transformations of the products,[4] a wide range of synthetic applications can be readily envisaged. The simplicity of the procedures, the low cost of the reagents, and the good chemical and optical yields of the products, make this a useful alternative to existing methods.[5, 6]

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[13] The corresponding pyroglutamic acids were isolated from the diastereomerically pure nickel(II) complexes (as depicted in Scheme 2) and their spectroscopic and optical properties compared with the literature data (ref. [6]).

[14] Exceptions were cases where α-(*R*) absolute configuration is a consequence of the Cahn-Ingold-Prelog priority (see ref. [16]) and also some aldol addition reactions where the α-(*R*) stereochemistry is a result of the rearrangement of the corresponding aldol products forming the β-hydroxy-coordinated complexes. For general patterns of reactivity and stereocontrol properties of complex (*S*)-**1**, see ref. [15].

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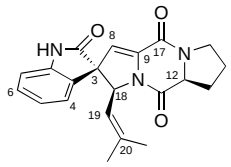
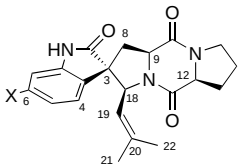
A Rapid Total Synthesis of Spirotryprostatin B: Proof of Its Relative and Absolute Stereochemistry**

Franz von Nussbaum and Samuel J. Danishefsky*

A variety of prenyl-containing alkaloids, clearly derived from the amino acids tryptophan and proline and linked by a diketopiperazine arrangement, have been isolated from natural sources.[1, 2] One such alkaloid which engaged our interest is spirotryprostatin A (**2**),[3] wherein a "prenylidene" group serves to meld N_b of the tryptophan to the β-carbon atom of the oxidized indolo sector. Recently, our laboratory



spirotryprostatin B (**1**)



X = OCH_3: spirotryprostatin A (**2**)
= H: 6-demethoxyspirotryprostatin A (**3a**)

disclosed the total synthesis of spirotryprostatin A (**2**).[4] Coproduced with the A compound in *Aspergillus fumigatus*, is spirotryprostatin B (**1**),[3] which lacks the C6-methoxyl substituent of the A congener **2**. Clearly, the additional site of unsaturation in the B system adds incremental complexity to the prospect of its total synthesis.

The relative stereochemistry of the A compound **2**, assigned on the basis of NMR "through space" connectivities between carbons 3, 18, 9, and 12 was fully corroborated by our total synthesis,[4] which also served to establish the absolute configuration. In the case of spirotryprostatin B (**1**), the "prolyl" center (C12) is spectroscopically isolated from the prenylated tryptophan domain (see Scheme 5). Thus, while the relative configurational arrangements at the C3 and C18 centers of the A and B alkaloids (**2** and **1**) are the same, the relationship between these diads and the proline-derived C12 atom could not be asserted with rigor in the latter compound. As we shall show, the surmize of the original discoverers of spirotryprostatin B (**1**) turns out to be correct.

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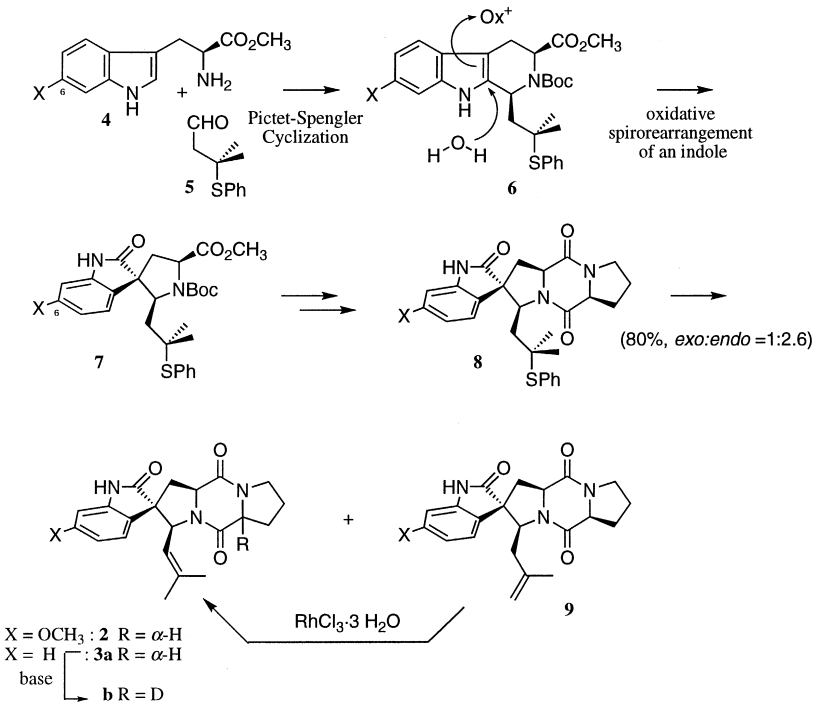
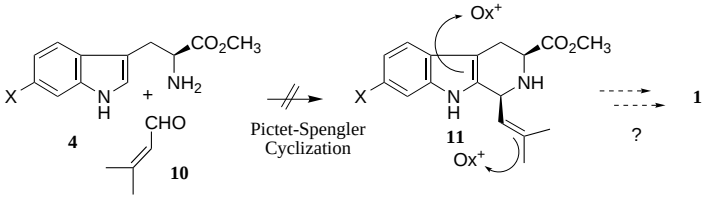
In addition to learning how to overcome the significant challenge posed by the presence of the C8–C9 double bond of the B system, total synthesis[5] emerged as the only way for our laboratory to gain access to appropriate quantities of this compound for assessment of its potential utility as a cancer chemotherapeutic agent (reportedly operating by cell cycle inhibition at the G2/M phase).

We naturally gave thought to reaching our goal by retracing and modifying our route to the A system **2**, which also had led to a total synthesis of 6-demethoxyspirotryprostatin A (**3a**). However, the routes we had used to reach the A compound **3a** (see Scheme 1), while reasonably concise (13 steps), were plagued by several difficult specificity issues necessitating



Scheme 1. Synthesis of spirotryprostatin A (**2**). Ox^+ = oxidant, Boc = *tert*-butoxycarbonyl.

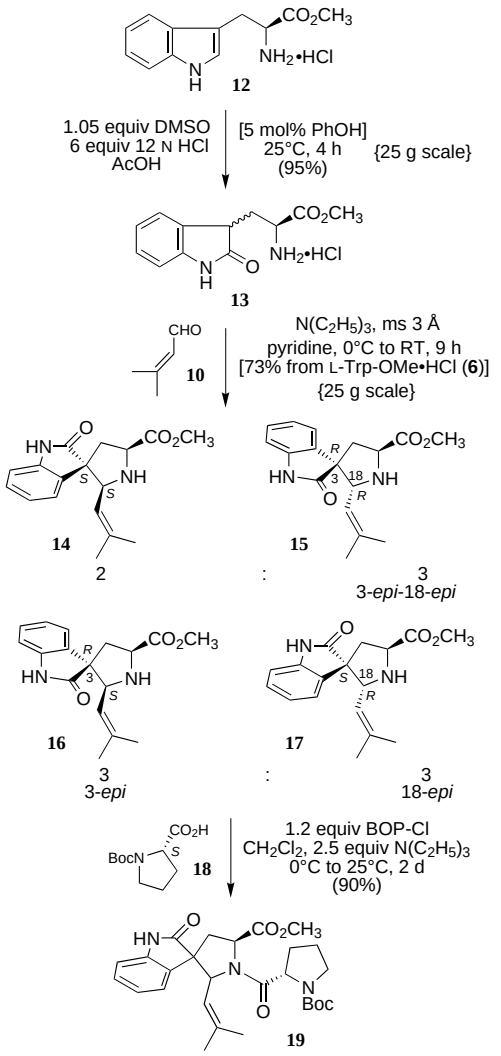
awkward management of functional groups. Moreover, attempted metal exchange at C9 in **3a**, bearing the intact diketopiperazine had resulted in metal hydrogen exchange at C12 (cf. **3b**), thus further complicating prospects for late stage introduction of the 8–9 double bond from a previously synthesized dihydro precursor.[6] Another serious factor complicating our synthesis of **2** was our failure to install the prenyl group by direct Pictet–Spengler incorporation of senecia aldehyde (**10**, Scheme 2). We and others[6] have not been able to reduce this apparently simple concept to practice



Scheme 2. Attempted Pictet–Spengler cyclizations with prenyl aldehyde (**10**).

in the face of the lability of **10**. We started by asking whether a Mannich reaction[7] on an oxindole would enable direct incorporation of the required prenyl function.

The readily available L-tryptophan methyl ester (**12**) was converted (95%) into the oxindole derivative hydrochloride **13** (Scheme 3).[8] Treatment of **13** with **10**, under the conditions shown, afforded a 73% yield of prenyl "insertion" products. Detailed NMR studies revealed that, at this stage, a four component mixture (**14**–**17**) had been generated.



Scheme 3. The Mannich route to the spirooxindole system. DMSO = dimethyl sulfoxide, ms = molecular sieves, RT = room temperature, BOP = Bis-(2-oxo-3-oxazolidinyl)phosphoryl.

Following chromatography, compound **15**, with the 3,18-bis-*epi* configuration (3*R*,18*R*), could be obtained in a homogenous state. The other three isomers coeluted. There was no advantage to be gained from independent forward processing of **15** since, under our acylation conditions with *N*-Boc-L-proline (**18**), extensive equilibration had occurred at C3 and C18. Indeed, attempted acylation of **15** with **18** afforded a product **19** whose NMR characteristics were the same as those of the mixture arising from conducting the prolinoylation on the **14**–**17** mixture. Identification of the components of the acylation mixture **19** (no stereoassignments intended) was no

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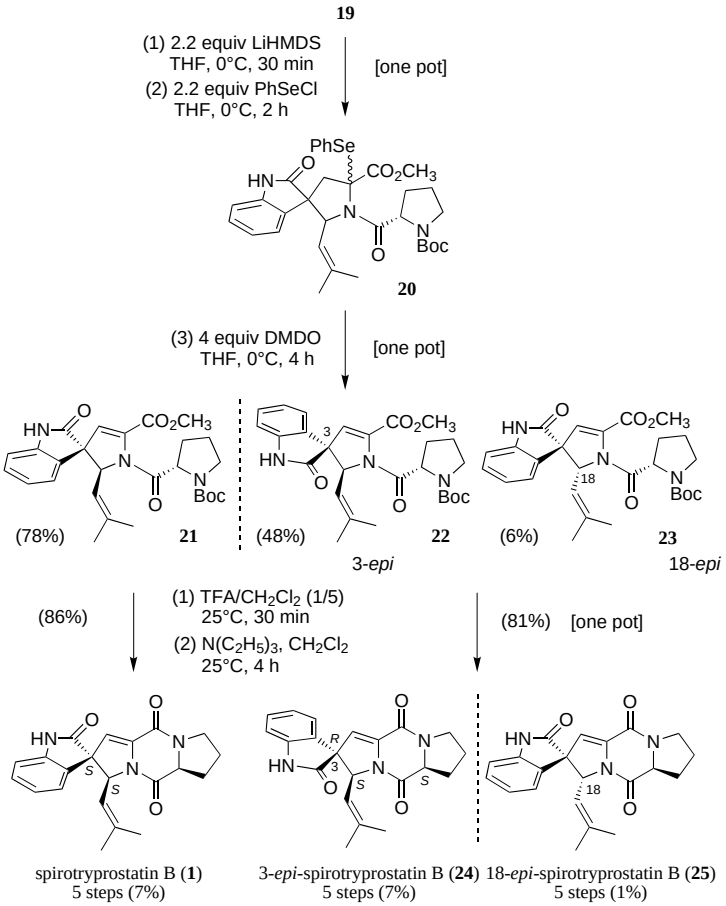
small matter. Given the serious complications in NMR spectral analysis of the prolinoylation products, further aggravated by the presence of the amidic and carbamate rotamers, we relied solely on mass spectroscopy to establish that coupling had actually occurred.

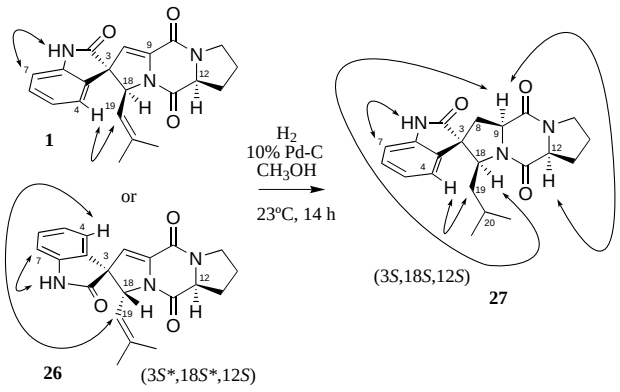
At this stage, we hoped to exploit the fact that in these acylation products, the enolizable tryptophyl α-center (C9) appears in the context of an ester (flanked by an *N*-acyl group). By contrast, the corresponding α-center in the proline sector (C12) is presented in an amide-like setting (this time flanked by a carbamoyl type nitrogen). In the event, treatment of **19**, with lithium 1,1,1,3,3,3-hexamethyldisilazane, followed by selenylation, led to a crude product presumed to be **20** by ESI-MS (Scheme 4). Upon exposure of the selenide **20** to the action of dimethyldioxirane, oxidative elimination occurred readily, yielding a mixture of the products **21**–**23**. Compound **21** could be separated from the latter two (**22** and **23**) by silica gel chromatography. The other fraction eluted as a mixture of **22** and **23**, substantially favoring the former according to NMR analysis. Fortunately, there had been a major simplification in converting the **14**–**17** mixture into the dehydro intermediates **21**–**23**. Thus, the pre-**2** stereoisomer **14** was the least prevalent isomer at the nonacylated stage (18% of a 4-component mixture). In the penultimate dehydro series it is approximately 40% (and readily purified) of a three component mixture, one member of which (**23**) is quite minor.



Scheme 4. Synthesis of spirotryprostatin B (**1**). HMDS = 1,1,1,3,3,3-hexamethyldisilazane, DMDO = dimethyldioxirane.

Deprotection of the prolyl nitrogen of **21** followed by triethylamine induced cyclization, afforded spirotryprostatin B (**1**), identical by the criteria of ^{1}H, COSY, HMBC, and ^{13}C NMR spectroscopy methods with the natural product.[9] Moreover, the CD spectrum and the optical rotation ($[\alpha]_D^{23}$ of synthetic material -165.4; $[\alpha]_D$ reported -162.1) established both materials to have the same absolute stereochemistry.

In order to confirm that spirotryprostatin B is **1**, rather than **26**, the fully synthetic compound was subjected to catalytic hydrogenation (Scheme 5). In this way, the proton at C9 would serve as a linkage site at the level of NMR analysis.



Scheme 5. Introduction of the 9-H NMR spectroscopy linker for nuclear Overhauser experiments.

Indeed, with greater than 90% *de*, catalytic hydrogenation of the fully synthetic compound afforded **27**. This compound, corresponding to tetrahydrospirotryprostatin B, exhibited all of the expected nuclear Overhauser enhancements (Scheme 5) between the α-protons (9-H, 12-H, and 18-H). Compound **27** could also be obtained by hydrogenation of 6-demethoxyspirotryprostatin A (**3a**) synthesized by the old route (Scheme 1). These findings rigorously establish spirotryprostatin B to have the relative and absolute stereochemistry implied in expression **1**.

Processing of the mixture of **22** and **23** in the same fashion, followed by chromatographic separation afforded 3-*epi*-spirotryprostatin B (**24**)[10] and 18-*epi*-spirotryprostatin B (**25**)[11] in a ratio of 7:1. Following careful chromatography, these compounds were obtained as homogenous entities. Missing at the end of the four step processing of mixture **14**–**17** was the 3,18-bis-*epi* compound which would have been derived from **15**. Its nonappearance in the sequence might reflect a failure of its precursor to undergo the oxidative dehydrogenation sequence or could be due to unfavorable stereoequilibration at an undetermined stage.

We note that while our total synthesis, as it is now conducted, lacks stereocontrol at carbons 3 and 18, it does produce copious quantities of **1**. (Currently we deliver approximately 500 mg per batch of this highly active compound, in five easily executed steps, starting with 5.5 g of trivially available **12** and resorting to only

two chromatographic purifications.) We expect to explore fresh possibilities for dealing with the last remaining stereochemical issues. However, chemical synthesis is already the preferred way to amass formidable quantities of **1**, and biological investigations reflecting its now ready availability are beginning in earnest.

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[5] Two other total syntheses of **1** have been accomplished, by groups headed by Prof. Robert Williams at Colorado State University (*J. Am. Chem. Soc.* **2000**, in press) and Prof. Lawrence Overman at the University of California, Irvine (personal communication). The work described herein was disclosed by S.D. at the H. C. Brown Award Symposium at the 219th National Meeting of the American Chemical Society, San Francisco, CA, on March 26, 2000. We thank Professor Williams and Professor Overman for sharing their results with us.

[6] a) D. M. Harrison, R. B. Sharma, *Tetrahedron* **1993**, *49*, 3165 – 3184; b) for a successful *N*-acyliminium Pictet – Spengler condensation, see: H. Wang, A. Ganesan, *Org. Lett.* **1999**, *1*, 1647 – 1649; H. Wang, A. Ganesan, *Tetrahedron Lett.* **1997**, *38*, 4327 – 4328.

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[8] W. E. Savige, A. Fontana, *Int. J. Pept. Protein Res.* **1980**, *15*, 285 – 297.

[9] Spirotryprostatin B (**1**): $[\alpha]_D^{23} = -165.4$ ($c = 0.92$, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 1.27$ (s, 3H, 22-H), 1.56 (s, 3H, 21-H), 1.92 – 2.03 (m, 2H, 13β-H, 14α-H), 2.01 – 2.15 (m, 1H, 14β-H), 2.48 (m, 1H, 13α-H), 3.57 (ddd, $J = 12.3$, 9.3, 3.0 Hz, 1H, 15α-H), 3.84 (dt, $J = 12.3$, 8.1 Hz, 1H, 15β-H), 4.34 (dd, $J = 10.0$, 5.8 Hz, 1H, 12-H), 5.21 (dm, $J = 8.9$ Hz, 1H, 19-H), 5.42 (d, $J = 8.9$ Hz, 1H, 18-H), 5.77 (s, 1H, 8-H), 6.85 (d, $J = 7.8$ Hz, 1H, 7-H), 6.99 (t, $J = 7.6$ Hz, 1H, 5-H), 7.06 (d, $J = 7.4$ Hz, 1H, 4-H), 7.23 (t, $J = 7.8$ Hz, 1H, 6-H), 7.69 (s, 1H, 1-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): $\delta = 18.22$ (C-22), 22.07 (C-14), 25.27 (C-21), 29.23 (C-13), 44.82 (C-15), 61.55 (C-12), 61.86 (C-3), 64.11 (C-18), 110.05 (C-7), 116.47 (C-8), 120.43 (C-19), 122.18 (C-5), 127.21 (C-3a), 127.71 (C-4), 129.04 (C-6), 138.16 (C-9), 138.28 (C-20), 140.70 (C-7a), 155.07 (C-17), 162.56 (C-11), 178.52 (C-2); HRMS (FAB): calcd for $C_{21}H_{22}N_3O_3$ $[M+H]^+$: 364.1661, found: 364.1650.

[10] 3-*epi*-Spirotryprostatin B (**24**): $[\alpha]_D^{23} = -250.6$ ($c = 0.87$, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 1.35$ (s, 3H, 22-H), 1.69 (s, 3H, 21-H), 1.93 – 2.03 (m, 2H, 13β-H, 14α-H), 2.11 (m, 1H, 14β-H), 2.41 (m, 1H, 13α-H), 3.53 (ddd, $J = 12.7$, 9.7, 3.6 Hz, 1H, 15α-H), 3.84 (dt, $J = 12.6$, 7.8 Hz, 1H, 15β-H), 4.23 (dd, $J = 10.4$, 5.7 Hz, 1H, 12-H), 5.28 (d, $J = 9.3$ Hz, 1H, 18-H), 5.56 (dm, $J = 9.2$ Hz, 1H, 19-H), 5.86 (s, 1H, 8-H), 6.82 (d, $J = 7.7$ Hz, 1H, 7-H), 7.07 (t, $J = 7.6$ Hz, 1H, 5-H), 7.20 (d, $J = 7.3$ Hz, 1H, 4-H), 7.25 (t, $J = 7.3$ Hz, 1H, 6-H), 7.40 (s, 1H, 1-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): $\delta = 18.19$ (C-22), 22.00 (C-14), 25.83 (C-21), 29.54 (C-13), 44.71 (C-15), 61.63 (C-12), 62.30 (C-3), 66.59 (C-18), 109.81 (C-7), 115.84 (C-8), 119.78 (C-19), 123.36 (C-5), 124.07 (C-4), 129.44 (C-6), 131.32 (C-3a), 136.84 (C-20), 138.36 (C-9), 140.15 (C-7a), 154.65 (C-17), 163.38 (C-11), 175.56 (C-2); HRMS (FAB): calcd for $C_{21}H_{22}N_3O_3$ $[M+H]^+$: 364.1661, found: 364.1675.

[11] 18-*epi*-Spirotryprostatin B (**25**): $[\alpha]_D^{23} = -24$ ($c = 0.14$, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 1.23$ (s, 3H, 22-H), 1.66 (s, 3H, 21-H), 1.96 – 2.05 (m, 2H, 13β-H, 14α-H), 2.13 (m, 1H, 14β-H), 2.45 (m, 1H, 13α-H), 3.55 (ddd, $J = 12.4$, 9.3, 3.3 Hz, 1H, 15α-H), 3.84 (dt, $J = 12.7$, 8.5 Hz, 1H, 15β-H), 4.38 (dd, $J = 10.0$, 5.8 Hz, 1H, 12-H), 5.27 (d, $J = 9.3$ Hz, 1H, 18-H), 5.49 (dm, $J = 9.3$ Hz, 1H, 19-H), 5.83 (s, 1H, 8-H), 6.81 (d, $J = 7.7$ Hz, 1H, 7-H), 7.07 (t, $J = 7.6$ Hz, 1H, 5-H), 7.25 (m, 2H, 6-H, 4-H), 7.47 (s, 1H, 1-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): $\delta = 18.11$ (C-22), 22.01 (C-14), 25.87 (C-21), 28.79 (C-13), 45.12 (C-15), 61.66 (C-12), 62.39 (C-3), 66.74 (C-18), 109.69 (C-7), 114.88 (C-8), 118.18 (C-19), 123.38 (C-5), 124.51 (C-4), 129.43 (C-6), 129.04 (C-6), 130.60 (C-3a), 137.92 (C-20), 140.59 (C-7a), 154.61 (C-17), 162.59 (C-11), 175.27 (C-2); HRMS (FAB): calcd for $C_{21}H_{22}N_3O_3$ $[M+H]^+$: 364.1661, found: 364.1650.

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