Natural Products Synthesis

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Total Synthesis of Tryprostatins A and B**

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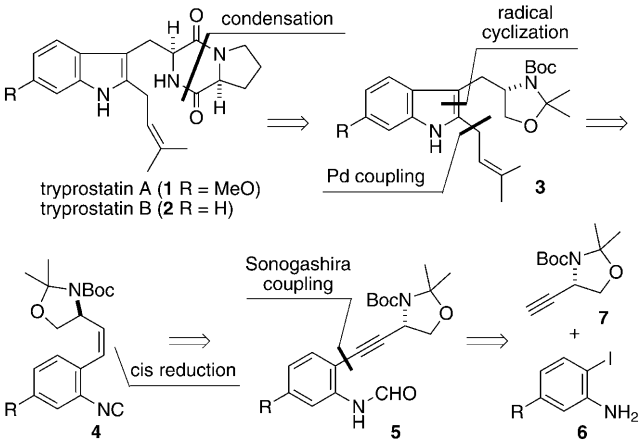
Multiple-drug resistance toward cell-cycle inhibitors frequently becomes an obstacle in cancer chemotherapy. One strategy for circumventing this problem is to develop antimitotic agents that operate by a new mode of action. Two such compounds, tryprostatins A and B, were isolated in 1995 from the fermentation broth of *Aspergillus fumigatus* BM939 by Osada and co-workers.[1] Tryprostatin A holds great promise, because it selectively arrests the cell cycle at the mitotic phase in tsFT210 cells.[2] Interesting biological activity combined with an apparently simple structure has stimulated the synthetic community, and several total syntheses have already been reported.[3,4]

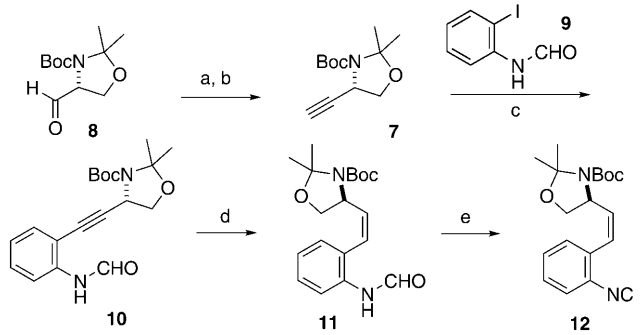
Our research group has long been involved with the radical-mediated construction of a variety of indole cores and the application of these methods to natural product synthesis.[5,6] We envisioned that our method could also be applied to the synthesis of the 2,3-disubstituted indole moiety of the tryprostatins. The interesting structural features as well as the therapeutic potential of tryprostatin A analogues indicated in a recent structure–activity-relationship study[4e] prompted us to launch a program toward the synthesis of tryprostatins.

We reasoned that radical-mediated cyclization and subsequent palladium-mediated coupling with a prenyl-group donor would enable facile construction of the 2,3-disubstituted indole core structure **3** (Scheme 1). The requisite *ortho*-alkenyl isocyanide **4** would be prepared from the alkyne **5**, which in turn could be accessed by Sonogashira coupling of the aromatic iodide **6** with the terminal alkyne **7**. To enable synthesis of the target compounds with high enantiomeric purity, the alkyne **7** derived from the Garner aldehyde (**8**)[7] was employed as a latent amino acid unit to be incorporated into the diketopiperazine.

Initially, we focused our efforts on the synthesis of the isocyanide **12** as a radical-cyclization precursor (Scheme 2). The alkyne **7** was prepared from **8** according to the known protocol[8] with a slight modification. After the Sonogashira coupling between **7** and 2-iodoformanilide (**9**), partial reduction of the triple bond was examined. Whereas the use of the Lindlar catalyst, Pd/C, nickel boride,[9] or diimide[10] resulted in no reaction or overreduction, treatment with $Zn/LiCuBr_2$ in ethanol[11] gave the desired product **11** along with the corresponding amine. The use of 2,2,2-trifluoroethanol[12] as the solvent suppressed the undesired solvolysis and improved the yield of **11** to 99%. Subsequent dehydration with bis(trichloromethyl) carbonate (triphosgene) gave the *ortho*-alkenyl isocyanide **12** and thus set the stage for a radical-mediated cyclization.



Scheme 1. Retrosynthetic analysis. Boc = *tert*-butoxycarbonyl.



Scheme 2. Synthesis of the *ortho*-alkenyl isocyanide **12**: a) CBr_4, PPh_3, Et_3N, CH_2Cl_2, −60–0°C, 88%; b) EtMgBr, THF, 0°C, 98%; c) **9**, CuI, $[PdCl_2(PPh_3)_2]$, Et_3N, room temperature, 97%; d) Zn, $BrCH_2CH_2Br$, CuBr, LiBr, THF, CF_3CH_2OH, 70°C, 99%; e) triphosgene, pyridine, CH_2Cl_2, 0°C, 87%.

When isocyanide **12** thus obtained was subjected to our previously established radical-cyclization conditions,[5a] the desired 5-exo adduct **13** was obtained in moderate yield after acidic treatment with silica gel along with a considerable amount of products **14** and **15** of a 6-*endo* cyclization–cleavage process (Scheme 3). We were aware of the tendency of this imidoyl-radical cyclization to give a mixture of the 5-*exo* and 6-*endo* products if the intermediate radical of the

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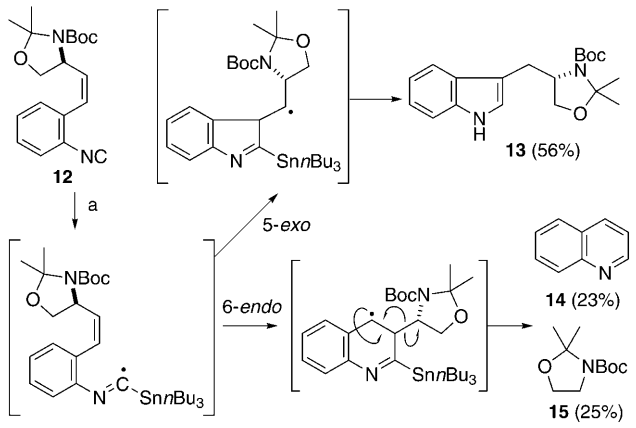
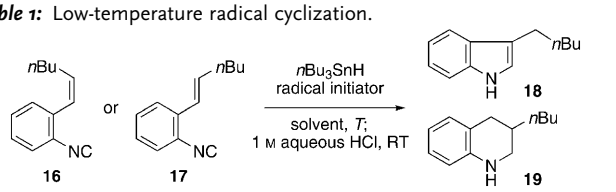


Scheme 3. Troublesome radical-mediated cyclization: a) nBu$_3$SnH (2.0 equiv), AIBN (15 mol%), MeCN, 100°C; silica gel, room temperature. AIBN = azobisisobutyronitrile.

5-*exo* cyclization is not stabilized by the neighboring substituent.[5] Although we showed in our previous studies that the 5-*exo* cyclization is dominant under conditions of kinetic control,[5d] generally applicable conditions for the formation of 2-stannylindoles had yet to be determined. In an attempt to reduce the reaction temperature, we employed 2,2′-azobis(4-methoxy-2,4-dimethylvaleronitrile) (V-70, **20**) as a radical initiator with a lower decomposition temperature.[13] To assess the potential of V-70, we chose substrates without radical-stabilizing substituents. When subjected to the reported conditions (AIBN, 100°C), substrates **16** and **17** afforded a mixture of the 5-*exo* adduct **18** and the 6-*endo* adduct **19**, and cyclization of the *trans* isomer **17** proved to be more difficult than that of the *cis* isomer **16** (Table 1, entries 1 and 4).[14] However, the use of V-70 at 30°C virtually suppressed the formation of the 6-*endo* adduct: after acidic treatment, indole **18** was obtained in good yield as the dominant product, regardless of the configuration of the substrate (Table 1, entries 3 and 6). Thus, we established reliable conditions for imidoyl-radical-mediated indole synthesis.

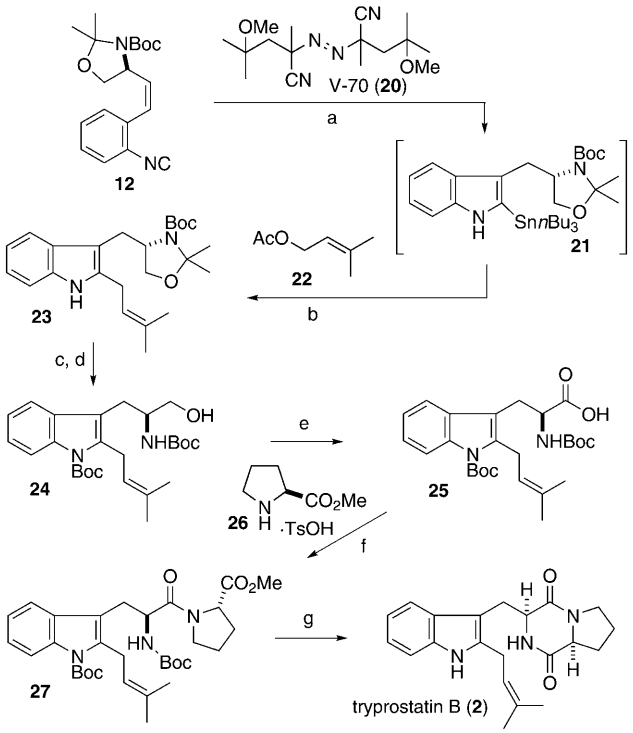
Table 1: Low-temperature radical cyclization.



Entry	Substrate	Initiator	Solvent	T [°C]	**18** [%][d]	**19** [%][d]
1[a]	**16**	AIBN	MeCN	100	72	18
2[b]	**16**	AIBN	toluene	100	64	23
3[c]	**16**	V-70	toluene	30	84	2
4[a]	**17**	AIBN	MeCN	100	51	33
5[b]	**17**	AIBN	toluene	100	48	34
6[c]	**17**	V-70	toluene	30	77	6

[a] See reference [5a] for the reaction conditions. [b] Reaction conditions: nBu$_3$SnH (2.0 equiv), AIBN (15 mol%), toluene (0.2 M), 1 h, 100°C; 1 M aqueous HCl, room temperature. [c] Reaction conditions: nBu$_3$SnH (2.0 equiv), V-70 (**20**, 15 mol%), toluene (0.2 M), 3 h, 30°C; 1 M aqueous HCl, room temperature. [d] Yield of the isolated product.

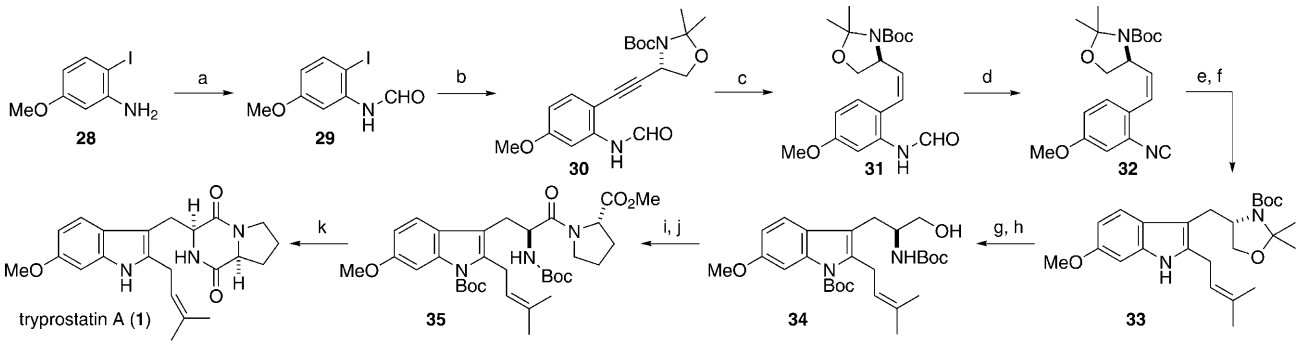
When this method was applied to the radical cyclization of the isocyanide **12**, complete selectivity was observed for the cyclization of the imidoyl radical to give the 2-stannylindole **21** (Scheme 4). Although we previously reported a one-pot Stille-type coupling reaction between aromatic or vinylic halides and 2-stannylindoles generated in situ for the



Scheme 4. Total synthesis of tryprostatin B: a) nBu$_3$SnH (3.0 equiv), V-70 (**20**, 20 mol%), toluene, 30°C; b) **22**, LiCl (3.0 equiv), [Pd$_2$(dba)$_3$] (10 mol%), AsPh$_3$ (40 mol%), DMF, 80°C, 73%; c) Boc$_2$O, DMAP, MeCN, room temperature, 99%; d) TFA/THF/H$_2$O (4:2:1), 0°C, 88%; e) TEMPO, PhI(OAc)$_2$, MeCN, phosphate buffer (pH 6.8), room temperature, 91%; f) **26**, HATU, iPr$_2$NEt, CH$_2$Cl$_2$, room temperature, 88%; g) NMP, reflux, 89%. dba = dibenzylideneacetone, DMAP = 4-dimethylaminopyridine, DMF = *N*,*N*-dimethylformamide, HATU = *O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexafluorophosphate, TEMPO = 2,2,6,6-tetramethylpiperidin-1-oxyl, TFA = trifluoroacetic acid.

preparation of 2,3-disubstituted indoles,[5a] no such coupling reactions with allylic systems have been reported. When [Pd(PPh$_3$)$_4$] was used as the catalyst, the desired product was obtained in only 29% yield with prenyl acetate (**22**) as the coupling partner. Extensive investigations revealed that a combination of [Pd$_2$(dba)$_3$], triphenylarsine, and lithium chloride gave the best results and afforded the desired product **23** in 82% yield in a one-pot process from the isocyanide **12**.

Once construction of the indole moiety was complete, **23** was converted into the corresponding amino acid **25** in a three-step sequence consisting of protection of the indole with a Boc group, hydrolysis of the acetonide, and oxidation of the resulting alcohol **24** to the carboxylic acid **25** with TEMPO. After condensation with L-proline methyl ester



Scheme 5. Total synthesis of tryprostatin A: a) HCO_2H, Ac_2O, CH_2Cl_2, 0 °C, 98 %; b) **7**, CuI, $[PdCl_2(PPh_3)_2]$, Et_3N/THF, room temperature, 98 %; c) Zn, $BrCH_2CH_2Br$, CuBr, LiBr, THF, CF_3CH_2OH, 70 °C, 94 %; d) triphosgene, pyridine, CH_2Cl_2, 0 °C, 85 %; e) nBu_3SnH (3.0 equiv), V-70 (20 mol %), toluene, 30 °C; f) **22**, LiCl (3.0 equiv), $[Pd_2(dba)_3]$ (10 mol %), $AsPh_3$ (40 mol %), DMF, 80 °C, 80 %; g) Boc_2O, DMAP, MeCN, room temperature, 92 %; h) TFA/THF/H_2O (4:2:1), 0 °C, 96 %; i) TEMPO, $PhI(OAc)_2$, phosphate buffer (pH 6.8), MeCN, 0 °C, 91 %; j) **26**, HATU, iPr_2NEt, CH_2Cl_2, room temperature, 77 %; k) NMP, reflux, 78 %.

(**26**), the remaining transformations involved the removal of the two Boc groups in **27** and cyclization to construct a diketopiperazine core structure. However, the conventional method for removing Boc groups under acidic conditions caused substantial decomposition of the substrate. Thus, we attempted thermal removal of the Boc groups.[15] After extensive optimization, this transformation was carried out by heating the substrate under reflux in *N*-methylpyrrolidinone (NMP). Under these conditions, spontaneous cyclization occurred to give tryprostatin B (**2**) in 89 % yield. Thus, tryprostatin B was synthesized in 11 steps from **8** in 33 % overall yield on a half-gram scale.

Having established a generally applicable protocol for the preparation of 2-stannylindoles with V-70, we undertook the total synthesis of tryprostatin A (**1**) to showcase the synthetic utility of this approach (Scheme 5). The synthesis commenced with the preparation of **28** from 4-methoxy-2-nitroaniline in a two-step, slightly modified sequence, including the Sandmeyer reaction and iron-mediated reduction.[4c,16] By following the route established for tryprostatin B, we synthesized tryprostatin A in 30 % overall yield from **28** on a half-gram scale.

In summary, optimization of the radical-mediated indole synthesis by the use of V-70 made it possible to access 2-stannyl 3-substituted indoles with substituents at C3 that cannot effectively stabilize the radical intermediate. In combination with a subsequent palladium-mediated coupling reaction at C2, the radical cyclization enables the preparation of a variety of 2,3-disubstituted indoles, such as tryprostatins A and B. Further investigations into the synthesis and biological activity of tryprostatin analogues are under way.

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[14] Dihydroquinoline intermediate underwent further reduction to afford tetrahydroquinoline **19**.

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[16] See the Supporting Information for a detailed description of the preparation of **28**.

