1871–1873; b) N. Yoshikawa, Y. M. A. Yamada, J. Das, H. Sasai, M. Shibasaki, *J. Am. Chem. Soc.* **1999**, *121*, 4168–4178.

[7] The absolute configuration was determined by Mosher's method; see J. A. Dale, H. S. Mosher, *J. Am. Chem. Soc.* **1973**, *95*, 512–519. The enantiomeric excess was determined by HPLC analysis using DAICEL CHIRALPAK AD (2-propanol/hexane 1/100).

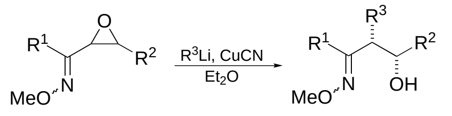
[8] a) T. Mandai, T. Matsumoto, M. Kawada, J. Tsuji, *Tetrahedron Lett.* **1993**, *34*, 2161–2164; b) J. Tsuji, T. Sugiura, I. Minami, *Synthesis* **1987**, 603–606.

[9] a) K. Harada, H. Urabe, F. Sato, *Tetrahedron Lett.* **1995**, *36*, 3203–3206; b) F. Sato, H. Ishikawa, M. Sato, *Tetrahedron Lett.* **1981**, *22*, 85–88. This method could lead to the fragment for the synthesis of epothilone B (**2**).

[10] For related work with oximes, see E. J. Corey, L. S. Melvin, Jr., M. F. Haslanger, *Tetrahedron Lett.* **1975**, 3117–3120.

[11] We can also prepare *syn*-aldol from *cis*-enone by this method.[13] Through coupling with the stereocontrolled catalytic asymmetric epoxidation of the *cis*- and *trans*-enones recently developed in our group,[5] it is possible to prepare *syn*- and *anti*-aldol from *cis*- and *trans*-enones, respectively, with excellent *ee* values (Table 1).

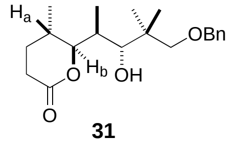
Table 1. Epoxide opening of *cis*-epoxy-oximes by cuprate reagents.



Entry	R^1	R^2	R^3	Yield [%]
1	methyl	pentyl	methyl	77
2	propyl	pentyl	methyl	72
3	phenethyl	pentyl	methyl	85
4	phenethyl	pentyl	butyl	80
5	phenethyl	pentyl	phenyl	80
6	phenethyl	pentyl	vinyl	50
7	pentyl	phenethyl	methyl	81

[12] The relative configuration of the alcohol resulting from reduction of **21** with Me$_4$NBH(OAc)$_3$ was determined by analyzing the NOE of **22**, and the relative configuration of the methyl group of **22** resulting from allylation of **20** was further determined by transforming **22** into **31** and analyzing the coupling constant between H$_a$ and H$_b$ (10.2 Hz).

[13] The absolute configurations were determined by Mosher's method,[8] and the enantiomeric excesses were determined by HPLC analysis using DAICEL CHIRALPAK AD (2-propanol/hexane 1/100). The relative configurations were determined by transforming **24** into **32** and **25** into **33** (cyclization of **27** and **34**, respectively), and analyzing their NOEs.



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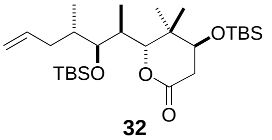

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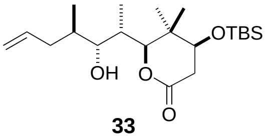

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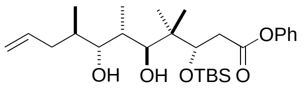

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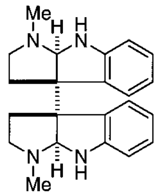
[14] R. Göttlich, K. Yamakoshi, H. Sasai, M. Shibasaki, *Synlett* **1997**, 971–973.

Enantioselective Construction of Vicinal Stereogenic Quaternary Centers by Dialkylation: Practical Total Syntheses of (+)- and *meso*-Chimonanthine**

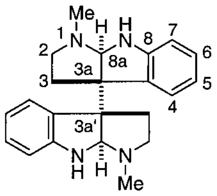
Larry E. Overman,* Jay F. Larrow, Brian A. Stearns, and Jennifer M. Vance

Enantioselective formation of vicinal stereogenic quaternary carbon centers is one of the more formidable challenges in contemporary organic synthesis.[1,2] Here we report a powerful method for creating such arrays by reaction of achiral dienolates with a chiral, enantiopure dielectrophile. The method was developed to address the vicinal stereogenic quaternary benzylic carbon centers that are the signature structural feature of indole alkaloids containing the hexacyclic 3a,3a′-bispyrrolidino[2,3-*b*]indoline unit.

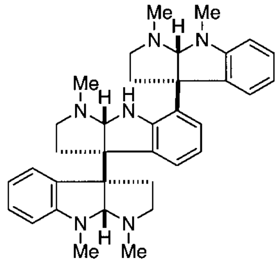
The chimonanthines are the simplest members of this family and are found in nature in all three stereochemical motifs: *meso*-chimonanthine (**1**)[3] and (−)-chimonanthine[4] from plant sources; (+)-chimonanthine (**2**) from the skin of a Colombian frog[5] and from plant sources.[6] In addition, many



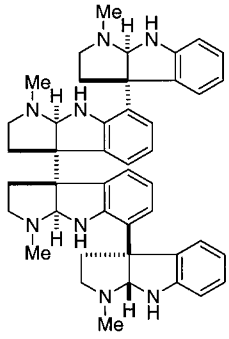
1: *meso*-chimonanthine



2: (+)-chimonanthine



3: (−)-idiospermuline



4: quadrigemine C

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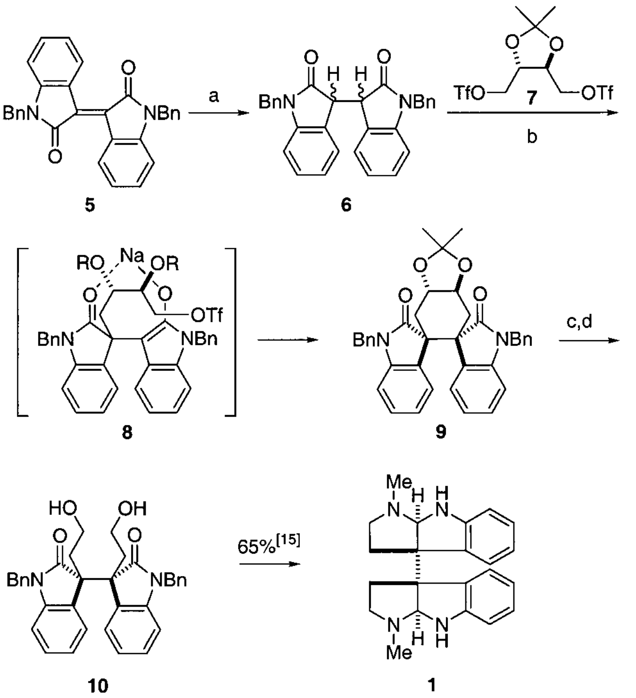
Supporting information for this article is available on the WWW under http://www.wiley-vch.de/home/angewandte/ or from the author.

more complex indole alkaloids contain the 3a,3a′-bispyrrolidino[2,3-*b*]indoline moiety. For example, (−)-idiospermuline (**3**) consists of a (−)-chimonanthine fragment functionalized with one additional pyrrolidinoindoline unit,[7] while the dodecacyclic alkaloid quadrigemine C (**4**) is proposed to have a *meso*-chimonanthine core functionalized with two pyrrolidinoindoline units of the same absolute stereochemistry.[8]

Early synthetic efforts directed towards this class of indole alkaloids produced largely the racemic C_2-symmetric isomers by oxidative dimerization of indole[9] or oxindole derivatives,[10] or by dialkylation of dihydroisoindigo.[11] We have reported a stereoselective approach to *meso*-bispyrrolidinoindoline alkaloids by samarium-mediated reductive dialkylation of an isoindigo,[12] and more recently a flexible approach to either the *meso* or C_2 isomers using an intramolecular Heck reaction cascade was presented.[13] Herein we disclose a highly efficient synthesis of 3a,3a′-bispyrrolidino[2,3-*b*]indolines having either *meso* or C_2 symmetry. The approach utilizes the nature of a metal dienolate (chelated or nonchelated) to control the relative configuration, while exploiting the chirality of a tartrate-derived dielectrophile to control absolute stereochemistry.

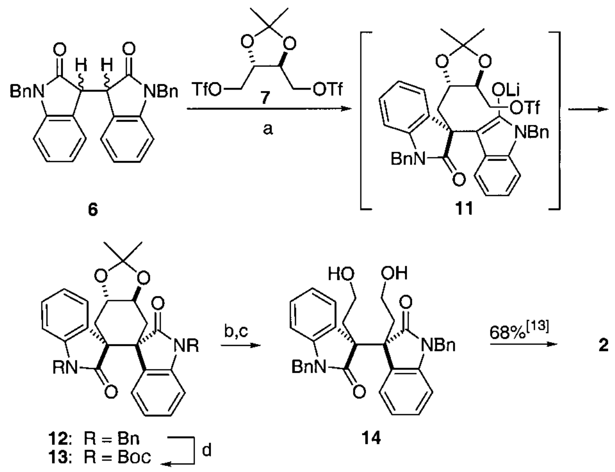
Isoindigo **5**,[12] which is available in two steps and 85 % yield from commercially available oxindole and isatin, was converted into the corresponding dihydroisoindigo **6** by reduction with zinc in acetic acid at room temperature (Scheme 1). Alkylation of the disodium dienolate of **6** with 1.1 equiv of ditriflate **7**[14] in THF at −78 °C provided in 92 % yield a single pentacyclic product **9**, having the *cis* relationship of the two spirooxindole groups. The acetonide was removed by exposure of **9** to camphorsulfonic acid (CSA) and MeOH to liberate the cyclohexanediol, which was cleaved with $Pb(OAc)_4$, and the resulting labile dialdehyde was reduced with $NaBH_4$ to provide diol **10**. This intermediate could be converted into *meso*-chimonanthine (**1**) in six steps and 65 % overall yield.[12, 15]



Scheme 1. a) Zn, AcOH, room temperature, 83 %; b) NaHMDS (2.1 equiv), THF, −78 °C, 92 %; c) CSA, $MeOH/CH_2Cl_2$, 100 %; d) $Pb(OAc)_4$, PhH; $NaBH_4$, MeOH, 92 %. Bn = benzyl, HMDS = 1,1,1,3,3,3-hexamethyldisilazane, OTf = trifluoromethansulfonate (triflate),

The critical dialkylation step of the approach to **1** summarized in Scheme 1 is more amenable to scale-up than the related step of our earlier route.[12] Use of the tartrate-derived dielectrophile **7** avoids problems with competing S_N2' cyclization that are seen in the reaction of the disodium dienolate of **6** with *cis*-1,4-dichloro-2-butene.[12] Stereoselection in the second alkylation step is consistent with preferential reaction of chelated intermediate **8**.[12, 16]

The outcome of the dialkylation reaction was dramatically altered by changing the dienolate counter ion and reaction solvent (Scheme 2). When the reaction of **6** with ditriflate **7** was carried out with LiHMDS (2.1 equiv) in THF containing 10 % DMPU at −78 °C, the major product formed was the C_2-symmetric bisoxindole **12**, which could be isolated in 55 % yield. Under these conditions, the other possible C_2 product was generated in trace amounts only (1–3 %), while isomer **9** having the spirooxindole groups *cis* was formed in 20 % yield. Both of these minor isomers were easily removed from **12** by chromatography.



Scheme 2. a) LiHMDS (2.1 equiv), THF/DMPU, −78 °C, 55 %; b) CSA, $MeOH/CH_2Cl_2$, 100 %; c) $Pb(OAc)_4$, PhH; $NaBH_4$, MeOH, 90 %; d) *t*BuLi, Et_2O, −78 °C then O_2; Boc_2O, DMAP, CH_2Cl_2, 61 %. Boc = *tert*-butoxycarbonyl, DMAP = 4-dimethylaminopyridine, DMPU = 1,3-dimethylhexahydro-2-pyrimidinone.

The relative stereochemistry of **12** was established by removing the benzyl groups with *tert*-butyllithium/oxygen[17] followed by *N*-acylation with di-*tert*-butyl dicarbonate to provide BOC derivative **13**. Single-crystal X-ray analysis of this derivative established the relative stereochemistry of **12**.[18] Using the same sequence employed to elaborate **9**, C_2-symmetric bisoxindole **12** was converted in two steps and 90 % overall yield into **14** ($[\alpha]_D^{23} = +235$, $c = 0.9$ in $CHCl_3$). This intermediate was processed in six additional steps to complete the first enantioselective total synthesis of (+)-chimonanthine (**2**; 68 % overall yield; $[\alpha]_D^{23} = +274$, $c = 0.5$ in EtOH).[13, 19]

In summary, the dialkylation approach described herein constitutes a practical way to prepare either the *meso* or enantiopure C_2 stereoisomers of 3a,3a′-bispyrrolidino[2,3-*b*]-indolines on large scales. Starting from commercially available oxindole and isatin, the total syntheses of *meso*- (**1**) and (+)-chimonanthine (**2**) were accomplished in 39 and 21% overall yield respectively. The alkylation of the dilithium dienolate of **6** in 10% DMPU/THF is notable in producing one of the two possible C_2 products with high selectivity (20:1). This alkylation reaction is a rare example of high diastereoselectivity arising from the combination a prostereogenic enolate with a chiral electrophile containing an sp^3 carbon center.[20]

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[1] Recent reviews of asymmetric synthesis of quaternary centers: a) E. J. Corey, A. Guzman-Perez, *Angew. Chem.* **1998**, *110*, 402–415; *Angew. Chem. Int. Ed.* **1998**, *37*, 388–401; b) K. Fuji, *Chem. Rev.* **1993**, *93*, 2037–2066.

[2] For an excellent recent strategy, see R. M. Lemieux, A. I. Meyers, *J. Am. Chem. Soc.* **1998**, *120*, 5453–5457.

[3] Y. Adjibadé, B. Weniger, J. C. Quirion, B. Kuballa, P. Cabalion, R. Anton, *Phytochemistry* **1992**, *31*, 317–319.

[4] H. F. Hodson, B. Robinson, G. F. Smith, *Proc. Chem. Soc. London* **1961**, 465–466.

[5] T. Tokuyama, J. W. Daly, *Tetrahedron* **1983**, *39*, 41–47.

[6] L. Verotta, T. Pilati, M. Tatò, E. Elisabetsky, T. A. Amador, D. S. Nunes, *J. Nat. Prod.* **1998**, *61*, 392–396.

[7] R. K. Duke, R. D. Allan, G. A. R. Johnston, K. N. Mewett, A. D. Mitrovic, C. C. Duke, T. W. Hambley, *J. Nat. Prod.* **1995**, *58*, 1200–1208.

[8] F. Guéritte-Voegelein, T. Sévenet, J. Pusset, M.-T. Adeline, B. Gillet, J.-C. Beloeil, D. Guénard, P. Potier, R. Rasolonjanahary, C. Kordon, *J. Nat. Prod.* **1992**, *55*, 923–930.

[9] E. S. Hall, F. McCapra, A. I. Scott, *Tetrahedron* **1967**, *23*, 4131–4141.

[10] a) R. Göschke, J. B. Hendrickson, R. Rees, *Tetrahedron* **1964**, *20*, 565–579; b) C. L. Fang, S. Horne, N. Taylor, R. Rodrigo, *J. Am. Chem. Soc.* **1994**, *116*, 9480–9486.

[11] T. Hino, S. Yamada, *Tetrahedron Lett.* **1963**, 1757–1760.

[12] J. T. Link, L. E. Overman, *J. Am. Chem. Soc.* **1996**, *118*, 8166–8167.

[13] L. E. Overman, D. V. Paone, B. A. Stearns, *J. Am. Chem. Soc.* **1999**, *121*, 7702–7703.

[14] Ditriflate **7** is available on a large scale by reaction of (+)-2,3-*O*-isopropylidene-L-threitol with triflic anhydride and iPr$_2$NEt in Et$_2$O at 0°C. Both enantiomers of this intermediate are available from (+)- and (−)-tartaric acid: E. A. Mash, K. A. Nelson, S. Van Deusen, S. B. Hemperly in *Organic Syntheses, Vol. 68* (Ed.: J. D. White), Wiley, New York, **1990**, pp. 92–103.

[15] Diol **10** had previously been converted into *meso*-chimonanthine.[12] The efficiency of this conversion is increased to 65% by utilizing the conditions we recently developed to convert a related chiral diol into (−)-chimonanthine.[13]

[16] Consistent with this interpretation, stereoselection was eroded in the presence of a sodium-selective cryptand (Kryptofix 221).

[17] R. M. Williams, E. Kwast, *Tetrahedron Lett.* **1989**, *30*, 451–454.

[18] Crystallographic data (excluding structure factors) for the structure reported in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication no. CCDC-134670. Copies of the data can be obtained free of charge on application to CCDC, 12 Union Road, Cambridge CB2 1EZ, UK (fax: (+44) 1223-336-033; e-mail: deposit@ccdc.cam.ac.uk).

[19] Literature value:[6] $[\alpha]_{\mathrm{D}}^{23} = +264.5$ ($c = 1.0$ in EtOH).

[20] a) A recent survey of stereoselective C–C bond formation cites no examples in sections dealing with alkylation reactions with chiral electrophiles; see D. S. Matteson, *Methoden Org. Chem. (Houben-Weyl) 4th ed. 1996*, Vol. E21/2, pp. 1077–1118; b) Interconversion of atropisomers of the dienolates derived from **6** is likely to be slow under the alkylation conditions. Whether the 20:1 selectivity arises solely in the initial alkylation step or is enhanced by selective partitioning of the minor diastereomer in the cyclization step has not yet been determined.

How Strong Is the Coordination Bond between a Histidine Tag and Ni–Nitrilotriacetate? An Experiment of Mechanochemistry on Single Molecules**

Matteo Conti, Giuseppe Falini, and Bruno Samorì*

A number of extensively used methods of increasing importance in molecular biology are based on the ability of the nickel(II) ion, when chelated by nitrilotriacetate (nta), to selectively bind proteins containing stretches of consecutive histidine residues (His). Hochuli et al. established this field of applications when they discovered that proteins containing isolated histidines lead to complexes that are less stable than those arising from proteins having two consecutive His tags at one terminus (2 × His-tag).[1] Indeed, a stretch of six consecutive His tags (6 × His-tag) is now commonly appended to the primary sequence of recombinant proteins, making it possible to isolate them selectively from a flow of crude cell lysate by means of a Ni–nta-functionalized chromatographic matrix[2a,b] and to immobilize them on biosensors for biomolecular interaction analysis.[3] The stability of the anchoring of the His-tagged proteins is challenged in both cases by the frictional force exerted on them by the flow. Such interplay between external forces and chemical processes can now be transferred to the single-molecule level thanks to recent developments in nanoscale manipulation techniques.[4, 5] Single-molecule experiments can give crucial insights into binding processes that cannot be accessed by solution methods. In the single-molecule world, bond making and breaking becomes a digital "yes-or-no" process. Determination of binding constants by traditional means provides only information that is averaged on the entire ensemble of the molecules and on the overall time scale of the experiment.

We simulated the process involved in the Ni–nta protein separation techniques by reproducing the approach, binding, stretching, and unbinding under external forces between individual His tags and individual Ni–nta chelating groups in a single-molecule experiment with the scanning force microscope (SFM, see Figure 1).[6] This experiment revealed that: 1) the probability that the encounters between a 6 × His-tag and a Ni–nta group develop in a stable bond is much higher than that for a 2 × His-tag; 2) both tags can make either of two types of complex with markedly different stabilities and with different "energy landscapes" along their force-driven disso-

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