A Highly Stereoselective Total Synthesis of Hispidospermidin: Derivation of a Pharmacophore Model

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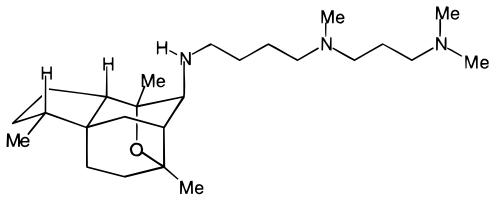
Abstract: The total synthesis of the title compound has been accomplished. Among the key steps were (i) a conjugate addition−Robinson annulation-type sequence (see **4**), (ii) intramolecular carbomercuration (see **3**), (iii) a reduction−ketonization sequence (see **25**), (iv) cycloetherification of an unactivated methylene group (see **28**), and reductive amination (see **1**). A highly preliminary SAR profile suggests that the functional cytotoxic pharmacophore of hispidospermidin involved a presentation of spermidine derivative **36** via linkage to a ball-like hydrophobic cage to its target.

Isolation, Structure, and Biological Activity

The role of the phospholipase C (PLC)-mediated transduction pathway in cell proliferation was an important factor in setting the stage for discovery of the natural product, hispidospermidin (**1**; Figure 1). A screen for active inhibitors of PLC, involving incubation of phosphoinositol with phospholipase C in the presence of a sample of microbial culture broth, was instituted by workers at Nippon Roche.[1a] Hispidospermidin, isolated from fermentation of the microorganism *Chaetospaeromena hispidulum* with the aid of this bioassay, was found to inhibit 50% of the phospholipase C activity at a concentration of 16 μM (IC_{50}). The inhibition is dose-dependent and is apparently selective for this enzyme. Hispidospermidin was also cytotoxic to HeLa cells at an IC_{50} of 36 μM.[1a]

The structural elucidation of hispidospermidin was accomplished by close analysis of data from a variety of high-field NMR experiments, including ^{1}H, ^{13}C, ^{1}H−^{1}H COSY, ^{13}C−^{1}H COSY, NOESY, and long-range *J* C−H resolved 2D spectroscopy.[1] The absolute configuration of hispidospermidin was assigned by application of Mosher's method to the primary amine derived from **1**.

The potential importance of a compound such as hispidospermidin from the point of view of drug exploration is connected to the role of the enzyme PLC in mediating the hydrolysis of phosphatidylinositol 4,5-bisphosphate (PIP_2).[2] Cleavage of PIP_2 leads to the production of two important second messengers, diacylglycerol (DAG) and inositol 1,4,5-triphosphate (IP_3). DAG activates protein kinase C (PKC), which, in turn, mediates the phosphorylation of other signaling proteins, thereby governing cell division and proliferation. Concurrently, IP_3 levels control the release of intracellular calcium, affecting the activity profiles of a variety of proteins and enzymes.[3] In principle, inhibition of PLC could well alter the signaling system of the target, thereby affecting its cell cycle progression.



hispidospermidin (**1**)

Figure 1.

Synthetic Planning

Quite aside from our biology-driven interest in a naturally occurring PLC inhibitor, the striking architecture of hispidospermidin invites proposals for its synthesis. Particularly challenging in this regard was the goal of implementing an efficient construction of the novel caged sector of the molecule. An early generalized view of our synthetic plan is depicted in Scheme 1.[4−6]

While we were not certain at the inception of the project as to how or when the spermidine side chain at C11 would be introduced, it was presumed that a way could be found via ketone **2**. This compound emerged as the focus of the planning exercise. It was thought that a hydrindenone of the type **4**, bearing an angular butynyl function, could provide a useful framework for future elaboration. While the particulars would be worked out by experimentation, from a global perspective it was necessary to introduce a β-hydrogen at C9, a β-methyl at

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(1) (a) Yanagisawa, M.; Sakai, A.; Adachi, K.; Sano, T.; Watanabe, K.; Tanaka, Y.; Okuda, T. *J. Antibiot.* **1994**, *47*, 1. (b) Ohtsuka, T.; Itezono, Y.; Nakayama, N.; Sakai, A.; Shimma, N.; Yokose, K.; Seto, H. *J. Antibiot.* **1994**, *47*, 6.

(2) (a) Potter, B. V. L.; Lampe, D. *Angew. Chem., Int. Ed. Engl.* **1995**, *34*, 1933. (b) Magerus, P. W.; Connolly, T. M.; Deckmyn, H.; Ross, T. S.; Bross, T. E.; Ishii, H.; Bansal, V. S.; Wilson, D. B. *Science* **1986**, *234*, 1519.

(3) Berridge, M. J.; Irvine, R. F. *Nature* **1984**, *312*, 315.

(4) For a preliminary communication, see: Frontier, A. J.; Raghavan, S.; Danishfsky, S. J. *J. Am. Chem. Soc.* **1997**, *119*, 6686.

(5) For a thorough treatment of this work, see: Frontier, A. J. Ph.D. Thesis, Columbia University, 1999.

(6) For an asymmetric synthesis of hispidospermidin, see: Overman, L. E.; Tomasi, A. L. *J. Am. Chem. Soc.* **1998**, *120*, 4039.

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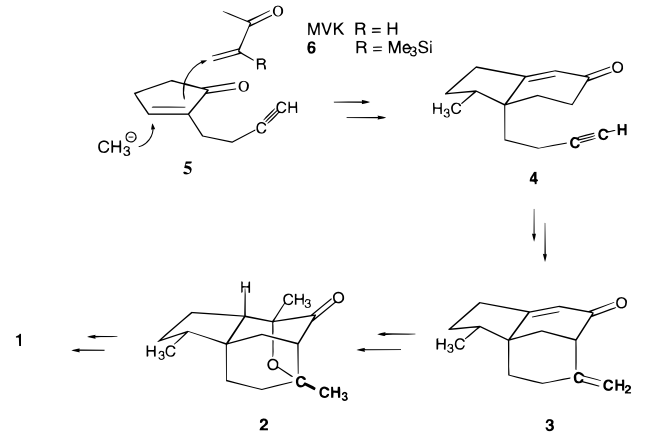
Scheme 1



C10, and a β-disposed polyamine at C11.[7] Placement of an α-oxygen at C10 for the purpose of bridging C10 and C2 would be necessary, as would provision for a bond to connect C12 and C2. The cage of **2** would be completed by addition of the α-oxygen, introduced at C10, to C2. These transformations could constitute a unique approach in that a terminal ethynyl linkage ultimately would have emerged as the quaternary C-methyl moiety (see boldface carbons in structures **2** and **4**).

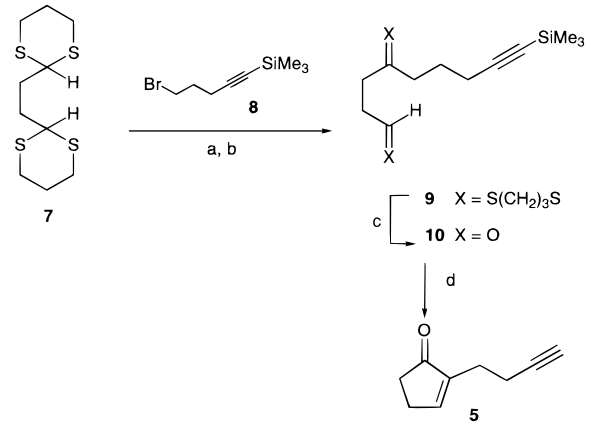
Critical to the success of our prospectus would be the optimal phasing of these steps to achieve the desired stereodirectionality. We were concerned about potential difficulties in maintaining tight stereochemical control in operations at C9 and C10 of hydrindenone **4**. Accordingly, carbon matrix **3**, which could arise from **4** by the joining of carbons 2 and 12 was considered an attractive milestone target. The pronounced cuplike character of its bicyclo[3.3.1]nonane domain would help to provide more significant stereochemical guidance.

Many strategies were considered to lead us to ketone type **4**. In the end, the preferred design was rather classical in conception. It was anticipated that conjugate addition to cyclopentenone **5** would generate a site-specific enolate that could be trapped with an appropriate vinyl ketone electrophile. It was hoped that the alkylation event at the α-carbon would occur from the face opposite the methyl substituent installed by the conjugate addition step. Cyclization of the resultant diketone, through completion of the Robinson annulation sequence, would then provide the desired enone (see **4**).

We were mindful that methyl vinyl ketone (MVK) itself is highly reactive and has a tendency to polymerize under aprotic basic conditions of the kind that would be critical for maintenance of the integrity of the kinetic enolate arising from the 1,4-addition step. Fortunately, several surrogates have been developed that are less vulnerable to polymerization, while providing access to the same ultimate products as does MVK with a given ketone substrate. We hoped to use the Stork-Ganem variant **6**, which bears a trimethylsilyl group at the α-position of the enone, as our MVK equivalent.[8] It had an established record as an electrophile that gives rise to an adduct that can be subjected to site-specific alkylation en route to Robinson annulation-type products.

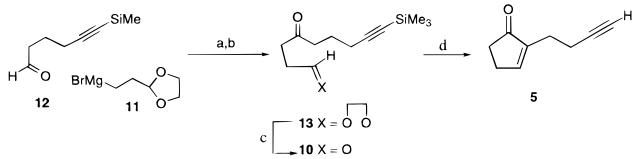
We now consider the synthesis of cyclopentanone **5**, which was unknown at the time. At first glance, from the perspective of the power of modern organic synthesis, this compound could be construed to be a rather simple target. For our purposes it would be necessary to prepare substantial quantities of **5** if it were to serve as the launching point for our investigations. In practice, meeting this need was not at all simple and a variety of initiatives failed. Of the many approaches surveyed, only two were encouraging enough to warrant further study in detail. In the first, butyllithium-induced deprotonation of **7** (see Scheme 2) was followed by alkylation with bromide **8** to give dithioketal **9**. This method suffered greatly from the insolubility of the lithio species derived from **7**. As a result, the alkylation of **7** could only be carried out on limited amounts of material, and after removal of the thioketal and the alkynyl trimethylsilyl group, we were left with insufficient amounts of cyclopentenone **5**. Eventually, this problem became an insurmountable obstacle to progress.

Scheme 2[a]



[a] Reagents and conditions: (a) *n*-BuLi, −30 °C, and then (b) bromoalkyne **8** (1.5 equiv) (85−93%). (c) CAN, acetone. (d) 1% NaOH/Et_2O (1:2), 3 days, 25−50% (two steps). ×b0

Scheme 3[a]



[a] Reagents and conditions: (a) 2 equiv of **11**, −78 °C. (b) PDC (77% from **12**). (c) 1:1 THF/3 N HCl. (d) Et_2O/1% NaOH (3:1), room temperature, 3 days (50% from **13**).

The protocol that eventually proved to be most amenable to large-scale processes started with condensation of the known Grignard reagent **11**[9] with aldehyde **12**[10] (see Scheme 3). The product carbinol was oxidized (PDC) to afford **13** (77% yield from **12**). Deprotection, as shown, gave keto aldehyde **10**, thereby converging with the bisdithiane route (Scheme 2). Aldolization of **10** (accompanied by desilylation) afforded **5** (50% from **13**).[11] The procedure allowed conversion of 32 g of aldehyde **12** to ∼9 g of **5**. This route, which requires six steps from 5-hexyn-1-ol to provide cyclopentenone **5**, is hardly ideal. However, it sufficed for our purposes.

With the required cyclopentenone in hand, we turned to the next phase of our scheme. Addition of lithium dimethylcuprate to **5** was followed by trapping of the metalloenolate with enone **6** (see Scheme 4). Treatment of the resulting crude adduct with

(7) Carbon numbering anticipates the hispidospermidin numbering system introduced in ref 1.

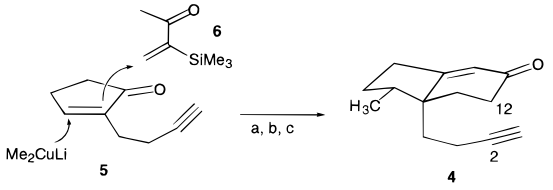
(8) (a) Stork, G.; Ganem, B. *J. Am. Chem. Soc.* **1973**, *95*, 6152. (b) Boeckmann, R. K., Jr.; Blum, D. M.; Ganem, B.; Halvey, N. *Org. Synth. Collect. Vol. VI* **1988**, 1033.

(9) Büchi, G.; Wüest, H. *J. Org. Chem.* **1969**, *34*, 1122.

(10) Bierer, D. E.; Kabalka, G. W. *Org. Prep. Proc. Int.* **1988**, *20*, 63.

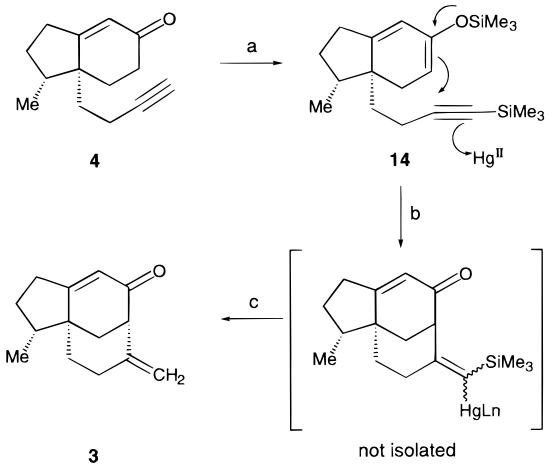
(11) Stork, G.; Ozorio, A. A.; Leong, A. Y. W. *Tetrahedron Lett.* **1978**, 5175.

Scheme 4[a]



[a] Reagents and conditions: (a) Me_2CuLi, −78 °C. (b) **6** (2 equiv), −35 °C. (c) MeOH/4% KOH (4:1), reflux 18 h (22−55% from **5**).

Scheme 5[a]



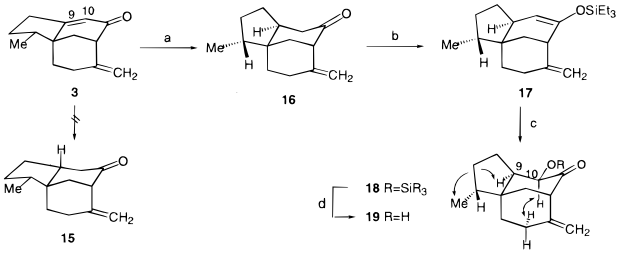
[a] Reagents and conditions: (a) LHMDS, NEt_3, TMSCl, −78 °C. (b) $HgCl_2$, HMDS. (c) NaI, 5.0 N HCl (87% from **4**).

KOH and methanol effected conversion to the desired hydrindenone **4**.[12] Close examination of the ^{1}H NMR spectrum of the crude product indicated that the reaction sequence had produced ~10% of its diastereomer. In light of the fact that a single methyl group is responsible for the stereochemical guidance, the lack of greater specificity was not surprising. The overall yield for the three-step sequence, when conducted on a multigram scale, tended to range from 25 to 30%. For smaller scale preparations, yields of 50−55% of **4** could be realized routinely.

Our next goal was that of progressing from the relatively planar arrangement in **4** to the cuplike tricyclic structure **3**. Attainment of this end would require the fashioning of a carbon−carbon bond between the positions destined to become C2 and C12 of the eventual hispidospermidin target. For this purpose, we planned to employ a cyclization strategy based on intramolecular carbomercuration of the terminal alkynyl linkage.[13] Our thought had been to create nucelophilic character at C12 by means of fashioning the C11−C12 cross-conjugated silyl enol ether. It was hoped that electrophilic attack of the terminal butynyl function would trigger the required cyclization. After our work was well in progress, publications by Huang and Forsythe provided examples of related chemistry.[14]

We commenced with enol silylation of **4** under experimental conditions anticipated to provide kinetic control in the enolate formation step (see Scheme 5). Not unexpectedly, the terminal alkynyl carbon was also silylated in the process. No extensive attempts were made to purify the primary product of this reaction. Rather, reaction of **14** with mercury(II) chloride in the presence of hexamethyldisilazane, followed by acid-induced demercuration with sodium iodide/aqueous HCl afforded an 87% yield of **3**.

Scheme 6[a]



[a] Reagents and conditions: (a) Li/NH_3, *t*-BuOH, −33 °C (85%). (b) Et_3SiOTf, NEt_3. (c) *m*-CPBA, $NaHCO_3$. (d) TBAF (79% from **16**).

With much of the functionality required to gain access to target system **2** seemingly well in hand, attentions were directed to the α,β-unsaturated ketone system. We hoped to exploit this linkage for introduction of the β-methyl, and α-hydroxyl groups at C10, in addition to a β-disposed proton at C9. We envisioned attack by the properly configured C10 tertiary hydroxyl group upon the terminal methylene group as completing the construction of the bridged ether system with emergence of a neopentyl-type methyl group at C2. It was noted that such a ring closure could well benefit from the enforced proximity of the hydroxyl and exo methylene centers in cyclization precursors. We turned first to the matter of introduction of a β-configured proton at C9.

Our thoughts for reaching this goal included the possibility of Birch-like reduction of the α,β-unsaturated ketone of **3**. The hope was that the angular β-carbanionoid species arising from reduction of **3** would assume a pretransoid conformation prompting protonation from the exo face of the local bicyclo[3,3,1]nonanone system (see **15**), as observed in the analogous reductions of many fused octalone systems.[15] On the other hand, it could also be argued that the sense of hybridization of the carbanion-like species at C9 would reflect the emergence of the cis hydrindanone structure since, in general, hydrindanones tend to be more stable in cisoid fusions.[16] In the event, reduction of **3** through the action of lithium in ammonia (with *tert*-butyl alcohol as the proton source) led to isolation of a dihydro product in 85% yield (see Scheme 6). At this stage it was difficult to offer a definitive assignment as to the fusion mode of this compound.

With this important issue unresolved, we probed the preparation of α-functionalized derivatives of the Birch product. The presence of the three-carbon bridge at position 12 dictated that enolization of the C11 ketone occur toward C10. Thus, formation of the silyl enol ether was uneventful, and oxidation in a Rubottom-like process was effected through its reaction with *m*-chloroperoxybenzoic acid.[17] This reaction was followed by workup with sodium bicarbonate. The silyl function, which had migrated to the newly introduced alcohol from the Rubottom reaction (see **18**), was cleaved through the action of tetra-*n*-butylammonium fluoride. At this point we had in hand a compound whose stereochemistry could be assigned. Thus, 1D NMR and NOE difference experiments revealed that the hydrogen at C9 of this Rubottom product is syn to the secondary methyl group. Furthermore, the proton at C10 is in close spatial proximity to a proton allylic to the C13 methylene group. These

(12) Boeckmann, R. K., Jr.; Blum, D. M.; Ganem, B. *Org. Synth. Collect. Vol. VI* **1988**, 666 and references therein.

(13) Drouin, J. D.; Boaventura, M.-A.; Conia, J.-M. *J. Am. Chem. Soc.* **1985**, *107*, 1726.

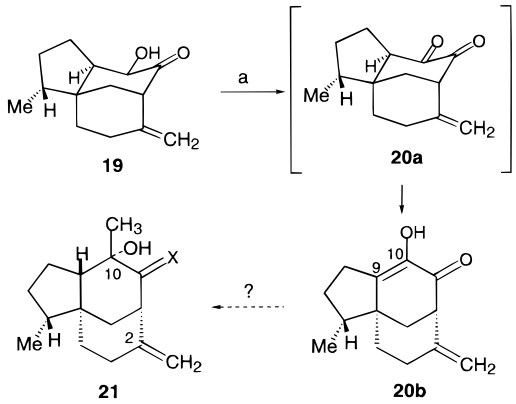
(14) (a) Huang, H.; Forsyth, C. J. *J. Org. Chem.* **1995**, *60*, 2773. (b) Huang, H.; Forsyth, C. J. *J. Org. Chem.* **1995**, *60*, 5746.

(15) (a) Stork, G.; Darling, S. D. *J. Am. Chem. Soc.* **1960**, *82*, 1513. (b) Stork, G.; Darling, S. D. *J. Am. Chem. Soc.* **1964**, *86*, 1761.

(16) Caine, D. *Org. React. (N. Y.)* **1976**, *23*, 1.

(17) Rubottom, G. M.; Gruber, J. M. *J. Org. Chem.* **1978**, *43*, 1599.

Scheme 7[a]



[a] Reagents and conditions: (a) TFAA, DMSO, NEt_3 (80%).

data served to define the stereochemistry of the desilylated oxidation product to be that shown as **19**. This compound would have been derived from the cis-fused Birch reduction product **16** (by way of its silyl enol ether **17**).

Prior to clarification of the stereochemistry of the Birch reduction product **16**, many attempts were made to advance toward hispidospermidin via the α-hydroxyketone **19**. These attempts were frustrated by a variety of α-ketol shifts and adventitious oxidations, targeted at the labile C10, C11 enediol linkage. Once it was established that the fusion was actually cis, we considered the possibility of exploiting diosphenol **20b** (Scheme 7), a compound identified as the product of inadvertent oxidation of **19** (vide infra), during the above investigations. It was possible to efficiently access **20b** through a Swern-type protocol,[18] presumably through the primary oxidation product, **20a**. Once again, the presence of the three-carbon bridge had served to control the sense of enolization of the α-diketone function.

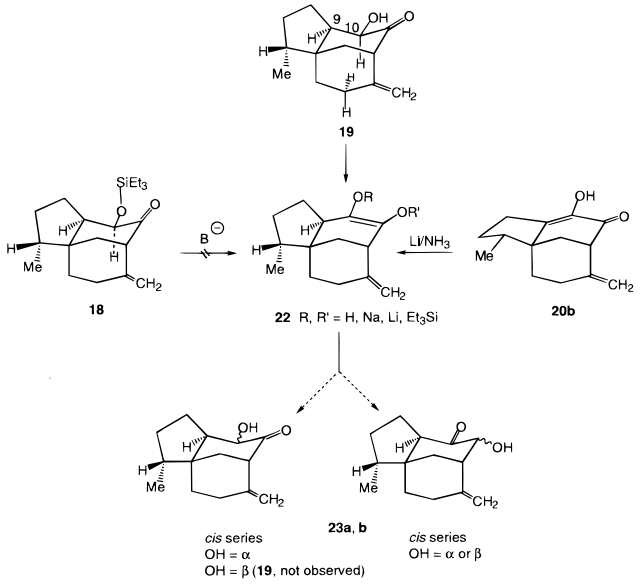
As a consequence of sequential Rubottom and Swern oxidations, we had achieved two potentially important goals. The unwanted α-stereochemistry chemistry at C9 resulting from the Birch reduction had been eliminated, and a potentially useful oxygen was introduced at C10. With **20b** in hand, we set our sights on the installation of a *β*-hydrogen at C9 and a ketone at C10, while maintaining the critical oxygen at carbon 11 (see **21**).

In executing the Rubottom reaction on the silyl enol ether, now known to be **17**, we were able to purify **18**, where the C10 silyloxy group was still intact. Thinking, at the time, that this compound might possibly be in the trans series, we attempted to introduce a methyl group at C10 by deprotonation of the α-siloxyketone at C10 and alkylation of the resultant enolate. Remarkably, a variety of attempts to achieve the required deprotonation were unsuccessful. Apparently, the presence of the siloxy function at C10 of **18** and the steric hindrance of the three-carbon bridge conspired to prevent access of a variety of strong bases to achieve the required deprotonation (Scheme 8).

Another source of difficulty and confusion at the time, arose in attempting to deprotonate the hydroxy ketone (now known to be **19**) at C10. Upon treatment of this substance with various bases, it underwent conversion to a new series of isomeric products. The minor product was the previously mentioned diosphenol **20b**. Other products were two isomeric hydroxy ketones, shown as **23a** and **23b**, in a 2:1 ratio. The structures of these compounds, which are isomers of **19**, have not been

(18) Kawada, K.; Gross, R. S.; Watt, D. S. *Synth. Commun.* **1989**, *5&6*, 777.

Scheme 8



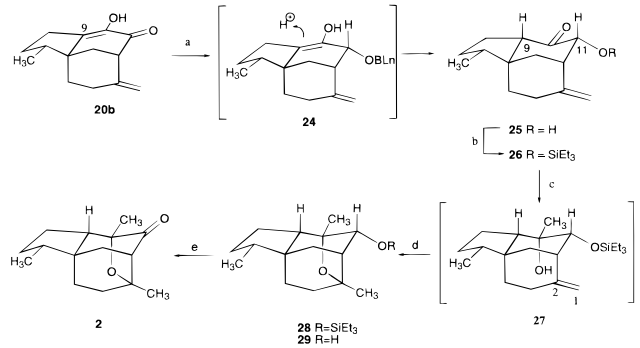
established. None of the original **19** could be detected in the reaction mixture. Indeed, when the solvent was carefully degassed, the oxidation step leading to the diosphenol could be substantially suppressed, resulting in the formation of hydroxyketones **23a** and **23b** in somewhat improved yields. Even within the context of the cis-fusion mode, permuting the ketones between C10 and C11 and the stereoecenter of the carbinol still allows for a pool of three isomeric ketols in addition to **19**. It was interesting to encounter the same mixture following attempted Birch reduction of **20b**. The most concise interpretation of this finding was that Birch reduction of **20b** had given a cis-fused enolate (cf. **22**) which, upon C-protonation, led to the same **23a/b** mixture as was available through treatment of **19** with strong bases.

Since the Birch reduction of **3** or **20b** had failed to provide access to the required trans hydrindanone arrangement, we turned to a new strategy for advancement. Presumably, existence of the C9−C10 enol in diosphenol **20b** is critically dependent on the presence of the C11 ketone. We supposed that reduction of this ketone (see intermediate **24**) would be followed very rapidly by ketonization of the C9−C10 enol. *Indeed, it would be this ketonization step that would establish the configuration at C9 and hence the junction modality.*

One could hardly be unmindful that the protonation step of the Birch reduction (see **3** → **16**) had occurred from the α-face of C9 to provide the cis junction. Similarly, Birch reduction of **20b** had also apparently occurred with exclusive formation of cis junction by means of proton attachment to the α-face of C9 (see discussion pertinent to Scheme 8 above). Nonetheless, we still felt that possibilities for reaching the trans fusion by *β*-protonation *during spontaneous ketonization of a C9−C10 enol* were reasonable. In the Birch reduction case, the stereochemistry at C9 arises from transition states where rehybridization of this center to the sp^3 state is well advanced prior to the protonation event. However, 1,2-reduction followed by ketonization, in contrast to the Birch reduction cases (cf. **3** and **20b**) would establish the stereochemistry at C9 during a kinetic protonation of the trigonal center of the enol (see **24**).

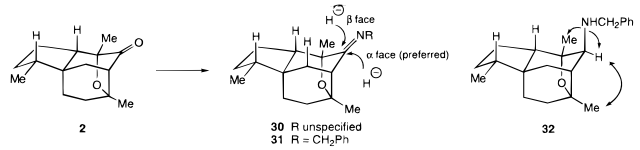
*In the event, treatment of **20b** with sodium borohydride led to the clean formation of a dihydroproduct which was, indeed, the desired **25** (see Scheme 9).* It was possible to preserve the keto group at C10 with rapid quenching of the reaction mixture

Scheme 9[a]



[a] Reagents and conditions: (a) $NaBH_4$, 30 s (84% based on recovered **20b**). (b) Et_3SiOTf, NEt_3 (93%). (c) MeMgI, Et_2O, 0 °C. (d) 1.0 N HCl in Et_2O, and then 5 N aqueous HCl (56% from **26**). (e) Jones reagent (92%).

Scheme 10



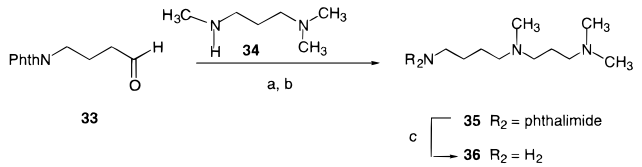
before over-reduction could occur. We note that **25** had not been encountered in the equilibration studies starting with **18** or **19** discussed above. The stereochemical assignment shown in **25** was supported by a 1D NOE experiment. Irradiation of the C9H enhanced the resonance of C11H, thereby establishing their cis relationship. While the stereochemistry at C11 was per se not important from an operational point of view (as it was necessary to convert this center to an amine), it was most helpful that a single stereoisomer was produced.

Our next goal was that of nucleophilic methylation at C10. The hydroxyl function of **25** was first protected (see triethylsilyl ether **26**). Treatment of **26** with methylmagnesium iodide led to the formation of a single compound, presumed to be **27** (cf. subgoal **21**). This presumption gained further support from the fact that this substance, in the presence of trace amounts of acid, gave rise to a saturated cyclic ether bearing a methyl group at C2 (see **28**). In practice, the etherification was promoted through the action of HCl (in ether) on **27**. Subsequent treatment with aqueous HCl resulted in desilylation to give **29**. Finally, oxidation of **29**, as shown, gave rise to ketone **2**.

Two possibilities were considered for reaching hispidospermidin from the compounds already in hand. One thought anticipated conversion of the alcohol function in **29**, to a suitable leaving group form (i.e., mesylate). At this point, the axial spermidine side chain would be installed by displacement of the leaving group from C11 with inversion of configuration. The alternative approach would start from **2**, which would be converted to the Schiff base of type **30** (Scheme 10). The latter, upon reduction, would hopefully produce the axial amine. We were well cognizant of the fact that in achieving such a result, it would be necessary for hydride delivery to occur from the α-face of the imine. While considering this requirement, it will be noted that the configurations at carbons 9 and 10 en route to these compounds had been established by *β*-face proton attachments to the convex surface of the bicyclo[3,3,1]nonane substructures.

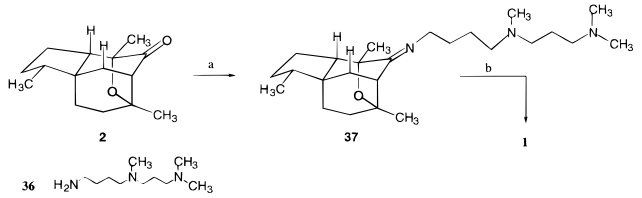
However, it was felt that a fundamental distinction in the context of the tetracyclic cage structure of type **30** could be exploited. In this latter system, in contrast to the tricyclic

Scheme 11[a]



[a] Reagents and conditions: (a) **34** (1.2 equiv), NaOAc/HOAc (pH 7), $NaCNBH_3$, MeOH (73% from **33**). (c) NH_2NH_2 (1 equiv), MeOH (81%).

Scheme 12[a]



[a] Reagents and conditions: (a) **36** (6.0 equiv), toluene, PPTS (cat.), reflux (Dean−Stark), 3 days. (b) $NaCNBH_3$, MeOH, pH 4 (60% from **2**).

structures bearing the bicyclo[3,3,1]nonane, α-face attack upon the imine would be occurring in a direction outside the environment of the hindered cagelike lattice. By contrast, *β*-face hydride attack on **30** could encounter abutments from the axial protons at carbons 9 and 13. Moreover, α-face attack could, in principle, benefit from a favorable interaction of the reducing agent with the ether oxygen of the cage.

We hoped to gain some early information as to the likelihood of success of the reductive amination strategy via a model system. Accordingly, ketone **2** was condensed with benzylamine, thereby giving rise to Schiff base **31**. Reduction of the latter with sodium borohydride in methanol in fact provided **32** bearing an axial benzylamino group. The stereochemistry in **32** was demonstrated by the NOE correlations depicted in Scheme 10.

Reassured by this finding, we then turned to the synthesis of the spermidine analogue **35** (see Scheme 11). This was accomplished, as indicated, starting with phthalimido butyraldehyde **33**.[19] Condensation of this aldehyde with diamine **34** under reductive conditions led to **35**.[20,21] Cleavage of the phthalamide function[22] exposed the required primary amine (see **36**).

In the final step, ketone **2** was condensed with triamine **36** (see Scheme 12). The crude Schiff base **37** was subjected to reduction with sodium cyanoborohydride. This reaction gave an 82% yield of DL-hispidospermidin. The identity of the fully synthetic hispidospermidin with natural material follows from the congruence of the proton (400 MHz), the ^{13}C NMR, and IR spectra of the synthetic racemate and those measured from an authentic specimen of **1** kindly provided by Nippon Roche. That hispidospermidin was reached through this route, of course, serves to independently corroborate the soundness of the assignments to all the intermediates along the synthesis route.

Though the total synthesis goal had been accomplished, there were still opportunities to explore interesting chemistry which

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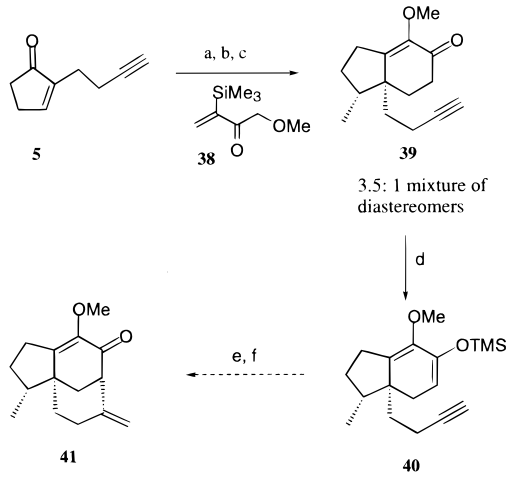
(20) Borch, R. F.; Hassid, A. I. *J. Org. Chem.* **1972**, *37*, 1673.
(21) Stevens, R. V.; Beaulieu, N.; Chan, W. H.; Daniewski, A. R.; Takeda, T.; Waldner, A.; Williard, P. G.; Zutter, U. *J. Am. Chem. Soc.* **1986**, *108*, 1039.
(22) Ford, H.; Chang, C.-H.; Behrman, E. J. *J. Am. Chem. Soc.* **1981**, *103*, 7773.

Scheme 13[a]



[a] Reagents and conditions: (a) Me_2CuLi, −78 °C. (b) **38** (2 equiv), −35 °C. (c) NeOH/4% KOH (4:1), reflux 18 h (30% from **5**). (d) LHMDS, NEt_3, TMSCl, −78 °C. (e) $HgCl_2$, HMDS. (f) NaI, 5.0 N HCl.

might well improve the route. In this regard, we sought to develop a more direct route to diosphenol **20b** by conducting the conjugate addition annulation sequence *with a trapping agent bearing the desired oxygen substituent* at the C10 position.

Based on literature precedents, Robinson annulation with methoxymethyl vinyl ketone can be conducted,[23] but no examples of conjugate addition followed by trapping with α-oxygenated methyl vinyl ketone equivalents were found. It seemed reasonable to attempt the annulation with vinyl ketone **38** (Scheme 12), the oxygenated analogue of our successful annulating agent. The synthesis was performed in a manner analogous to the parent case.[8b]

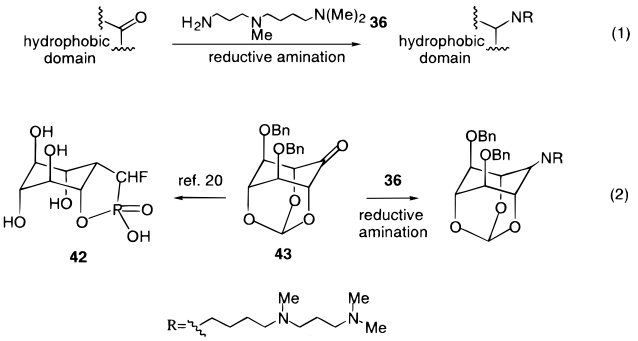
The Robinson annulation sequence was then performed as before (see Scheme 13), to good effect. The yield of bicyclic enone (∼30%) **39** was acceptable for the moment. However, the trapping of the annulating agent by the enolate was apparently significantly less selective (3.5:1; via **38** vs 8:1 via **6**).

The α-methoxyenone **39** was then converted to enol silane **40**. Unfortunately, treatment of this compound with mercury chloride did not lead to the desired tricyclic enone **41**. Apparently, the methoxy substituent of **40** interferes with the reactivity of the silyl enol ether in some way. It is not clear whether the effect is electronic, steric, or derived from some kind of unproductive sequestration of the mercury salt. This result was disappointing, but the success of the annulation was notable in that it appears to be the first example of the trapping of an α-oxygenated Stork-type annulating agent after conjugate addition.

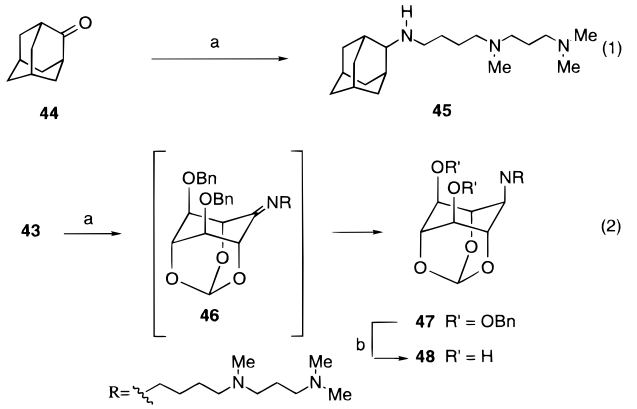
With the total synthesis of hispidospermidin accomplished, attentions turned to trying to gain at least a preliminary insight into the gross structural features that influence the mode of action of the agent. Already, assay results had shown that the spermidine side chain plays a key role in phospholipase C inhibition.[1a] We wondered about the role of the caged system in this regard. Thus, in the original report of Othsuka, spermidine itself (cf. the N-methylated spermidine present in **1**) was able to inhibit the action of PLC at an IC_{50} of 59 μM. Hence, we posed the question as to the consequence of attaching the N-methylated spermidine subunit to other types of carriers. It

(23) Wenkert, E.; Golob, N. F.; Sathe, S. S.; Smith, R. A. J. *Synth. Commun.* **1973**, 205 and references therein.

Scheme 14



Scheme 15[a]



[a] Reagents and conditions: (a) **36** (3.0 equiv), toluene, PPTS (cat.), reflux (Dean−Stark), 3 days, and then $NaCNBH_3$, MeOH, pH 4. (b) Na, NH_3, −33 °C.

was decided to probe two very different models in this regard. In one model, we hoped to supplant the caged domain of hispidospermidine with other caged hydrophobic moieties (Scheme 14, eq 1). The expectation would be to assemble such structures by reductive amination of appropriate ketones with **36** (cf. **2** → **1**).

In another initiative, we took note of the fact that cyclic phosphate **42**, derived from ketone **43**, is itself a weak (6.2 mM) inhibitor of phosphoinositol hydrolysis by PLC.[24] Moreover other PIP_2 analogues have been shown to inhibit the action of PLC, presumably by competing with the natural substrate.[25] Accordingly, we undertook to link ketone **43** to spermidine **36** to create a hybrid system containing spermidine and substrate "look-alike" domains (Scheme 14, eq 2).

In practice, reductive amination of commercially available adamantanone (**44**) with **36** provided **45** in 67% yield (Scheme 15, eq 1). In a parallel effort, reductive amination of ketone **43** with **36** provided a 77% yield of **47** as a single diastereomer. Reductive cleavage of the benzyl protecting groups was accomplished through the action of potassium in ammonia to afford **48** (Scheme 15, eq 2). We note that the stereochemical course of the imine reduction step (**43** → **47**) was not rigorously determined. However, in the light of the similar topology of imine intermediate **46** with that of **37**, we would expect the two benzyloxy groups to direct reduction of the imine syn to the *m*-dioxane moiety, leading to **46** and thence **48**.

Preliminary cytoxicity studies were performed with synthetic hispidospermidine, adamantyl analogue **45**, and myoinositol

(24) Campbell, A. S.; Thatcher, G. R. J. *Tetrahedron Lett.* **1991**, *32*, 2207.

(25) Ryan, M.; Smith, M. P.; Vinod, T. K.; Lau, W. L.; Keana, J. F. W.; Griffith, O. H. *J. Med. Chem.* **1996**, *39*, 4366.

look-alike **48** against a variety of cell lines. The extensive data so gathered will be published elsewhere in detail. We tested the antiproliferative effects of these compounds on a panel of human cancer cell lines. In TSU-Pr1 (prostate), A431 (epidermoid), Colo-205 (colon), and MDA MB-468 (mammary) cells, the adamntyl system **45** gave IC_{50} values that were weaker than those of **1** by factors ranging between 2 and 12. By contrast, the myoinositol analogue **48** was virtually inactive. The data with respect to the breast cancer cell line MCF7 were typical (IC_{50}: **1,** 6 μM; **36**, 60 μM; **45,** 18 μM; **48**, >300 μM. Thus, it appears that much of the activity arises from the spermidine domain. While the nature of the spermidine effect has been a long-term matter of discussion, it now seems likely that the effect is significantly enhanced by a suitably positioned caged hydrophobic domain. The values of these and related compounds prepared through straightforward synthesis as cytotoxic agents are currently being evaluated.

Summary

In summary, the total synthesis of racemic hispidospermidine (**1**) has been accomplished. Among the key steps were intramolecular carbomercuration (see formation of **3**) and reductive conversion of diosphenol **20b** to the required hydroxy ketone **25**, bearing the all critical trans junction stereochemistry connecting rings A and B. The ability to conduct this transformation to achieve stereochemical control at a late stage allowed us to exploit the cis-fused tricyclic structure **16** by a variant of the Robinson annulation (see formation of **4**). The conversion of **16** to the required diosphenol **20a** was readily achieved through Rubottom oxidation of **16**, followed by Swern oxidation of **19**.

Cyclization of the tertiary alcohol **27** to the unactivated exo methylene group is favored by the enforced proximity of the two functions. Finally, introduction of the spermidine side chain is accomplished by hydride addition from the concave surface of the constrained tetrahydrofuran concave ring, presumably due to interferences from the bridging cyclohexane ring (see formation of **32** and **1**).

The reductive amination of spermidine with ketones **38** and **44** (adamantanone) led to analogue probe structures **45** and **48**. Of these, the former, bearing a cagelike lipid structure linked to spermidine performed similarly to hispidospermidine at the level of IC_{50} values in a variety of cell lines. By contrast, the inositol analogue, **48**, bearing a much less hydrophobic carrier domain proved to be much less potent. These data suggest the possibility of discovering a new class of cytoxic agents through joining strategic polyamines to highly lipophilic structures, through the simple chemistry of reductive amination. Studies on these matters are continuing.

Experimental Section

Preparation of 1-[1,3]Dioxolan-2-yl-9-(trimethylsilanyl)non-8-yn-4-one (13). Magnesium turnings (8.23 g, 339.0 mmol) in THF (100 mL) were stirred under argon during the addition of 2-(2-bromoethyl)-1,3-dioxolane (52.5 g, 290.2 mmol) in THF (150 mL) via addition funnel. The addition was carried out at a rate such that the reaction remained at or below 35 °C (~9 h) and then cooled to −78 °C. Aldehyde **12**[24] (32.5 g, 193.5 mmol) in THF (150 mL) was added dropwise over 4 h to the solution of Grignard reagent **11**, and the reaction was allowed to warm to room temperature and stir 8 h. The reaction was quenched with aqueous NH_4Cl (50 mL), and the solvent was removed under reduced pressure. The residue was taken up in Et_2O (750 mL) and washed successively with NH_4Cl (200 mL) and brine (200 mL), dried over $MgSO_4$, filtered, and concentrated. The crude product was used directly in the next reaction.

Crude alcohol was dissolved in CH_2Cl_2 (1 L), and pyridinium dichromate (80.0 g, 213.0 mmol) was added in a single portion. The reaction mixture was stirred vigorously at room temperature for 48 h and then filtered through Celite, rinsing the flask and the cake of Celite repeatedly with Et_2O (6 × 50 mL). The filtrate was concentrated under reduced pressure. Flash chromatography (20% ethyl acetate/ hexane) gave ketoacetal **13** (40.0 g, 77% over two steps): IR 2940, 2880, 2160, 1705 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 4.91 (t, J = 4.3 Hz, 1H), 3.95 (t, J = 7.1 Hz, 2H), 3.85 (t, J = 7.1 Hz, 2H), 2.55 (ddd, J = 10.8, 7.3, 3.5 Hz, 4H), 2.25 (t, J = 6.9 Hz, 2H), 1.98 (ddd, J = 11.8, 7.5, 4.3 Hz, 2H), 1.78 (t, J = 7.1 Hz, 2H), 0.14 (s, 9H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 209.3, 106.3, 103.3, 85.3, 64.9, 64.8, 41.1, 36.5, 27.6, 22.4, 19.1, 0.1; high-resolution mass spectrum (EI) m/z calculated for $C_{14}H_{24}O_3Si$ 268.1495, found 268.1491.

Preparation of 2-(3-Butynyl)-2-cyclopenten-1-one (5) (via Keto Aldehyde 10). Ketoacetal **13** (40.0 g, 149.2 mmol) was dissolved in THF/3 N HCl (1:1, 600 mL) and stirred for 10 h. The solution was poured into Et_2O (600 mL) and separated layers. The aqueous layer was extracted with Et_2O (3 × 100 mL). The combined organics were washed with saturated aqueous $NaHCO_3$ (300 mL) and brine (300 mL), dried over $MgSO_4$, and filtered, and the solvent was removed under reduced pressure with care, due to the volatile nature of the product., to give keto aldehyde **10**, which was used in the next reaction without further purification.

Crude keto aldehyde **10** was dissolved in Et_2O (750 mL) and 1% aqueous NaOH (250 mL), and the resultant biphasic mixture was stirred vigorously at room temperature for 3 days. The reaction mixture was poured into a separatory funnel, and the layers were separated. The organic layer was washed with brine (200 mL), dried over $MgSO_4$, and filtered. The solvent was removed by distillation at atmospheric pressure to avoid loss of the volatile product. The residue was purified by flash chromatography (30% Et_2O/pentane) to give cyclopentenone **5** (9.0 g, 45% over two steps) which crystallized at −20 °C: IR 3275 (w), 2900, 1690 (s) cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.47 (s, 1H), 2.60 (m, 2H), 2.40 (m, 6H), 1.95 (s, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 208.7, 158.3, 143.5, 83.0, 68.7, 33.9, 26.1, 23.5, 16.4; high-resolution mass spectrum (EI) m/z calculated for $C_9H_{10}O$ 134.0732, found 134.0746.

Preparation of Bicyclic Enone 4.[11] A solution of copper iodide (3.5 g, 18.6 mmol) and molecular sieves was suspended in Et_2O (40 mL). This mixture was cooled to 0 °C, and methyllithium (1.6 M, 9.3 mL) was added via syringe. A yellow precipitate was formed. The reaction mixture was cooled to −78 °C, and cyclopentenone **5** (2.0 g, 14.9 mmol) in Et_2O (12 mL) was added via cannula. The flask was rinsed with an additional portion of Et_2O (5.0 mL) which was then added to the reaction mixture. The resultant solution was allowed to warm slowly to −30 °C, and silyl butenone **6**[10b] (4.2 g, 30.0 mmol) in Et_2O (20 mL) was added via syringe pump over 1 h (reaction was monitored by TLC; sometimes additional silyl butenone was required to complete the reaction). The reaction was stirred an additional 30 min while warming to −10 °C and was quenched with saturated aqueous NH_4Cl solution containing NH_4OH and adjusted to pH 8 (5 mL). The resultant solution was poured into an Erlenmeyer flask containing Et_2O (100 mL) and NH_4Cl/NH_4OH solution (100 mL) and stirred vigorously for 3 h. The layers were separated, and the organic layer was washed successively with NH_4Cl/NH_4OH solution (2 × 50 mL) and brine (50 mL), dried over $MgSO_4$, filtered, and concentrated. The crude product was used directly in the next reaction.

The crude diketone in MeOH (10 mL) was added dropwise via syringe to a solution of 4% aqueous KOH (50 mL) in methanol (200 mL). The resultant cloudy brown mixture was heated to reflux for 18 h, at which point the solution had become a clear red. The organic solvent was removed in vacuo, and the residue was diluted with EtOAc (150 mL). The layers were separated, and the aqueous layer was extracted with EtOAc (3 × 15 mL). The combined organic layers were washed with brine (50 mL), dried over Na_2SO_4, filtered, and concentrated. The residue was purified by flash chromatography on silica (0−20% ethyl acetate/hexanes) to give bicyclic enone **4** (1.3 g, 43% yield over two steps): IR 3280 (w), 2890 (w), 2060, 1625 (w) cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) 4.3:1 mixture of diastereomers δ 5.87 (s, 1H), 2.58−2.71 (m, 1H), 2.34−2.58 (m, 3H), 2.16−2.31 (m, 3H), 1.52−

2.04 (m, 7H), 0.88 and 1.07 (two pairs of doublets, $J = 7.0$ Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 198.9, 176.6, 123.0, 83.9, 68.8, 46.7, 46.5, 33.7, 33.2, 30.9, 29.6, 29.3, 15.9, 13.6; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{14}H_{18}O$ 202.1358, found 202.1355.

Preparation of Tricyclic Enone 3 (via Silyl Enol Ether 14). Enone **4** was dissolved in anhydrous benzene, and solvent was removed in vacuo (3 ×). To a solution of **4** (690 mg, 3.4 mmol) in 10 mL of anhydrous CH_2Cl_2 at −78 °C was added lithium hexamethyldisilazane (1.0 M in THF, 7.2 mL, 7.17 mmol) and triethylamine (1.0 g, 10.2 mmol). After stirring for 5 min at −78 °C, chlorotrimethylsilane (1.9 g, 10.2 mmol) was added via syringe. The resultant solution was warmed to 0 °C over 1 h, quenched with saturated aqueous $NaHCO_3$ (2 mL), and partitioned between EtOAc and saturated aqueous $NaHCO_3$ (50 mL each). The aqueous layer was extracted with EtOAc (2 × 30 mL), and the combined organic layers were washed with brine, dried over Na_2SO_4, filtered, and concentrated. The residue was dissolved in anhydrous benzene, and the solvent was removed in vacuo (3×) to give crude silyl enol ether **14**.

A solution of mercuric chloride (1.0 g, 3.8 mmol) and hexamethyldisilazane (0.4 mL, 1.9 mmol) in 10 mL of anhydrous CH_2Cl_2 at room temperature was warmed to 35 °C and stirred for 30 min. A solution of **14** in 2 mL of anhydrous CH_2Cl_2 was introduced all at once via cannula. The flask was rinsed with 0.5 mL of anhydrous CH_2Cl_2 and added to the reaction mixture. The resultant mixture was stirred at 35 °C for 0.5 h. The solution was cooled to 0 °C, and aqueous HCl (5.0 N, 2.5 mL) and NaI (1.53 g, 10.2 mmol) were added with vigorous stirring. After 0.5 h, the reaction mixture was warmed to 35 °C and stirred for 1 h. The mixture was quenched with solid $NaHCO_3$, stirred for 15 min, and then partitioned between CH_2Cl_2 and saturated aqueous $NaHCO_3$ (60 mL each). The aqueous layer was extracted with CH_2Cl_2 (2 × 40 mL), and the combined organic layers were washed with brine, dried over Na_2SO_4, filtered, and concentrated. Flash chromatography (10% ethyl acetate/hexanes) gave tricyclic enone **3** (600 mg, 87%): IR 3060 (w), 3020 (w), 2920 (s), 2860, 2840, 1660 (s), 1630 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 10:1 mixture of diastereomers δ 6.03 (s, 1H), 4.89 (s, 1H), 4.75 (s, 1H), 3.15 (s, 1H), 2.67 (m, 1H), 2.52 (m, 1H), 2.29 (m, 2H), 1.94 (m, 3H), 1.79 (m, 1H), 1.52 (m, 2H), 1.43 (m, 1H), 1.00 (d, 6.8 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 199.6, 176.5, 144.3, 123.4, 110.4, 53.8, 46.2, 43.8, 40.9, 29.6, 29.4, 28.5, 26.1, 12.7; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{14}H_{18}O$ 202.1358, found 202.1357.

Preparation of Ketone 16. Ammonia (50 mL) was condensed into a flame-dried three-necked flask at −78 °C. Lithium (150 mg, 21.7 mmol) was added in small pieces to the stirring ammonia. The solution was stirred vigorously until it became deep blue (∼15 min). A solution of tricyclic enone **3** (1.29 g, 6.4 mmol) and *tert*-butyl alcohol (520 mg, 7.0 mmol) in 15 mL of anhydrous ether was added via cannula. The resultant mixture was allowed to warm to −35 °C over 10 min and then stirred at −35 °C for 30 min. The lithium was quenched by the dropwise addition of isoprene (∼5 mL), indicated by the disappearance of the deep blue color. Solid ammonium chloride (∼500 mg) was added carefully, and the solution slowly warmed to room temperature, with evaporation of the ammonia. The residue was taken up in ether (150 mL), brine (60 mL) was added, and the layers were separated. The aqueous layer was extracted with ether (3 × 50 mL). The combined organic layers were dried over Na_2SO_4 and filtered, and the solvent was removed in vacuo. The residue was chromatographed on silica (10% ethyl acetate/hexanes) to give 800 mg of ketone **16** and 148 mg of recovered enone **3**, 72% yield combined: IR 3060 (w), 2920 (s), 2840, 1700 (s), 1640 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.82 (s, 1H), 4.80 (s, 1H), 3.00 (s, 1H), 2.61 (dd, $J = 16.1$, 6.2, 1H), 2.30 (m, 2H), 2.05 (m, 3H), 1.91 (dt, $J = 13.0$, 2.3, 1H), 1.73 (m, 3H), 1.44 (m, 2H), 1.28 (m, 2H), 0.96 (d, 7.0 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 215.0, 146.6, 110.6, 54.3, 54.2, 45.7, 45.4, 40.7, 37.4, 34.6, 34.3, 33.2, 29.2, 14.4; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{14}H_{20}O$ 204.1514, found 204.1520.

Preparation of α-Hydroxyketone 19 (via Silyl Enol Ether 17). To ketone **16** (725 mg, 3.6 mmol) in 35 mL of CH_2Cl_2 at −78 °C was added triethylamine (1.1 g, 10.7 mmol) followed by triethylsilyltrifluoromethanesulfonate (1.4 g, 5.4 mmol). The resultant solution was warmed to room temperature over ∼2 h. The reaction mixture was poured into saturated aqueous $NaHCO_3$ (50 mL) and extracted with CH_2Cl_2 (3 × 30 mL). The combined organic layers were washed with brine (40 mL), dried over Na_2SO_4, filtered, and concentrated. The crude enol ether **17** was used directly in the next reaction.

A solution of silyl enol ether **17** (3.55 mmol) in CH_2Cl_2 (40 mL) containing solid $NaHCO_3$ (2.5 g) was cooled to −30 °C. *m*-CPBA (715 mg, 4.14 mmol) was added, and the reaction mixture was allowed to warm to 0 °C over 1 h. Pentane (80 mL) was added, and the solution was filtered. The filtrate was concentrated in vacuo to ∼5 mL of solvent, which was diluted with THF (20 mL). The resultant solution was cooled to 0 °C, TBAF (7.1 mL of 1.0 M in THF, 2.0 equiv) was added, and the reaction stirred for 1 h. The reaction was diluted with CH_2Cl_2 (75 mL) and poured into saturated aqueous $NaHCO_3$. The layers were separated, and the aqueous layer was extracted with CH_2Cl_2 (2 × 30 mL). The combined organics were washed with brine (40 mL), dried over Na_2SO_4, filtered, and concentrated. The residue was chromatographed on silica (10% ethyl acetate/hexanes) to give 620 mg (79%) of the α-hydroxyketone **19**: IR 3460 (br), 3050 (w) 290 (s), 2850 (s), 1700 (s), 1625 (w) cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 8:1 mixture of diastereomers; δ 4.88 (d, $J = 1.5$ Hz, 1H), 4.86 (d, $J = 1.5$ Hz, 1H), 4.63 (dd, $J = 8.3$, 3.1 Hz, 1H), 3.52 (d, $J = 3.1$ Hz, 1H), 3.39 (t, $J = 3.2$ Hz, 1H), 2.81 (ddd, $J = 19.6$, 8.3, 2.2 Hz, 1H), 2.62 (m, 2H), 2.06 (m, 2H), 1.88 (m, 2H), 1.68 (m, 2H), 1.42 (dt, $J = 13.6$, 2.5 Hz, 1H), 1.38 (m, 1H), 1.22 (m, 1H), 0.91 (d, $J = 7.3$ Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 211.4, 142.2, 113.7, 74.3, 56.0, 50.7, 44.2, 43.2, 40.0, 32.6, 30.6, 29.9, 24.7, 18.1; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{14}H_{20}O_2$ 220.1463, found 220.1468.

Preparation of Diosphenol 20b. A solution of methyl sulfoxide (430 μL, 8.2 mmol) in CH_2Cl_2 (25 mL) was cooled to −78 °C. Trifluoroacetic anhydride (850 μL, 6.0 mmol) was added via syringe, and the resultant mixture was stirred for 2 h at −78 °C. α-Hydroxyketone **19** (620 mg, 2.82 mmol) in CH_2Cl_2 (3 mL) was added via cannula, and the reaction was stirred an additional 0.5 h at −78 °C. Triethylamine (2.9 mL, 21.0 mmol) was added via syringe, and the reaction was allowed to warm to 0 °C over 1 h. The reaction was diluted with CH_2Cl_2 (45 mL), washed with water (25 mL), saturated aqueous $NaHCO_3$ (25 mL), and brine (25 mL), dried over Na_2SO_4, filtered, and concentrated. The residue was chromatographed on silica (10% ethyl acetate/hexanes) to give 488 mg (80%) of diosphenol **20b**: IR 3380, 3060 (w), 2920 (s), 2850, 1710, 1670, 1640 (s) cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$); 8:1 mixture of diastereomers δ 5.85 (s, 1H), 4.92 (s, 1H), 4.80 (s, 1H), 3.27 (m, 1H), 2.60 (m, 2H), 2.32 (m, 2H), 2.04−1.92 (m, 3H), 1.80 (m, 1H), 1.62−1.41 (m, 3H), 0.97 (d, $J = 5.9$ Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 194.7, 143.8, 143.2, 141.9, 111.0, 52.6, 46.2, 44.8, 30.6, 28.3, 26.3, 25.4, 12.5; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{14}H_{18}O_2$ 218.1307, found 218.1305.

Preparation of α-Hydroxyketone 25. A solution of diosphenol **20b** (170 mg, 0.78 mmol) in 8 mL of anhydrous methanol was cooled to 0 °C. $NaBH_4$ (30 mg, 0.78 mmol) was added in one portion. The reaction mixture was allowed to stir 30 s and then poured into 1 N HCl (20 mL). The quenched reaction was extracted with methylene chloride (3 × 15 mL), and the combined organic layers were washed with brine (15 mL), dried over Na_2SO_4, filtered, and concentrated. Flash chromatography (10% ethyl acetate/hexane) gave diosphenol **20b** (45 mg, 26%) and α-hydroxyketone **25** (100 mg, 58%): IR 3460 (br), 3050 (w), 2920 (s), 2869 (s), 1700 (s), 1680 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.83 (m, 2H), 4.24 (d, $J = 6.1$ Hz, 1H), 3.52 (s, 1H), 3.27 (m, 1H), 2.53 (apparent t, $J = 10$ Hz, 1H) 2.14 (dd, $J = 15.7$, 5.9 Hz, 1H), 2.02−1.72 (series of m, 7H), 1.5 (m, 1H), 1.36 (m, 2H), 0.89 (d, $J = 6.8$ Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 212.0, 146.8, 111.0, 167.2, 50.3 (2C), 50.1, 44.6, 39.3, 29.8, 27.8, 26.0, 19.1, 13.7; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{14}H_{20}O_2$, 220.1463, found 220.1467.

Preparation of α-Silyloxyketone 26. A solution of α-hydroxyketone **25** (240 mg, 1.1 mmol) in 10 mL of anhydrous CH_2Cl_2 was cooled to −20 °C. Triethylamine (456 μL, 3.3 mmol) and triethyltrifluoromethanesulfonate (373 μL, 1.65 mmol) were added sequentially via syringe. After 0.5 h, the reaction was diluted with CH_2Cl_2 (40 mL) and washed with H_2O (20 mL), saturated aqueous $NaHCO_3$ (20 mL), and brine (20 mL). The organic layer was dried over Na_2SO_4, filtered, and concentrated. Flash chromatography (5% ethyl acetate/hexane) gave

α-silyloxyketone **26** (326 mg, 89%): IR 3040 (w), 2900 (br), 2869 (s), 1700 (s), 1680 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 4.72 (s, 1H), 4.67 (s, 1H), 4.26 (d, 5.9 Hz, 1H), 3.02 (m, 1H), 2.41 (apparent t, J = 10 Hz, 1H) 2.09 (dd, J = 15.4, 6.1 Hz, 1H), 2.03−1.84 (m, 4H), 1.77−1.68 (m, 3H), 1.47 (m, 1H), 1.33 (m, 2H), 0.96 (t, J = 7.9 Hz, 9 H), 0.86 (d, J = 6.9 Hz, 3H), 0.63 (m, 6H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 210.2, 147.5, 110.1, 109.9, 109.7, 77.9, 59.5, 52.2, 52.1, 49.2, 44.7, 40.0, 39.9, 29.9, 28.0, 26.1, 19.3, 13.8, 6.8, 5.0; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{20}H_{34}O_2SiK$ 373.1965, found 373.1974.

Preparation of Alcohol 29. To a solution of α-silyloxyketone **26** (62.0 mg, 0.19 mmol) in 2 mL of anhydrous THF at room temperature was added methylmagnesium iodide (3.0 M in Et_2O, 122 μL, 1.9 mmol). After 5 min the reaction was complete. The solution was treated with anhydrous HCl (1.0 M in Et_2O, 5 mL) and stirred for 0.5 h. Aqueous HCl (5.0 M, 0.5 mL) was added, and the reaction was stirred an additional 5 h. The solution was partitioned between ether and water (15 mL each), and the aqueous layer was extracted with ether (2 × 10 mL). The combined organic layers were washed with saturated aqueous $NaHCO_3$ and brine (15 mL each), dried over anhydrous $MgSO_4$, filtered, and concentrated. Flash chromatography (20% ethyl acetate/ hexane) gave 29 mg (56%) of alcohol **29** as a white solid: IR 3380 (br), 2920 (s), 2840 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 3.65 (s, 1H), 2.19 (d, J = 5.5 Hz, 1H), 1.90−1.60 (series of m, 5H), 1.50 (m, 4H) 1.38 (s, 3H), 1.28 (m, 2H), 1.21 (s, 3H), 1.08 (d, J = 12.5 Hz, 1H), 0.84 (d, J = 6.7 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 85.2, 83.9, 82.6, 59.0, 50.2, 43.6, 43.2, 33.5, 32.3, 30.3, 29.2, 20.5, 17.9, 16.3, 14.1; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{15}H_{24}O_2$; 236.1781, found 236.1779.

Preparation of *N*-[3-(Dimethylamino)propyl]-*N*-methyl-1,4-butanediamine (36). *N*,*N*′,*N*′-Trimethylethylenediamine **34** (1.39 g, 12.0 mmol), NaOAc trihydrate (1.30 g, 15.8 mmol), and $NaCNBH_3$ (0.90 mg, 14.3 mmol) were dissolved in MeOH (50 mL). Aldehyde **33**[19] (2.17 g, 10.0 mmol) was added to the reaction mixture. The resultant solution was adjusted to pH 7 with HOAc and stirred for 24 h at room temperature. Acetone (5 mL) was added followed by 5 N HCl until the pH was 1−2. The solvent was removed under reduced pressure, and the residue was taken up in H_2O (15 mL) and washed with Et_2O (3 × 4 mL). The aqueous layer was adjusted to pH 8.5 with 1 N NaOH and extracted once with ether. The aqueous layer was then extracted with CH_2Cl_2 (6 × 5 mL), checking the pH after each extraction and adjusting to pH 8.5 if necessary. Solid NaCl was added to the aqueous layer and it was extracted again with CH_2Cl_2 (2 × 5 mL). The combined CH_2Cl_2 extracts were dried over $NaSO_4$, filtered, and concentrated to give pure amine **35** (2.29 g, 73%), which was used directly in the next reaction. The spectral data for this material were identical to that previously reported for this compound, prepared by an alternative method.[22]

Dephthaloylation of amine **35** was performed as described in the literature[22] to give title amine **36** (81%). The spectral data of the product were identical to that previously reported for amine **36**.[22]

Preparation of Ketone 2. Alcohol **29** (107 mg, 0.45 mmol) in acetone (4.5 mL) was cooled to 0 °C, and Jones reagent was added until the solution remained red. The resultant solution was stirred at 0 °C for 2 h, with Jones reagent being added whenever necessary to maintain a red solution. When the starting material was consumed (monitored by thin-layer chromatography), 2-propanol was added dropwise to consume excess oxidizing reagent (indicated by a change from red to green solution). The solution was diluted with EtOAc (40 mL), washed with brine (10 mL), dried over Na_2SO_4, filtered, and concentrated. Flash chromatography (5% ethyl acetate/hexane) gave the ketone **2** (97 mg, 92%): IR 2930 (s), 2840, 1750 (s) cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 2.25 (d, J = 5.3 Hz, 1H), 2.02 (dd, J = 12.9, 5.8 Hz, 1H), 1.94−1.82 (m, 2H), 1.77−1.30 (series of m, 9H), 1.23 (s, 3H), 1.15 (s, 3H), 0.85 (d, J = 6.7 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 215.2, 81.7, 77.7, 62.6, 50.8, 43.5, 42.2, 35.9, 31.3, 30.7, 28.0, 19.7, 18.2, 15.2, 14.2; high-resolution mass spectrum (EI) *m*/*z* calculated for $C_{15}H_{22}O_2$, 234.1620, found 234.1610.

Preparation of Hispidospermidin (1). Representative Procedure for Reductive Amination. A solution of ketone **2** (97 mg, 0.41 mmol), triamine **36** (228 mg, 1.23 mmol), and pyridinium *p*-toluenesulfonate (5 mg, 0.02 mmol) in anhydrous toluene (4.5 mL) was heated to reflux under a Dean−Stark trap filled with 4-Å molecular sieves. After 3 days, the toluene was removed under reduced pressure. The residue (crude **37**) was dissolved in anhydrous methanol (4.5 mL), and $NaCNBH_3$ (52.0 mg, 0.82 mmol) was added. The solution was adjusted to pH 6 with 1 N HCl in ether; it became cloudy at this point. After stirring for 36 h at room temperature, concentrated HCl was added to quench excess reducing agent. The methanol was removed under reduced pressure, and the residue was dissolved in H_2O (6 mL). It was washed with CH_2Cl_2 (3 × 2 mL), and then the aqueous layer was adjusted to pH 10 with a NaOH pellet. The aqueous layer was extracted with CH_2Cl_2 (4 × 3 mL), and the combined organic layers were dried over Na_2SO_4 and filtered, and the solvent was removed under reduced pressure. Flash chromatography (10% NH_3 in MeOH/CH_2Cl_2) gave **1** (100 mg, 60% from ketone). Identity was established on the basis of comparison of 400-MHz ^1H NMR, 75-MHz ^{13}C NMR, IR, and high-resolution mass spectral data with an authentic sample of hispidospermidin.

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Supporting Information Available: Experimental data of selected intermediates (**9**, **10**, **23a**,**b**, **32**, **38**, **39**, **45**, **47**, **48**). This material is available free of charge via the Internet at http://pubs.acs.org.

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