and 14.0 μM respectively (Figure 1).<sup>4</sup>



**Figure 1.** Spirotryprostatin A and B.

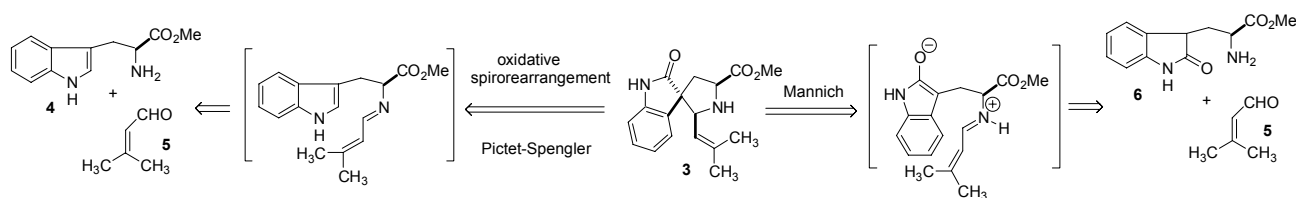
These natural products appear to be biosynthesized from senecialdehyde and a diketopiperazine ring derived from the amino acids L-tryptophan and L-proline. The novel structure and shortage of material for further pharmacological studies provide motivation for total synthesis of these novel compounds. Structurally, the most challenging features of **1** and **2** are the C3 spirooxindole quaternary center, the adjacent isobutenyl group at C18, and the conjugated C8-C9 double bond. This report focuses on several innovative strategies devised to construct stereoselectively the quaternary center at C3 and the adjacent center at C18.

## Spirooxindole Formation Through Imine Intermediates

Two groups have addressed the construction of the spirooxindole core through cyclization of imines generated from senecialdehyde **5** (Scheme 1). Ganesan and coworkers developed a biomimetic approach using a Pictet-Spengler condensation to form a tetrahydro-β-carboline, which could then undergo an oxidative spirorearrangement furnishing spirooxindole **3**.<sup>5</sup> In the second approach, Danishefsky and coworkers saw that a Mannich reaction between oxindole derivative **6** and senecialdehyde **5** would provide spirooxindole **3** in one step.<sup>6</sup>

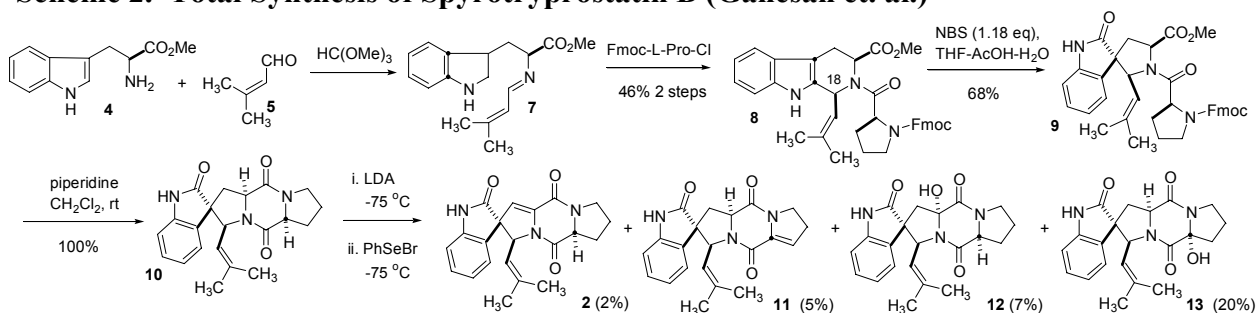
Condensation of aldehyde **5** and L-tryptophan methyl ester **4** with trimethyl orthoformate followed by acylation with Fmoc-L-Pro-Cl, gave tetrahydro-β-carboline **8** in 46% yield as a 1.4:1

### Scheme 1. Retrosynthetic analysis of the imine cyclization strategies.



mixture of C18 cis and trans diastereomers (Scheme 2). While condensations between aliphatic or aromatic aldehydes and tryptophan derivatives proceed smoothly to tetrahydro- $\beta$ -carboline with acid catalysis,  $\alpha,\beta$  unsaturated aldehydes were previously found to be unreactive.<sup>7</sup> However, Ganesan observed that acylation of the imine with proline generates a more reactive imine when compared to its protonated or tosylated analogs, and allows for cyclization to proceed faster than side reactions.<sup>8</sup>

### Scheme 2. Total Synthesis of Spirotryprostatin B (Ganesan et. al.)<sup>5</sup>

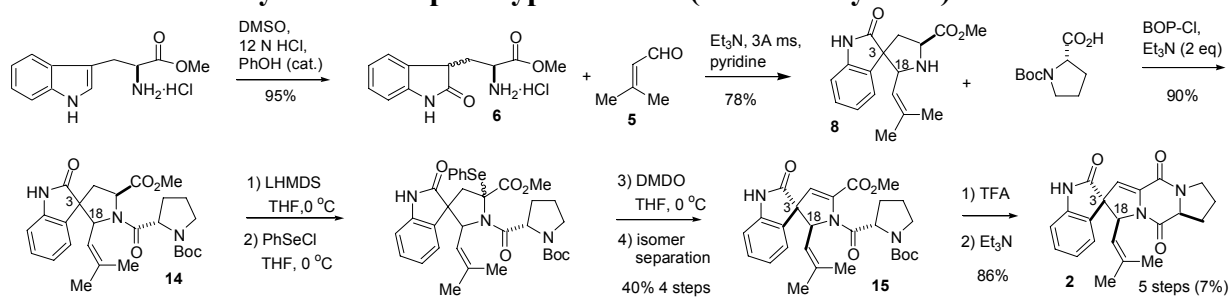


NBS oxidation in aqueous acetic acid<sup>9</sup> of the C18-C9 cis diastereomer furnished oxindole **9** as the major product. The spiro center configuration was created through NBS attack on the less hindered face of **8**, followed by a pinacol-like rearrangement. Fmoc deprotection and simultaneous diketopiperazine cyclization afforded dihydrospirotryprostatin B **10** in quantitative yield. Lithiation of **10** with excess LDA followed by selenylation with PhSeBr and acidic work-up resulted in an unexpected mixture of compounds **2** and **11-13**. While selenide eliminations under nonoxidative conditions with excess phenylselenenyl halide are known,<sup>10</sup> Ganesan rationalized these results by selenylation at both  $\alpha$ -positions of the diketopiperazine ring followed by iminium ion formation through elimination of the selenide. The iminium ion was then either deprotonated to form dehydro-diketopiperazines **2** and **11**, or captured by water to form alcohols **12** and **13**.

Danishefsky and coworkers used a Mannich reaction between oxindole **6** and aldehyde **5** to generate spirooxindole **8** as a mixture of C3 and C18 epimers (Scheme 3).<sup>6</sup> Although partial chromatographic separation could be achieved, the acylation conditions in the subsequent step epimerized the individual diastereomers to produce a mixture of C3 and C18 isomers. This racemization is due to the long reaction times required for the bulky acylating agent and allows for competition from retro Mannich and subsequent Mannich reactions.<sup>11</sup> Accordingly, the mixture of all four epimers of **8**

was acylated to furnish **14** in 90% yield. Introduction of the double bond at C8–C9 through selenylation and oxidative elimination generated a mixture of isomers. After separation of compound **15**, deprotection and Et<sub>3</sub>N-induced cyclization afforded **2** in 7% yield over five steps.

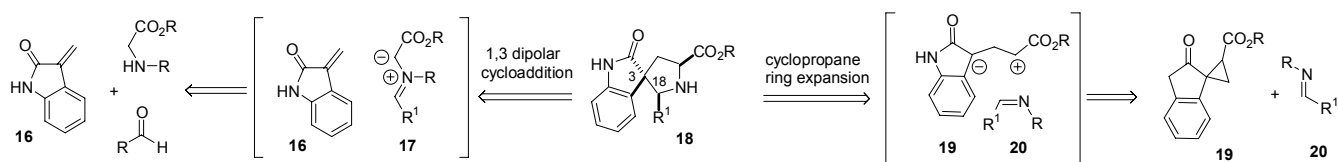
### Scheme 3. Total Synthesis of Spirotryprostatin B (Danishefsky et al.). 6



### Spirooxindole Ring Formation Through Annulation Reactions

Williams and Carreira have disclosed bond-constructing strategies toward the spirooxindole core involving annulation reactions (Scheme 4). Williams proposed a retrosynthetic analysis utilizing an asymmetric 1,3 dipolar cycloaddition with an azomethine ylide **16** and dipolarophile **17** to set simultaneously the correct absolute configuration of the spiro center at C3 and adjacent stereocenter at C18 in spirooxindole **18**.<sup>12</sup> In Carreira's retrosynthetic analysis, the cyclopropane ring  $\alpha$  to a carbonyl group acts as both an electrophile and an enolate in ring forming reactions with oxindole **19** and aldimine **20** to form spirooxindole **18**.<sup>13</sup>

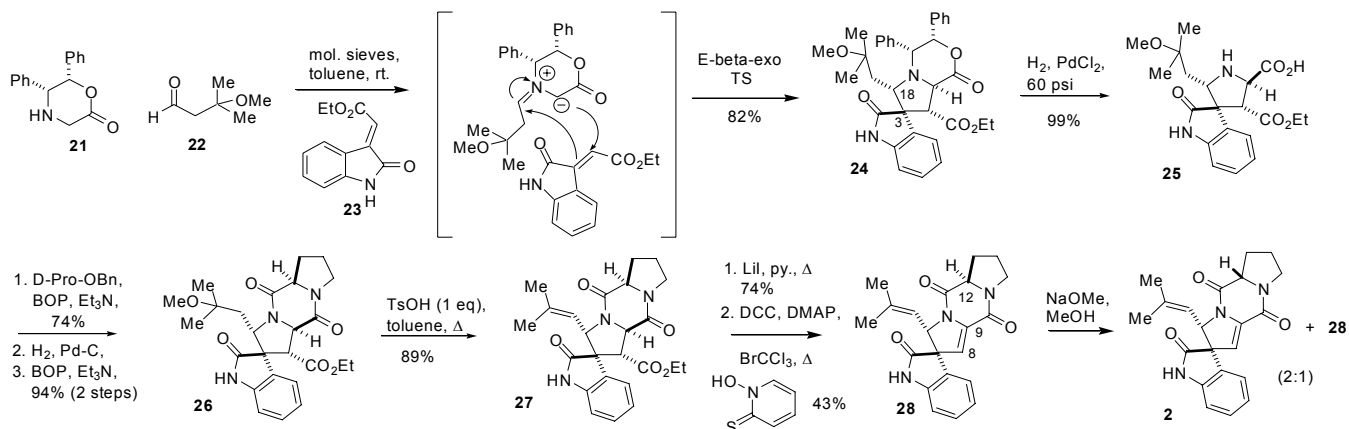
### Scheme 4. Retrosynthetic analysis of the ring annulation strategies.



In the Williams synthesis, the asymmetric [1,3] dipolar cycloaddition was achieved with a chiral azomethine ylide. Condensation of isovaleraldehyde with the commercially available, enantiomerically pure 5,6-diphenylmorpholin-2-one **21** generated the *E* azomethine ylides, and provided strong facial bias for approach of the dipolarophile opposite to the phenyl groups.<sup>14</sup> As the *E* geometry is required to achieve the correct stereochemistry C18, this azomethine ylide presented an ideal dipole substrate for creation of the spiropyrrolidine core. Accordingly, combination of oxazinone **21** with aldehyde **22** and dipolarophile **23** in toluene at room temperature in the presence of 3 Å mol. sieves afforded cycloadduct **24** as a single product in 82% yield (Scheme 5). The X-ray structure of **24** established that the desired absolute configuration of the spiro core and adjacent stereocenter had been achieved. This absolute stereochemistry can be explained through an “*E*-beta-*exo*” transition state, where “*E*” is the geometry of

the imine, “beta” refers to the approach of the dipolarophile from the top face as drawn, and “exo” is the orientation of the ester group on dipolarophile **23**.

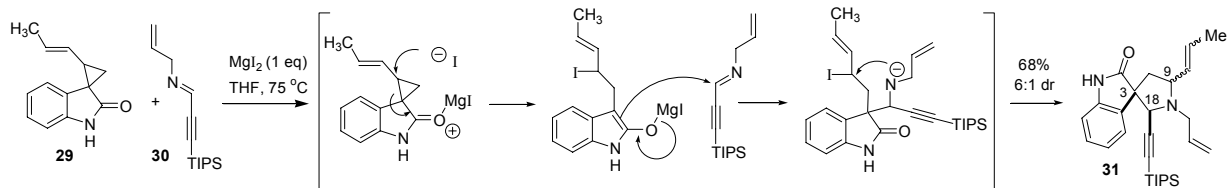
### Scheme 5. Total Synthesis of Spirotryprostatin B (Williams et al.). 12



Catalytic hydrogenation of **24** afforded amino acid **25** in almost quantitative yield. Peptide coupling with *D*-proline benzyl ester using BOP and Et<sub>3</sub>N generated a dipeptide. Removal of the benzyl group and formation of the *cis* diketopiperazine ring through BOP-promoted cyclization furnished **26** in excellent yield over 2 steps. Although *D*-proline has the incorrect configuration and requires a late stage epimerization, coupling with the *L*-proline benzyl ester proceeded in low yields. Reaction of **26** with TsOH in refluxing toluene, afforded **27** as a single double bond isomer. Formation of the carboxylic acid with LiI in refluxing pyridine, followed by Barton oxidative decarboxylation<sup>15</sup> generated 12-*epi*-spirotryprostatin B, **28**. Epimerization, by addition of NaOMe in MeOH, produced a 2:1 mixture of **2**:**28** which was separated by chromatography to give pure (-)-spirotryprostatin in 11% overall yield.

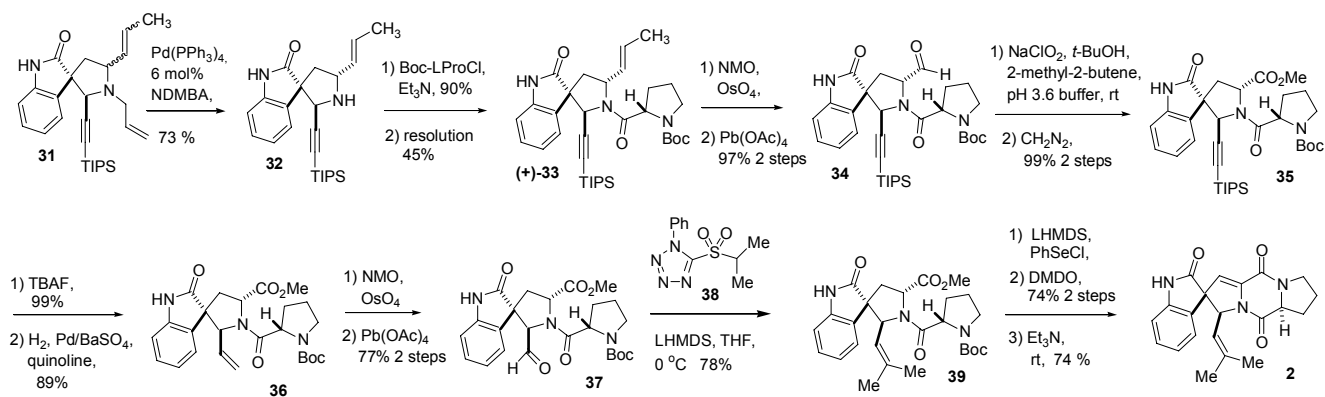
Carreira and coworkers approach to the diastereoselective construction of the spirooxindole core was achieved through a MgI<sub>2</sub>-promoted annulation of the 1,2 disubstituted cyclopropane **29** with aldimine **30** (Scheme 6).<sup>13</sup> Carreira proposed that the facile dissociation of MgI<sub>2</sub> generates a Lewis acid and base that work in synergy to open the cyclopropane.<sup>16</sup> Nucleophilic attack by I<sup>-</sup> onto the activated cyclopropane generates a magnesium enolate that reacts with the aldimine. The allylic iodide, created by the ring opening, can then be displaced by the anionic nitrogen to form the spirooxindole. Optimal reaction conditions were found to be the combination of spirocyclopropane **29** with imine **30** in THF at 75 °C in a sealed tube in the presence of 1 equiv of MgI<sub>2</sub> for 15 h. Spirooxindole **31** was obtained in a 68% yield as a 6:1 mixture of diastereomers, favoring the desired diastereomer.

### Scheme 6. Mechanism of the cyclopropane ring annulation reaction.



After the annulation, the C9 epimers were separated<sup>17</sup> and the allyl protecting group of the 9R isomer of **31** was removed to afford **32** (Scheme 7). Coupling with N-Boc-proline chloride gave racemic **33** and allowed for the separation of the enantiomers. Oxidative cleavage of the propenyl double bond generated aldehyde **34** in excellent yield over two steps. The aldehyde was then oxidized to the acid that was methylated to afford ester **35**. Removal of the triisopropylsilyl (TIPS) group produced a terminal alkyne, which was converted to alkene **36** through hydrogenation and oxidative cleavage generated aldehyde **37** in good yield. While previous investigators had found an aldehyde at C18 to be resistant to a variety of olefination procedures,<sup>18</sup> Carreria and coworkers discovered alkene **39** could be obtained in 78% yield without epimerization at C18 using the Kociensky modified Julia olefination with tetrazole **38**. The double bond at C8-C9 was then installed through selenylation followed by oxidative elimination. Removal of the Boc protecting group from proline allowed for diketopiperazine ring formation and purification provided (-)-spirotryprostatin B in 0.03% overall yield.

### Scheme 7. Total Synthesis Spirotryprostatin B (Carrera et al.).<sup>13</sup>

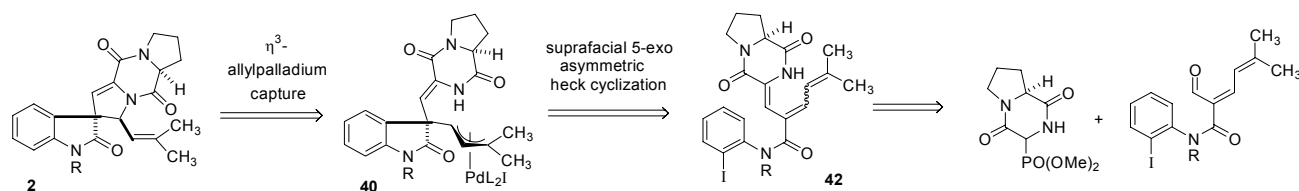


### Spirooxindole Ring Formation Through an Asymmetric Heck Reaction

In a distinctly different approach, Overman proposed coupling an intramolecular asymmetric Heck reaction with the bimolecular trapping of an  $\eta^3$ -allylpalladium intermediate by the proximal nitrogen of the diketopiperazine ring to set simultaneously the stereochemistry at C3 and C18 (Scheme 8).<sup>19</sup> The initial asymmetric Heck reaction with control of the appropriate chiral ligand would proceed to give 5-exo cyclization with syn insertion of the aryl palladium bond.<sup>20,21</sup> This would generate a geometrically defined, configurationally stable  $\eta^3$ -allylpalladium species **40** or **41**, were the geometry of the  $\eta^3$ -allylpalladium species is dictated by the internal double bond geometry of the conjugated triene, (*E*) or (*Z*)-**42**. Facial isomerization of the  $\pi$ -allyl palladium intermediate would be unfavorable due to the highly substituted termini. Trapping by nitrogen could then proceed either anti or syn to the metal

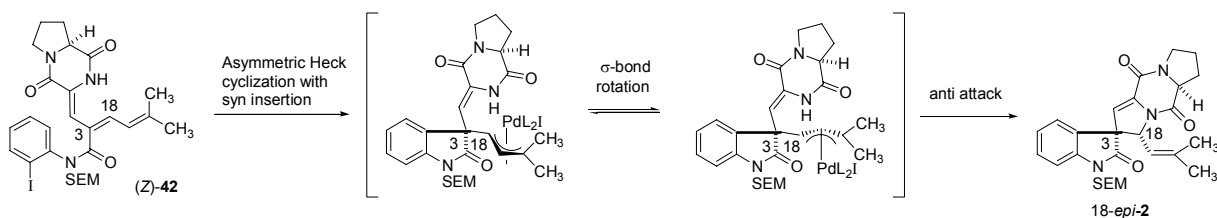
center to generate **2** or 18-*epi-2*. As the stereochemical outcome of attack by nitrogen was unknown, this strategy was equipped to deal with either result by simply changing the geometry of triene **42**.

**Scheme 8. Retrosynthetic analysis of the Heck cyclization and allylpalladium capture strategy.**



Without prior knowledge of the mode of nitrogen attack, triene (*Z*)-**42** was arbitrarily synthesized first. Cyclization in the presence of 20% Pd/(*S*)-BINAP and excess 1,2,2,6,6-pentamethylpiperidine (PMP) in DMA at 100°C produced a 6:1 mixture of 18-*epi-2* and 3-*epi-2* in 28% yield. This stereochemical outcome implies nitrogen attack anti to the metal center prior to stereomutation of the  $\eta^3$ -allylpalladium intermediate (scheme 9). It was then anticipated that cyclization of triene (*E,E*)-**42** under the same reaction conditions would produce spirotryprostatin B, **2**.

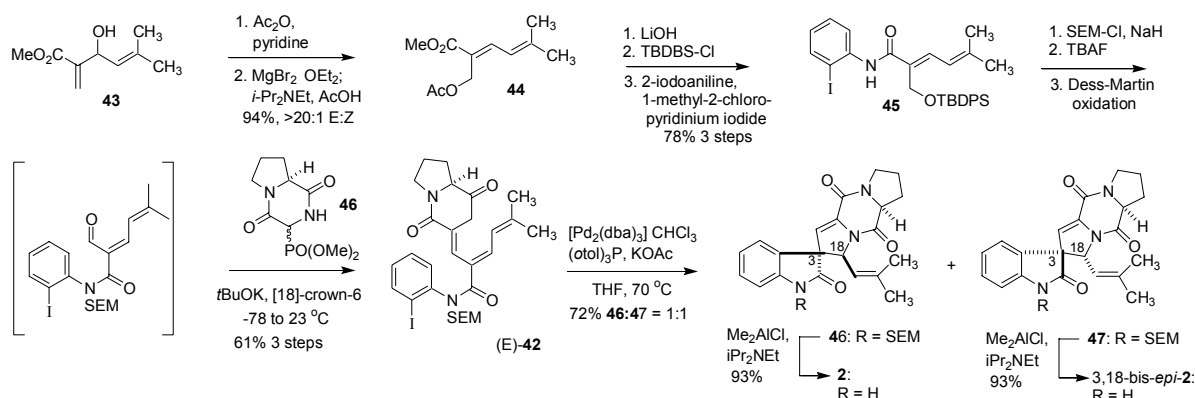
**Scheme 9. Direction of nitrogen attack.**



The synthesis of (*E*)-**42** began with the acylation of known alcohol **43**, which was synthesized from methyl acrylate and 3-methyl-2-butenal (Scheme 10). The acylated alcohol was then transformed into the primary allylic bromide, which was subsequently displaced by acetate to generate (*E*)-dienoate **44**. Saponification with LiOH and silylation with *t*-butyldiphenylsilyl chloride (TBDPS-Cl) generated a silylcarboxylic acid, which after coupling with 2-iodoaniline afforded **45**. The amide nitrogen was then protected with (2-trimethylsilyl)ethoxymethyl chloride (SEM-Cl) before cleavage of the TBDPS group with *n*-Bu<sub>4</sub>NF. The primary alcohol was then oxidized and coupled with diketopiperazine phosphonate **46** to generate (*E*)-**42** in isomerically pure form. Surprisingly, cyclization of (*E*)-**42** using Pd/(*S*)-BINAP under identical conditions as shown above led to the formation of 18-*epi-2*. When the same reaction was performed without palladium, *E* to *Z* isomerization of the internal bond of triene was observed. Attempts to find reaction conditions that would promote the asymmetric Heck reaction without isomerization were unsuccessful, and necessitated the use of milder achiral conditions. Cyclization of (*E*)-**42** with 10% [Pd<sub>2</sub>(dba)<sub>3</sub>]·CHCl<sub>3</sub>, 40% tri-*o*-tolylphosphane ((*o*-tol)<sub>3</sub>P), and excess KOAc in THF at 70°C afforded a 1:1 mixture of **2** and 3,18-*bis-epi-2* in 72% yield, again demonstrating nitrogen attack

anti to the metal center. Deprotection and chromatographic separation provided pure (-)-**2** in 9% overall yield.

### Scheme 10. Total Synthesis of Spirotryprostatin B (Overman et. al.).<sup>19</sup>



## CONCLUSION

Several innovative and conceptually distinct strategies toward the synthesis of the unique spirooxindole core of spirotryprostatin B have been highlighted. These methods include Pictet-Spengler condensation followed by an oxidative rearrangement, a Mannich reaction, an asymmetric 1,3 dipolar cycloaddition, an annulation of a cyclopropane, and an intramolecular Heck reaction. Although high stereocontrol in the formation of the C3 quaternary center was accomplished in several syntheses, these methods were also accompanied by low yielding functional group transformations and epimerizations in later stages. While these methods culminated in total syntheses, a truly general and high yielding strategy is still lacking. The opportunity for finding new reactions and strategies toward the construction of the unique structure of the spirotryprostatins still exists for future investigators.

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