Combinatorial Chemistry

Stereocontrolled Solid-Phase Synthesis of a 90-Membered Library of Indoline-Alkaloid-like Polycycles from an Enantioenriched Aminoindoline Scaffold**

*Zhonghong Gan, P. Thirupathi Reddy, Sophie Quevillon, Samuel Couve-Bonnaire, and Prabhat Arya**

With growing interest in the use of small molecules[1] for dissecting protein–protein interactions[2] and for understanding signaling pathways, the need for developing combinatorial methods to obtain small molecules that have stereochemical and skeletal diversity has also grown.[3,4] Owing to their structural complexity and the diversity of their functional groups, natural products are a source of bioactive lead compounds, and it is highly likely that libraries of small molecules that also display these features would serve as valuable tools.[5]

We initiated a combinatorial chemistry program that is aimed at providing indoline-alkaloid-like complex polycyclic compounds in a high-throughput manner.[6] Indole and indoline alkaloids belong to an important family of bioactive natural products, and several of these derivatives (**1**–**3**, see Figure 1) exhibit various biological responses. An important milestone in our approach is the development of a practical, enantioselective synthesis of a functionalized aminoindoline scaffold, **4**, that could be further utilized in introducing skeletal diversity.[7] This scaffold is highly unique and comprises four orthogonal protecting groups. Our plan was to utilize the phenolic hydroxyl group as an immobilization site in solid-phase synthesis. The remaining three functional groups could further be used in complexity-generating, diversity-oriented reactions. As shown in Scheme 1, aminoindoline **4** could be easily converted into **5**, which comprises a conjugated carboxylate ester. Following the immobilization of **5** through the phenolic hydroxyl group on a solid support to give **6** and upon selective removal of the indoline protecting group, the substrate could then be coupled to an amino acid to give **7**. A key step in our approach is the formation of a six-membered ring by a stereoselective, conjugate hetero (e.g. aza)-Michael reaction. This could provide the indoline-alkaloid-like tricyclic derivative **8**, in which the diversity could easily be introduced at four sites. A six-membered ring-closure strategy that involves the trapping of the primary amine by a conjugated carboxylic ester could also provide a general method to the synthesis of cyclic β-amino acids.[8]

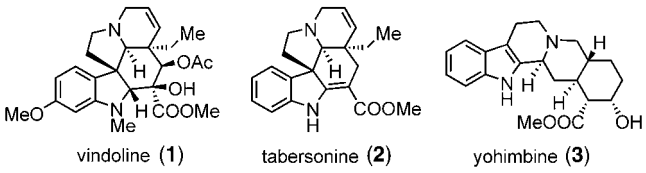
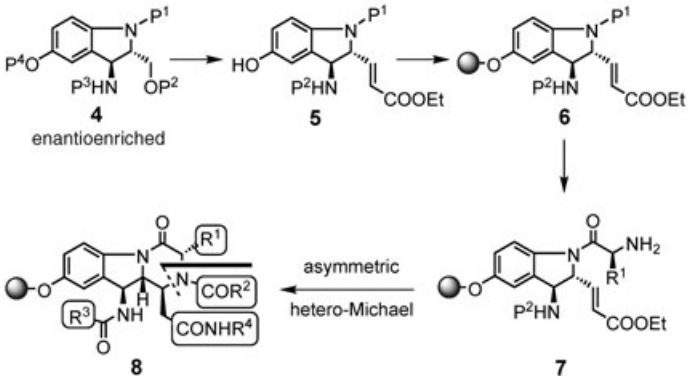


Figure 1. Examples of bioactive indole and indoline alkaloids.



Scheme 1. Natural-product-like and indoline-alkaloid-like complex polycyclic compounds. P = protecting group, R = diverse functional groups.

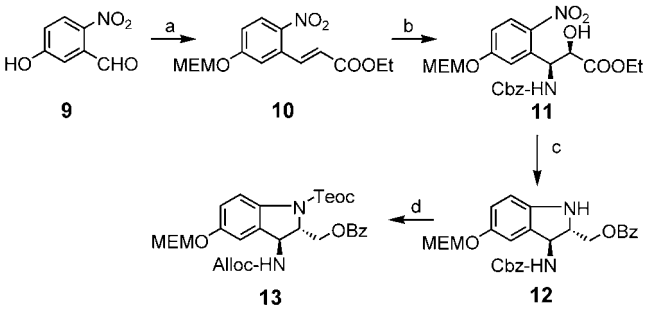
The enantioselective synthesis of aminoindoline derivative **13** is shown in Scheme 2. 5-Hydroxy-2-nitro-benzaldehyde (**9**) was converted into **10** in two steps that involve protection of the phenol and elongation of the carbon chain. Compound **10** was then subjected to an asymmetric aminohydroxylation reaction to give compound **11** in 79% yield (>92% *ee*).[9] The aminoindoline **12** was obtained from **11** in several steps that involved 1) reduction of the carboxylate ester (70%), 2) benzoyl-protection of the primary alcohol (OBz, 88%), 3) tosylation of the secondary alcohol (OTs, 87%), 4) selective reduction of the nitro group, and 5) cyclization under mild basic conditions (75% for two steps). Finally, compound **13** was obtained from **12** in three steps by which the indoline nitrogen was protected (Teoc, 98%), the Cbz protecting group was removed under hydrogenation, and the benzylic amine was protected with an Alloc group (85% for two steps, see Scheme 2). The overall sequence is very clean and the desired aminoindoline **13** could be obtained in

[*] Dr. Z. Gan, Dr. P. T. Reddy, S. Quevillon, Dr. S. Couve-Bonnaire, Dr. P. Arya
Chemical Biology Program
Steacie Institute for Molecular Sciences
National Research Council of Canada
100 Sussex Drive, Ottawa, Ontario, K1A0R6 (Canada)
Fax: (+1) 613-952-0068
E-mail: prabhat.arya@nrc.ca
S. Quevillon, Dr. P. Arya
Department of Chemistry
University of Ottawa
10 Marie Curie, Ottawa, K1N 6N5 (Canada)

[**] We sincerely thank Stuart Schreiber, Jared Shaw, and the DOS team members at the Broad Institute of Harvard and MIT for providing alkylsilyl-linker-based macrobeads and for their help in our solid-phase synthesis projects. This work was supported by a NRC genomics and health initiative program, special VP-NRC funds for chemical biology, the National Cancer Institute of Canada (NCIC), and the Canadian Institutes of Health Research (CIHR). Malgosia Daroszewska, Michael Barnes, and Don Leek are thanked for their technical support.

Supporting information for this article is available on the WWW under http://www.angewandte.org or from the author.

  DOI: 10.1002/ange.200462298



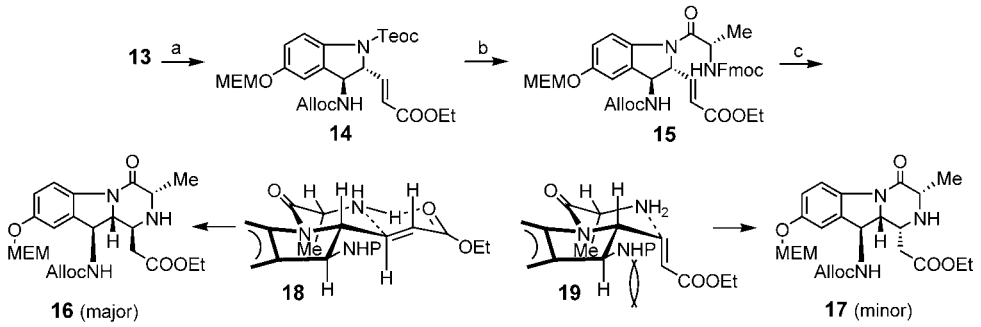
Scheme 2. a) 1) MEMCl, DIPEA, 97%; 2) $(EtO)_2P(O)CH_2COOEt$, NaH, 94%; b) Sharpless aminohydroxylation, 79%; c) 1) $LiBH_4$, 70%; 2) BzCl, pyridine, 88%; 3) TsCl, DMAP, 87%; 4) H_2, Lindlar cat.; 5) K_2CO_3, THF, 75% for two steps; d) 1) TeocCl, pyridine, 98%; 2) H_2, 10% Pd/C, MeOH; 3) AllocCl, pyridine, 85% for two steps. MEM = Methoxyethoxymethyl, DIPEA = diisopropylethylamine, Cbz = carbobenzyloxy, Bz = benzyl, Ts = *p*-toluenesulfonyl, DMAP = 4-dimethylaminopyridine, Teoc = (trimethylsilyl)ethoxycarbonyl, Alloc = Allyloxycarbonyl.

large quantities (5–10 g) in a short period. With a practical method in hand for this scaffold, the next step was to prepare **15**, a key starting material to explore the stereoselective, conjugate hetero-Michael approach.

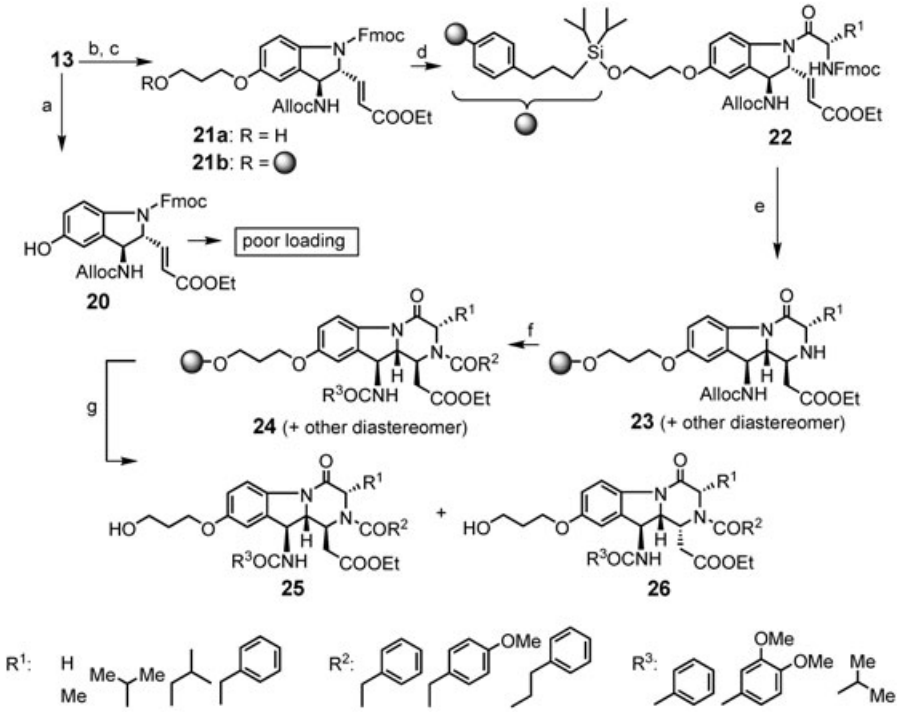
Scheme 3 shows our model study to obtain a tricyclic derivative by use of a stereoselective, conjugate hetero-Michael approach as the key step. Conversion of **13** into the conjugated ester **14** required the removal of the protecting group at the primary alcohol (99%), oxidation of the alcohol to the aldehyde, followed by a Wittig chain extension (93% for two steps). In a test study, upon removal of the Teoc group, the indoline amine was coupled with the amino acid chloride to give *N*-Fmoc-protected derivative **15** (88% for two steps). Upon removal of the Fmoc group (20% piperidine), we were delighted that the six-membered ring formed under these mild conditions. The primary amine generated in situ was easily trapped by the conjugated carboxylate ester in a stereoselective manner to give the cyclic compounds **16** (major) and **17** (minor) in 96% overall yield ($R^1 = Me$, > 10:1 ratio of two diastereomers, 82% d.r. for the major product). The stereochemistry of the new stereogenic center was assigned by NMR spectroscopy studies. The ease of the asymmetric, conjugate hetero-Michael approach to yield the tricylic product was a pleasant surprise and it opens a simple and very attractive approach to the synthesis of cyclic β-amino acids. The stereochemical preference of this reaction is postulated by the proposed transition states (**18** and **19**) that may account for the attack of the nucleophile from the β face. The conjugate hetero-Michael reaction is highly reproducible and gives the desired cyclic product(s) upon use of different amino acid derivatives.

The model solid-phase synthesis is shown in Scheme 4. Compound **20** was obtained from **13** in a few steps to perform solid-phase synthesis. In our hands, this scaffold gave poor loading with the use of (4-methoxyphenyl)diisopropylsilyl-propyl polystyrene beads (50–100 μm, loading 1.4 mmol g^{-1}).[10] At this stage, we decided to work with compound **21a**, in which a three-carbon spacer was introduced between the aromatic moiety and the primary alcohol group.[11] As expected, the loading of **21a** with commercially available silyl-linker-based beads and with the macrobeads (500–560 μm loading 1.29 mmol g^{-1})[12,13] worked very well to give product **21b** (1.12 mmol g^{-1}, 87% upon cleavage of the product from the macrobead support).

The next series of steps were then attempted on the solid support. The indoline amine was first deprotected of its Fmoc group and then coupled with the Fmoc-protected amino acid chloride (first diversity) to give the amino acid coupled product, **22**. Several attempts were then made to optimize the conditions for the solid-phase coupling reaction, and the use of collidine as a base gave the best results. The coupled product, **22**, was then treated with piperidine, and, as observed in the synthesis carried out in solution, we were pleased that the primary amine was trapped with the conjugated carboxyl ester to give the tricyclic derivative **23** during the removal of the Fmoc group. Once again, as with the solution-state synthesis, the in situ conjugate hetero-Michael reaction was highly reproducible in the solid phase. The mild conditions for this cyclization reaction are highly appealing and attractive to explore its potential in the generation of a molecule library. The stereochemical outcome of this reaction was found to be dependent upon the choice of the amino acid, and the ratio of the two diastereomers, **25** and **26**, varied from 5:1 to 1:1.[14] To complete the test sequence in the solid phase, **23** was then subjected to 1) an amide coupling reaction to introduce the second diversity, 2) removal of the Alloc protecting group to give the free amine, and 3) reaction with carboxylic acid chloride to introduce the third diversity and to give **24**. Finally, compounds **25** and **26** were obtained upon cleavage of the substrates from the support under



Scheme 3. a) 1) K_2CO_3, MeOH, 99%; 2) Dess–Martin periodinane; 3) $Ph_3P{=}CHCOOEt$, 93% for two steps; b) 1) TBAF; 2) Fmoc alanine chloride, pyridine, 88%; c) 20% piperidine, 96%. TBAF = tetra-*n*-butylammonium fluoride, Fmoc = 9-fluorenylmethoxycarbonyl.



Scheme 4. a) and b) see Supporting Information; c) (4-methoxyphenyl)diisopropylsilyl-propyl polystyrene macrobeads (500–560 μm, loading 1.29 mmol g^{-1}), 87% after cleavage from support; d) 1. 20% piperidine; 2. Fmoc amino acid chloride, collidine; e) 20% piperidine; f) 1. R^2COCl, pyridine; 2. $Pd(PPh_3)_4$; 3. R^3COCl, pyridine; g) pyridine–HF; 80–85% yield of **25** and **26** over 7 steps from **21b**.

desilylation conditions. Interestingly, compounds **25** and **26** were obtained from **21b** in seven steps in 80–85% overall yield. The overall sequence on the solid support utilizes very mild reaction conditions that include a crucial in situ conjugate hetero-Michael reaction to give functionalized aminoindoline-based, alkaloid-like, tricyclic derivatives. With alkylsilyl-linker-based macrobeads, this method was further utilized in the generation of a test library of 90 compounds (two diastereomers per well) by an IRORI split-and-mix-type encoded method. After completion of the library synthesis, the beads were taken out of the IRORI Kans and then subjected to desilylation cleavage conditions.[15]

To summarize, a library of indoline-alkaloid-like polycyclic compounds was synthesized on the basis of an enantiopure aminoindoline scaffold. The key reaction was the stereoselective, conjugate hetero-Michael reactions with nitrogen nucleophiles to obtain cyclic β-amino acid derived compounds. Furthermore, biological studies of this library in various cellular assays are in progress and the findings will be reported in due course.

Received: October 14, 2004
Published online: January 26, 2005

Keywords: combinatorial chemistry · cyclization · indoline alkaloids · protecting groups · solid-phase synthesis

[1] a) S. L. Schreiber, *Chem. Eng. News* **2003**, *81*, 51–61; b) T. Gura, *Nature* **2000**, *407*, 282–284; c) R. L. Strausberg, S. L. Schreiber, *Science* **2003**, *300*, 294–295; d) T. U. Mayer, *Trends Cell Biol.* **2003**, *13*, 270–277; e) J. R. Peterson, T. J. Mitchison, *Chem. Biol.* **2002**, *9*, 1275–1285.

[2] a) M. R. Arkin, J. A. Wells, *Nat. Rev. Drug Discovery* **2004**, *3*, 301–317; b) T. Berg, *Angew. Chem.* **2003**, *115*, 2566–2586; *Angew. Chem. Int. Ed.* **2003**, *42*, 2462–2481; c) D. L. Boger, J. Desharnais, K. Capps, *Angew. Chem.* **2003**, *115*, 4270–4309; *Angew. Chem. Int. Ed.* **2003**, *42*, 4138–4176; d) R. Breinbaur, I. R. Vetter, H. Waldmann, *Angew. Chem.* **2002**, *114*, 3002–3015; *Angew. Chem. Int. Ed.* **2002**, *41*, 2878–2890; e) K. Hinterding, D. Alonso-Díaz, H. Waldmann, *Angew. Chem.* **1998**, *110*, 716–780; *Angew. Chem. Int. Ed.* **1998**, *37*, 688–749; f) A. Huwe, R. Mazitschek, A. Giannis, *Angew. Chem.* **2003**, *115*, 2170–2187; *Angew. Chem. Int. Ed.* **2003**, *42*, 2122–2138; g) T. Klabunde, G. Hessler, *ChemBioChem* **2002**, *3*, 928–944.

[3] a) M. D. Burke, S. L. Schreiber, *Angew. Chem.* **2004**, *116*, 48–60; *Angew. Chem. Int. Ed.* **2004**, *43*, 46–58; b) M. D. Burke, E. M. Berger, S. L. Schreiber, *Science* **2003**, *302*, 613–618; c) S. Borman, *Chem. Eng. News* **2004**, *82*, 32–40.

[4] For a review on solution- and solid-phase library generation of natural product analogues and natural-product-like compounds, see: a) D. G. Hall, S. Manku, F. Wang, *J. Comb. Chem.* **2001**, *3*, 125–150; b) P. Arya, R. Joseph, D. T. H. Chou, *Chem. Biol.* **2002**, *9*, 145–156.

[5] For selected examples of solid-phase library synthesis on benzopyrans as a privileged substructure, see: a) K. C. Nicolaou, J. A. Pfefferkorn, G.-Q. Cao, *Angew. Chem.* **2000**, *112*, 750–755; *Angew. Chem. Int. Ed.* **2000**, *39*, 734–739; b) K. C. Nicolaou, G.-Q. Cao, J. A. Pfefferkorn, *Angew. Chem.* **2000**, *112*, 755–759; *Angew. Chem. Int. Ed.* **2000**, *39*, 739–743; c) K. C. Nicolaou, J. A. Pfefferkorn, A. J. Roecker, G.-Q. Cao, S. Barluenga, H. J. Mitchell, *J. Am. Chem. Soc.* **2000**, *122*, 9939–9953; d) K. C. Nicolaou, J. A. Pfefferkorn, H. J. Mitchell, A. J. Roecker, S. Barluenga, G.-Q. Cao, R. L. Affleck, J. E. Lillig, *J. Am. Chem. Soc.* **2000**, *122*, 9954–9967; e) K. C. Nicolaou, J. A. Pfefferkorn, S. Barluenga, H. J. Mitchell, A. J. Roecker, G.-Q. Cao, *J. Am. Chem. Soc.* **2000**, *122*, 9968–9976.

[6] P. Arya, C.-Q. Wei, M. L. Barnes, M. Daroszewska, *J. Comb. Chem.* **2004**, *6*, 65–72.

[7] For selected examples of combinatorial approaches to obtain skeletally diverse architectures, see: a) M. D. Burke, E. M. Berger, S. L. Schreiber, *Science* **2003**, *302*, 613–618; b) M. D. Burke, E. M. Berger, S. L. Schreiber, *J. Am. Chem. Soc.* **2004**, *126*, 14095–14104.

[8] For a review on β-amino acids, see: M. Liu, M. P. Sibi, *Tetrahedron* **2002**, *58*, 7991–8035.

[9] The % *ee* values were determined by chiral HPLC.

[10] The resin was purchased from Novabiochem.

[11] For the synthesis of **21a**, see the Supporting Information.

[12] J. A. Tallarico, K. M. Depew, H. E. Pelish, N. J. Westwood, C. W. Lindsley, M. D. Shair, S. L. Schreiber, M. A. Foley, *J. Comb. Chem.* **2001**, *3*, 312–318.

[13] The alkylsilyl-linker-based macrobeads were provided by ICCB, Harvard Medical School, Harvard University.

[14] The explanation for the decrease in diastereoselectivity during the solid-phase conjugate hetero-Michael reaction is not clear at this stage.

[15] For detailed information on library synthesis, see the Supporting Information.

