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Modular Access to Complex Prodiginines: Total Synthesis of (+)-Roseophilin *via* its 2-Azafulvene Prototropisomer

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Abstract

Ansa-bridged prodiginines are bioactive pigments produced by bacteria. Certain of these structures are reported to be antagonists of protein-protein interactions involved in apoptosis. We describe a new entry to alkaloids of this type, demonstrated with a concise asymmetric synthesis of (+)-roseophilin (**3**). Our route constructs the pyrrolophane motif *via* phosphoryl transfer-terminated macroaldolization and passes through a previously unexplored prototropic form of the natural product.

Ansa-bridged prodiginines are lipochromophores produced by both terrestrial and marine bacteria.[1] They derive from *seco* precursors consisting of a prodigiosin heterocycle harboring a long-chain *n*-alkane (e.g. **1**, Figure 1).[2,3] Medium/large rings are formed directly within these materials by way of net dehydrogenation (e.g. **1**→**2**). In the case of streptorubin B (**2**), a specialized non-heme Rieske oxygenase mediates cyclization, putatively *via* intermediate alkyl radical addition to the heterocyclic nucleus.[4,5] Metacycloprodigiosin, prodigiosin R1, and nonylprodigiosin are thought to be regioisomeric products of this remarkable chemistry. Polycyclic congeners derived from more extensive oxidation are also known (*vide infra*).

Our interest in these molecules derives from Shore's finding that streptorubin B potentiates apoptotic signaling in cell culture, reportedly through interactions with mitochondrial Bcl-2 proteins.[6,7] This discovery seeded development of obatoclax, a simplified prodigiosin analog currently being evaluated in humans as therapy for chronic lymphocytic leukemia.[8,9] To ascertain whether functionalized pyrrolophane variants can more selectively antagonize protein-protein contacts gating mitochondrial membrane permeability,[10] we sought generic access to the group. The goal was a modular synthetic route; one amenable to varied heterocyclic components and peripheral substitution. An assembly reminiscent of their biosynthesis was attractive, wherein the *ansa*-bridge would be installed late and in a manner such that the extent and position of its connectivity to the chromophore could be altered. We have reduced this strategy to practice with a concise total synthesis of (+)-roseophilin (**3**), arguably the most complex member of the group.

Roseophilin's distinct structure has drawn considerable attention.[2] It harbors two C-C σ-bonds connecting its hydrocarbon tail to the heterocyclic core, which itself is more highly oxidized relative to **2**. Fürstner's seminal synthesis of **3** constructs the target from two finished segments joined along the C_8-C_9 bond.[11] This blueprint has been influential.

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Supporting Information. Experimental details, characterization data, copies of ^{1}H and ^{13}C NMR spectra for new compounds, and HPLC data. This material is available free of charge via the Internet at http://pubs.acs.org.

Intense activity since has focused on the *ansa*-bridged azatricyclic component, resulting in many creative contributions and a number of formal syntheses.[12,13]

To place roseophilin within a larger target set, we chose different plans. It was useful to contemplate the stability of **3** relative to its 2-azafulvene prototropisomer **4** (Figure 1). Assuming the former was lower in energy, and a path connecting the two was available,[14] one might exploit **4** as an intermediate *en route* to **3**. This was desirable because the synthetic problem simplifies readily from **4**. Net hydration of its azafulvene reveals β-pyrrolyl ketone **5** as a potential precursor. Pyrrolophane **5** resembles simpler *ansa* prodiginines such as **2** and its carbonyl group is a versatile design handle. Options for large ring formations within keto prodigiosins **6** (Scheme 1) became apparent, as did means to establish absolute stereochemistry late in the sequence *via* controlled reduction. The question became how best to assemble achiral structures **6** from fragments, with an eye towards diversifying the route in subsequent iterations.

We targeted generic components **7** and **8**, and sought to link the two such that C_9 in **6** would be at the oxidation state of a ketone. The α-olefin in **8** would facilitate incorporating a third component (i.e. **9**) using alkene cross metathesis. Towards this end, we have developed syntheses of **7**[15] and **8**,[16] each beginning with pyrrole. Our new preparation of **7** occurs in five steps and permits X, Y, and R^1 to be controllably varied.[15] This route provides facile access to multigram quantities of specific roseophilin segment **10**.[17]

To construct a variant of **6** appropriate for synthesis of **3**, methoxyfuran **10** was lithiated at low temperature and the resultant organometallic was treated with $ZnBr_2$. Palladium-catalyzed carboxylation of the incipient zinc species provided carboxylic acid **11**.[18] Condensation with *N*-methylsulfonyl benzotriazole then afforded an active amide, which acylates 2-(8-nonenyl)pyrrole (**8**; n = 7) when aided by $TiCl_4$.[19] This Katritsky protocol scaled effectively and gave mixed bisheteroaryl ketone **12** in high yield.

We originally planned to convert **12** to an azafulvene (e.g. **6**), wherein Z would later participate in an internal cross coupling reaction *en route* to **5**. However, converting the ketone in **12** to either an enol sulfonate or a vinyl halide proved difficult. Attempts at the former resulted in *N*-sulfonylation. Finding means to exploit this outcome led to a new pyrrolophane synthesis.

Consistent with earlier observations, treatment of **12** with potassium hydride and diethyl chlorophosphite gave the *N*-phosphinyl derivative, which oxidized to phosphoramide **13** upon exposure to air.[20] Metathesis of **13** with isopropyl propenyl ketone (**14**)[21] then provided a chain-homologated enone, which was reduced *in situ* employing palladium catalyzed hydrosilylation.[22] Hydrolysis of the resultant silyl enol ether during workup afforded diketone **15** as an amber oil.

Analogous to sulfonyl-transfer reactions implicated in hydride reductions of *N*-tosyl-2-acylpyrroles,[23] the phosphoramide in **15** was intended as an internal trap for carbon nucleophiles added to the C_9 carbonyl. When **15** was deprotonated with KHMDS at low temperature, quenching the reaction with water returned starting material. The same was true when an equivalent of 18-crown-6 was added to the medium and the mixture warmed to rt prior to protonation. However, when the enolate formed from the crown ether/KHMDS combination was brought to 55 °C, we observed gradual formation of pyrrolophane **19** (Scheme 2). After 18 h, substrate **15** was fully consumed and macrocycle **19** was isolated in 66% yield. We speculate that **19** derives from the minor component in an initial equilibrium. Namely, one established between kinetic enolate **16** and hindered internal aldol salt **17**. At low temperature and as unmodified ion pairs, these species regenerate **15** upon protonation.

However, given sufficient energy in the presence of a potassium chelator, unimolecular *N* to *O* phosphoryl-transfer can stabilize the aldol adduct as β-phosphoryl ketone **18**. Subsequent elimination of potassium diethylphosphate affords **19**.

Relative to enone **19**, roseophilin (**3**) lies two electrons lower in oxidation state. Samarium diiodide can reduce the C_9-C_{22} olefin to provide **21**, albeit as a racemic mixture of diastereomers. Until recently, one may have been content with that outcome. Methods to controllably saturate electron rich tetrasubstituted enones are few. Fortunately, we were beneficiaries of a recent study by scientists at Eli Lilly. Through screening they identified a chiral rhodium complex and Lewis acid combination able to catalyze the partial hydrogenation of highly substituted chalcones.[24] Adapting this protocol to our system involved hydrogenating **19** (H_2, 100 bar) in the presence of a catalyst generated from $Rh(cod)_2OTf$ and a JosiPhos ligand.[25] Consistent with precedent, turnover required co-catalytic $Zn(OTf)_2$ and MeOH co-solvent. Under these conditions, we obtained *cis* β-pyrrolyl ketone **21** with high diastereoselectivity (>25:1). Furthermore, when the catalyst was formed using enantiopure bis-phosphine **20**, product (+)-**21** was isolated in 92% yield and 67% ee.[26,27]

Compound **21** is an oxygenated structural isomer of prodigiosin R1. It is also a hydrated form of roseophilin (**3**). Among conditions found to dehydrate **21**, catalysis by $[ReBr(CO)_3(thf)]_2$ was most effective.[29] 10 mol % of this Lewis acid smoothly induced cyclodehydration, affording unstable 2-azafulvene **22**. It was best not to handle **22**, but rather treat the material *in situ* with dry HCl and substoichiometric amounts of *t*-BuOH. This provided roseophilin hydrochloride directly (Scheme 2). With minimal handling, roseophilin was obtained in 32% overall yield from enone **19**. ^1H and ^{13}C NMR data for synthetic **3**.HCl are indistinguishable from that reported for the natural product and fully consistent with the structure assignment.

Intermediate **21** could also be desilylated with CsF. Dehydration of the product with catalytic $[ReBr(CO)_3(thf)]_2$ affords *iso*-roseophilin **4** (Scheme 3). This reactive substance could be characterized, although loss during isolation is significant. It degrades intractably on standing ($t_{1/2}$ < 1h at rt). Reduction of crude **4** with SmI_2 in MeOH afforded dihydro-roseophilin **23**; an air-sensitive molecule that DDQ converts cleanly to roseophilin. When $NaBH_4$ was used to reduce **4**, epimer **24** was formed in significant amounts. Upon standing in air (0.1 M $CHCl_3$ solution, rt, 18 h), a mixture **23** and **24** converted only to roseophilin. Epimer **23** was oxidized while **24** remained largely unchanged. DDQ treatment degraded **24** rather than form a diastereomer of **3**. Additional studies on these fascinating structures are ongoing.

In conclusion, we have completed the shortest synthesis of roseophilin to date. Phosphoryl-transfer terminated macroaldolization uniquely installs the *ansa*-bridge. It does so at an oxidation state where saturated asymmetry can be induced *via* reduction late in the sequence. We expect the route to accommodate changes in ring sizes and substitution patterns, providing analogs that would be otherwise difficult to prepare. Since the C_{23} substituent follows from the choice of metathesis partner **9**, and because our synthesis of heterocycle **10** tolerates varying halogen and alkoxy groups,[15] there is design flexibility at multiple points along the angled periphery of the polyheterocycle. We can test whether roseophilin and its relatives are ligands for anti-apoptotic Bcl-2 proteins and probe in detail whether the hetero-cyclic backbone is a scaffold upon which new α-helix mimetics can be developed. Work along these lines is ongoing, as are attempts to adapt the route to syntheses of other members of this important group of natural products.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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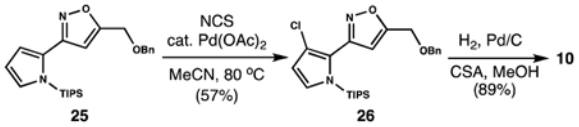
References

1. (a) Williamson NR, Fineran PC, Leeper FJ, Salmond GPC. Nat Rev Microbiol. 2006; 4:887–899. [PubMed: 17109029] (b) Williamson NR, Fineran PC, Gristwood T, Chawrai SR, Leeper FJ, Slamond GPC. Future Microbiol. 2007; 2:605–618. [PubMed: 18041902]
2. Fürstner A. Angew Chem, Int Ed. 2003; 42:3582–2603.
3. (a) Stanley AE, Walton LJ, Kourdi-Zerikly M, Corre C, Challis GL. Chem Commun. 2006:3981–3983.(b) Mo SJ, Sydor PK, Corre C, Alhamadsheh MM, Stanley AE, Haynes SW, Song L, Reynolds KA, Challis GL. Chem Biol. 2008; 15:137–148. [PubMed: 18291318] (c) Haynes SW, Sydor PK, Stanley AE, Song L, Challis GL. Chem Commun. 2008:1865–1867.
4. Sydor PK, Barry SM, Odulate OM, Barona-Gomez F, Haynes SW, Corre C, Song L, Challis GL. Nat Chem. 2011; 3:388–392. [PubMed: 21505498]
5. Wasserman HH, Shaw CK, Sykes RJ, Cushley RJ. Tetrahedron Lett. 1974; 15:2787–2790.
6. Murthy MSR, Steenarrt NAE, Johnson RA, Shore GC. PTC Int Appl WO 2001055131. 2001:267.
7. (a) Lessene G, Cazbotar PE, Colman PM. Nat Rev Drug Disc. 2008; 7:989–1000.(b) Kang MH, Reynolds CP. Clin Cancer Res. 2009; 15:1126–1132. [PubMed: 19228717]
8. (a) Nguyen M, Marcellus RC, Roulston A, Watson M, Serfass L, Madiraju SRM, Goulet D, Viallet J, Bélec L, Billot X, Acoca S, Purisma E, Wiegmans A, Cluse L, John-stone RW, Beauparlant P, Shore GC. Proc Natl Acad Sci USA. 2007; 104:19512–19517. [PubMed: 18040043] (b) Acoca S, Cui Q, Shore GC, Purisima EO. Proteins. 2011:2624–2636. [PubMed: 21721047]
9. Phase I/II of obatoclax (GX15-070) in untreated chronic lymphocytic leukemia (CLL): ClinicalTrials.gov Identifier: NCT00600964.
10. Obatoclax triggers apoptosis in Bax/Bak deficient embryonic fibroblasts. Its selectivity as a BH3 helix mimetic is thus a subject of debate. See: van Delft MF, Wei AH, Mason KD, Candenberg CJ, Chen L, Czabotar PE, Willis SN, Scott CL, Day CL, Cory S, Adams JM, Roberts AW, Huang DCS. Cancer Cell. 2006; 10:389–399. [PubMed: 17097561] Vogler M, Weber K, Dinsdale D, Schmitz I, Schulze-Osthoff K, Dyer MJS, Cohen GM. Cell Death Differ. 2009; 16:360–367. [PubMed: 18806758]
11. Fürstner A, Weintritt H. J Am Chem Soc. 1998; 120:2817–2825.
12. Total syntheses of **3**: Harrington PE, Tius MA. J Am Chem Soc. 2001; 123:8509–8514. [PubMed: 11525658] Boger DL, Hong J. J Am Chem Soc. 2001; 123:8515–8519. [PubMed: 11525659]
13. Formal syntheses and approaches to **3**: (a) Reference 2 and references therein. Occhiato EG, Prandi C, Ferrali A, Cuarna A. J Org Chem. 2005; 70:4542–4545. [PubMed: 15903343] Salamone SG, Dudley GB. Org Lett. 2005; 7:4443–4445. [PubMed: 16178554] Song C, Knight DW, Whatton MA. Org Lett. 2006; 8:163–166. [PubMed: 16381593] Bitar AY, Frontier AJ. Org Lett. 2009; 11:49–52. [PubMed: 19053717] Song C, Liu H, Hong M, Liu Y, Jia F, Sun L, Pan Z, Chang J. J Org Chem. 2012; 77:704–706. [PubMed: 22098172] Kerr DJ, Flynn BL. Org Lett. 2012; 14:1740–1743. [PubMed: 22455601]
14. A concerted sigmatropic shift of C_9-H to C_{23} within structure **4** is improbable. However, successive bimolecular protonation/deprotonation events appeared a viable path to **3** from **4**. The 3D structure of conformer **4** shown in Fig. 1 (Spartan; B3LYP) was rendered with CYLview 1.0b (http://www.cylview.org).
15. Isoxazolylpyrrole **25** was assembled in three steps from commercial dibromoformaldoxime, benzyl propargyl ether, and pyrrole. Substrate-directed, palladium catalyzed chlorination provided a single isomer of **26**. Hydrogenolysis of **26** and treatment of the resultant enaminone with CSA/

MeOH *in situ* afforded **10**. Full details of this route (5 steps, 19% overall yield) and application of related methods in syntheses of congeners **7** will be reported separately.



16. Pyrrole **8** (n = 7) has been reported previously, see: Aldrich LN, Dawson ES, Lindsley CW. Org Lett. 2010; 12:1048–1051. [PubMed: 20141121] We describe a shortened synthesis in the Supporting Information.

17. For previous syntheses of **10**, see: (a) Reference 11. Nakatani S, Kirihara M, Yamada K, Terashima S. Tetrahedron Lett. 1995; 36:8461–8464.Kim SH. PhD Thesis. Purdue University1998

18. Yeung CS, Dong VM. J Am Chem Soc. 2008; 130:7826–7827. [PubMed: 18510323]

19. Katritzky AR, Suzuki K, Singh SK, He HY. J Org Chem. 2003; 68:5720–5723. [PubMed: 12839468]

20. Lee K, Wiemer DF. J Org Chem. 1991; 56:5556–5560.

21. Oare DA, Henderson MA, Sanner MA, Heathcock CH. J Org Chem. 1990; 55:132–157.

22. Sumida Y, Yorimitsu H, Oshima K. J Org Chem. 2009; 74:7986–7989. [PubMed: 19761266]

23. Greenhouse R, Ramirez C, Muchowski JM. J Org Chem. 1985; 50:2961–2965.

24. Calvin JR, Frederick MO, Liard DL, Remacle JR, May SA. Org Lett. 2012; 14:1038–1041. [PubMed: 22288716]

25. Refinements are ongoing. For catalyst screening to date, see the Supporting Information.

26. The same reaction employing the antipode of **20** provided scalemic **5** enriched in the opposite enantiomer (70% yield, 65% ee).

27. A diastereomeric mixture of **5** (d.r. >25:1) enriched in the *cis* isomer epimerizes at C_{22} to afford largely the corresponding *trans* diastereomer (1:4 *cis:trans*) upon exposure to DBU (0.5 M THF, rt, 48 h).

28. Kuninobu Y, Tasuzaki T, Matsuki T, Takai K. J Org Chem. 2011; 76:7005–7009. [PubMed: 21761938]

29. The cyclodehydration protocol was optimized in a model system. For details, see Supporting Information.

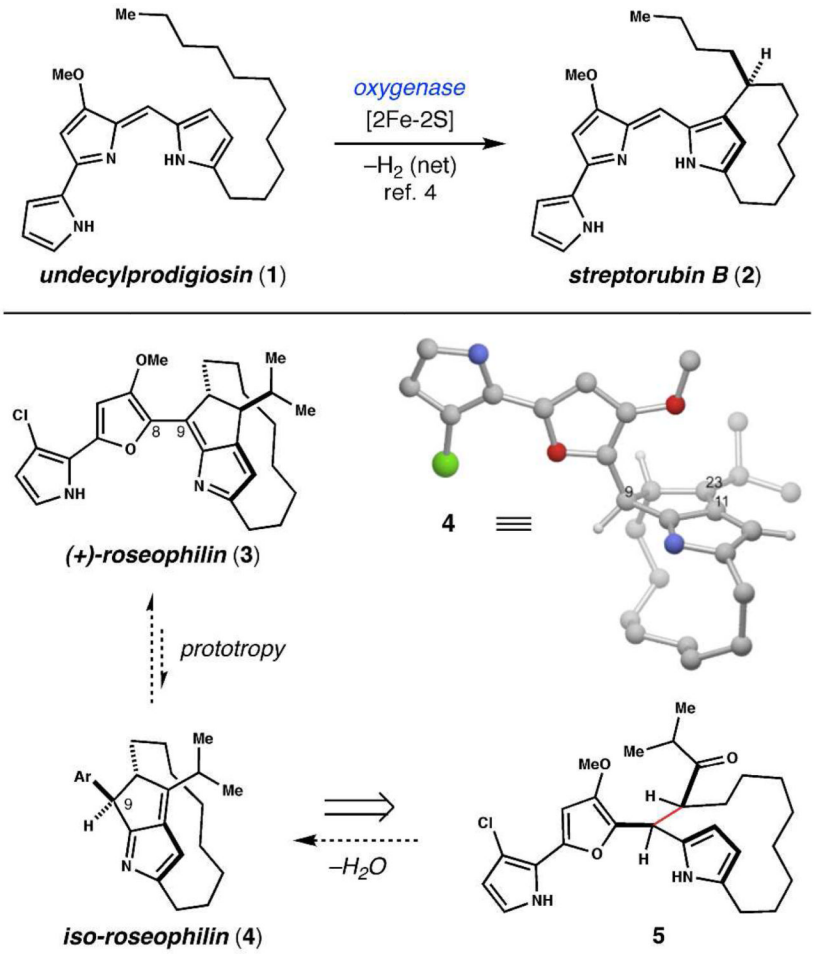
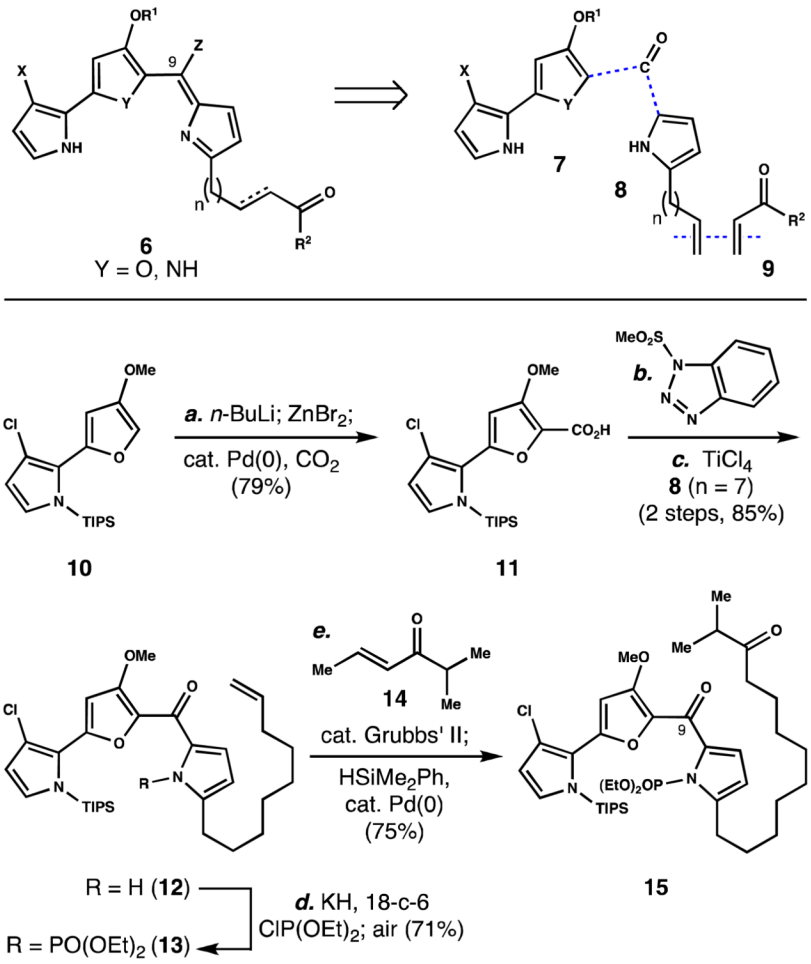


Figure 1.

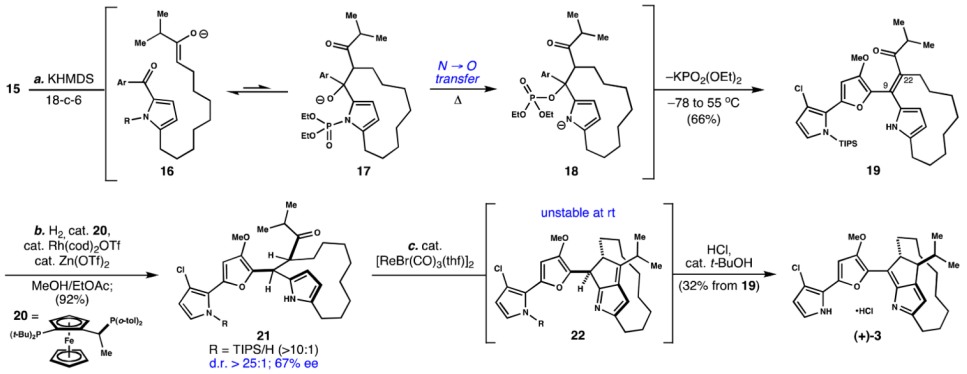
Ansa-bridged prodiginines are biosynthesized from *seco* hydrocarbons such as **1**. Roseophilin (**3**) and related structures can be approached in an analogous manner by exploiting the intermediacy of azafulvene prototropisomer **4**.



Scheme 1.

Design and assembly of *seco* precursors.

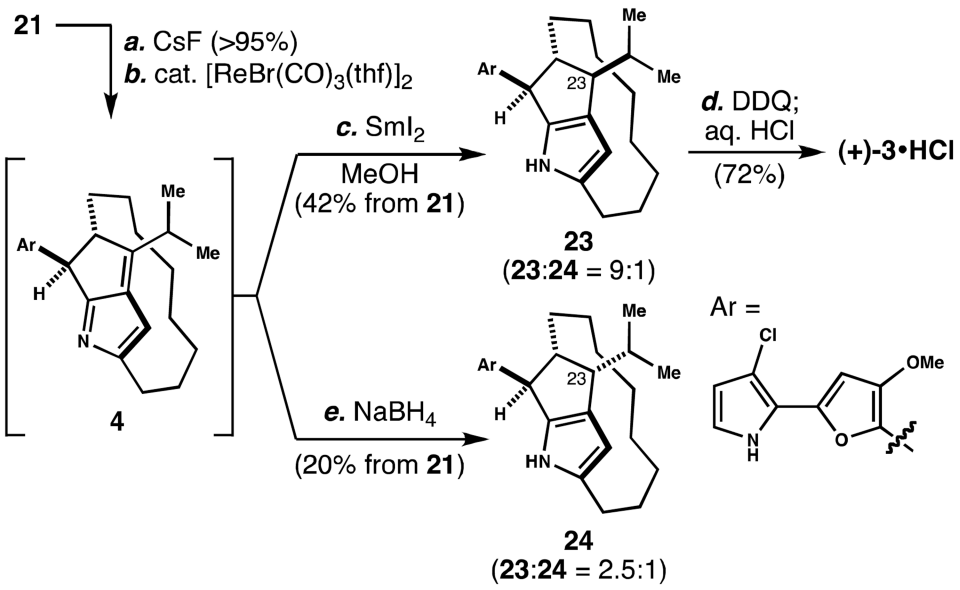
Reagents and conditions: (a) *n*-BuLi, THF, –78 °C, 30 min; $ZnBr_2$ (1.0 M in THF), -78 °C to 0 °C, 2 h; 5 mol % $Pd(OAc)_2$ 10 mol % PCy_3, CO_2 (1 atm), THF, rt, 24 h, 79%. (b) 1-(methanesulfonyl)-1*H*-benzotriazole, Et_3N, THF, reflux, 18 h. (c) **8** (n = 7), 2 equiv $TiCl_4$, CH_2Cl_2, 0 °C, 3 h, 85% from **10**. (d) KH, 18-c-6, THF, rt; $ClP(OEt)_2$, 0 °C, 1 h; air, 18 h, rt, 71%. (e) **14**, 5 mol % $PCy_3Cl_2(iMes)Ru{=}CHPh$, CH_2Cl_2, rt, 18 h; 5 mol % $Pd(OAc)_2$, 10 mol % PCy_3, $HSiMe_2Ph$, PhMe, 60 °C, 6 h, 75%.



Scheme 2.

Total synthesis of (+)-roseophilin (**3**).

Reagents and conditions: (a) 2.2 equiv KHMDS, 18-c-6, THF, -78 °C to 55 °C, 18 h, 66%.* (b) 5 mol % $Rh(cod)_2OTf$, 5 mol % **20**, 7.5 mol % $Zn(OTf)_2$, H_2 (100 bar), MeOH/EtOAc (1:1), rt, 24 h. (c) 10 mol % $[ReBr(CO)_3(thf)]_2$, $(CH_2Cl)_2$, 70 °C, 4 h; 25 mol % *t*-BuOH, HCl, dioxane, –78 °C to rt, 4 h, 32% from **19**. * Dephosphorylated **15** (5–7%) was also isolated in this experiment.



Scheme 3.

Reductions of *iso*-roseophilin (**4**).

Reagents and conditions: (a) CsF, THF, rt, 3 h, >95%. (b) 10 mol % $[ReBr(CO)_3(thf)]_2$, $(CH_2Cl)_2$, 70 °C, 4 h. (c) SmI_2, MeOH, THF, –78 °C to rt, 42% from **21**. (d) DDQ, CH_2Cl_2, rt, 5 h, 72%. (e) $NaBH_4$, THF, 50 °C, 3 h, 20 % from **21**.