J|A|C|S
ARTICLES
Published on Web 04/24/2002

Quasiracemic Synthesis: Concepts and Implementation with a Fluorous Tagging Strategy to Make Both Enantiomers of Pyridovericin and Mappicine

Qisheng Zhang, Alexey Rivkin, and Dennis P. Curran*

Contribution from the Department of Chemistry, University of Pittsburgh, Pittsburgh, Pennsylvania, 15260

Received January 15, 2002

Abstract: The concept of quasiracemic synthesis is introduced and illustrated with syntheses of both enantiomers of pyridovericin (whose absolute configuration is assigned as R) and mappicine. Like racemic synthesis, quasiracemic synthesis provides both enantiomers in a single synthetic sequence; however, separation tagging is used to ensure that quasiracemic mixtures can be analyzed, separated, and identified on demand. Fluorous tags of differing chain lengths are used to tag two enantiomeric starting materials. The resulting quasienantiomers are mixed to make a quasiracemate, which is then treated like a true racemate in successive steps of the synthesis. Fluorous chromatography is used to separate, or demix, the final quasiracemate into its two components, which are then detagged to provide (true) enantiomeric products. Quasiracemic synthesis is portrayed as the first and simplest of a series of mixture synthesis techniques based on separation tagging, and the prospects for using other types of separation tags are briefly evaluated.

Introduction

Chiral compounds abound in organic, medicinal, and natural products chemistry. Because biological activity and other properties are a function of the absolute configuration, the synthesis of chiral compounds in enantiopure form is of utmost importance. Today, there are two ways to make enantiopure (or enantioenriched) organic molecules: racemic synthesis followed by resolution, or asymmetric synthesis.[1] We introduce herein a third method, quasiracemic synthesis, which unites some of the key advantages of racemic and asymmetric synthesis.

Classical racemic synthesis makes both enantiomers of a target compound in a single synthesis, but separation (resolution) and identification of the final enantiomers pose large hurdles. Asymmetric synthesis employs enantiopure compounds, but two separate syntheses are needed if both enantiomers are desired. Like asymmetric synthesis, quasiracemic synthesis starts and finishes with enantiopure compounds, but like racemic synthesis it provides both enantiomers in a single synthesis. "Quasi-enantiomers" are used in place of enantiomers, and the separation and identification of the final quasienantiomers is ensured by a tagging strategy.[2]

We use the term quasienantiomers as defined qualitatively by Eliel.[1a] Quasienantiomers are not enantiomers because they are not isomers; "most" of one quasienantiomer is reflected into the other by a mirror, but the molecules differ in one part such that an atom or group of one quasienantiomer is reflected into a similar but not identical atom or group of the other. For example, (*S*)-2-chlorobutane and (*R*)-2-bromobutane are quasienantiomers. "Quasiracemic" follows as a mixture of equal parts of quasienantiomers.[3] The "method of quasiracemates" is a classical technique used to assign absolute configuration,[1] and innovative new resolution methods based on quasienantiomers have been introduced by Bergbreiter,[4] Vedejs,[5] and Zwanenburg.[6]

The tagging strategy of quasiracemic synthesis is shown in a very general way in Figure 1. Enantiomeric compounds (*R*)-**1** and (*S*)-**1** are tagged with different tags to provide "quasi-enantiomers" (*R*)-**2a** and (*S*)-**2b**. The quasienantiomers (*R*)-**2a** and (*S*)-**2b** are mixed to make a quasiracemic mixture, which is then taken through a series of steps to make a final tagged product mixture (*R*)-**3a**/(*S*)-**3b**. The mixture is then separated by a method that is selective based on the tag to provide the two pure quasienantiomers. The tag is finally removed to generate the two true enantiomers (*R*)-**4** and (*S*)-**4**.

Ideally, tags for quasiracemic synthesis behave as if they are identical during intermediate steps of the synthesis. This means that tagged quasienantiomers should behave like true enantiomers: they should have the same solubility, reactivity (toward

* Corresponding author. E-mail: curran+@pitt.edu.

(1) (a) Eliel, E. L.; Wilen, S. H. *Stereochemistry of Organic Compunds*; Wiley-Interscience: New York, 1994; p 1267. (b) Jacques, J.; Collet, A.; Wilen, S. H. *Enantiomers, Racemates and Resolutions*; Wiley: New York, 1981.

(2) The concept of separation tagging is generalized in: Curran, D. P. *Angew. Chem., Int. Ed. Engl.* **1998**, *37*, 1175.

(3) The terms "pseudoenantiomers" and "pseudoracemates" are often used with the same meanings, although pseudoracemate has another meaning (a crystalline form consisting of a solid solutiuon of two (true) enantiomers). See ref 1a, p 159.

(4) Bergbreiter, D. E.; Zhang, L. *J. Chem. Soc., Chem. Commun.* **1993**, 596.

(5) (a) Vedejs, E.; Chen, X. *J. Am. Chem. Soc.* **1997**, *119*, 2584. (b) Vedejs, E.; Rozners, E. *J. Am. Chem. Soc.* **2001**, *123*, 2428.

(6) ten Holte, P.; Thijs, L.; Zwanenburg, B. *Org. Lett.* **2001**, *3*, 1093.

10.1021/ja025606x CCC: $22.00

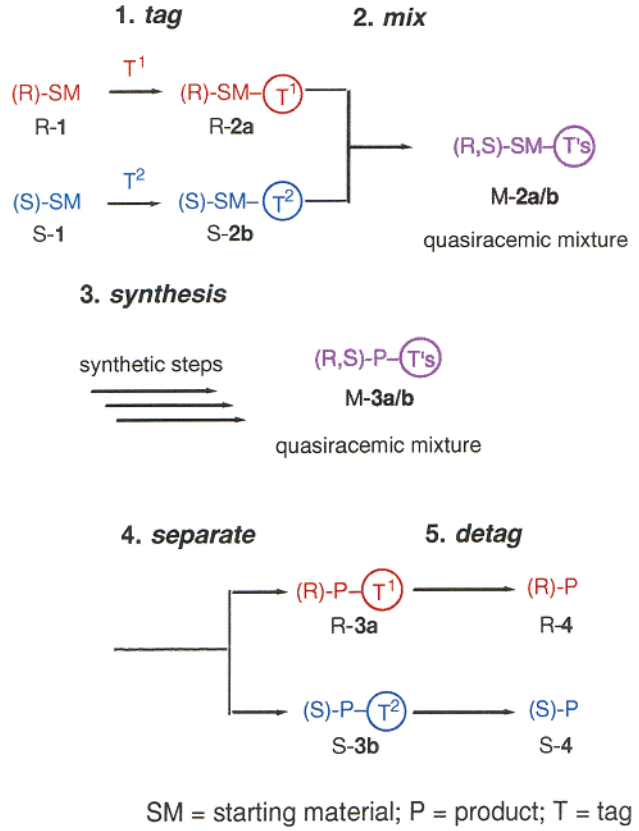


Figure 1. Quasiracemic synthesis by separation tagging.

achiral species), physical properties and spectral characteristics. They should also have similar, preferably identical, chromatographic properties on regular and reverse phase silica gel. However, tags must be chosen such that there is at least one "orthogonal" reaction or separation method that can be used to differentiate on demand the quasiracemic mixtures based on the tag.

While there may be no tag that exhibits "ideal" behavior, we posit that many tags will be close enough to ideal to be useful. To demonstrate quasiracemic synthesis, we selected fluorous tags bearing slightly different perfluorinated fragments. Fluorous chromatography is the separation method that is complementary to the tag.[7] Fluorous methods have been exploited recently in a diverse collection of settings with an underlying theme of separating fluorous-tagged molecules from organic molecules.[8] Here we instead capitalize on the ability to separate fluorous-tagged molecules from each other, or to prevent that separation, depending on whether fluorous or nonfluorous separation methods are used.

Quasiracemic syntheses of two natural products, pyridovericin **5** and mappicine **6**, are described below. Pyridovericin (Figure

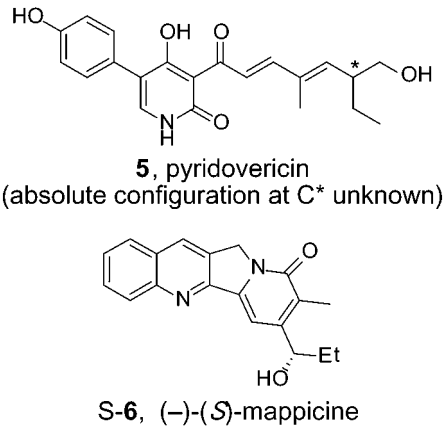


Figure 2. Structures of pyridovericin and mappicine.

2) was isolated from the entomopathogenic fungus *Beauveria bassiana* EPF-5 in 1998, and is an inhibitor of the protein tyrosine kinase.[9] This pyridone has one stereocenter of unknown absolute configuration along with a primary hydroxy group that serves as a convenient location for the tag. Initial goals were to synthesize both enantiomers of pyridovericin **5** in enantiopure form and to assign the absolute configuration of the natural product. As the work unfolded, only the second goal was met. To reach the first, we selected the antiviral agent mappicine **6**,[10] which has been made by a number of groups.[11] We have used an isonitrile cascade annulation[12] to make mappicine by racemic synthesis,[13] asymmetric synthesis,[14] solid-phase synthesis (with little success[15]), and solution phase parallel synthesis.[12] Herein we use the same strategy for the quasiracemic synthesis. The quasiracemic synthesis of mappicine along with the synthesis of analogues by "fluorous mixture synthesis" has been briefly communicated.[16]

Results and Discussion

Figure 3 shows the plan for the synthesis of pyridovericin **5**. This plan follows directly from a synthesis of tenellin by Rigby and Qabar.[17] Tenellin (not shown) is an isomer of pyridovericin with a hydroxyl group on the pyridone nitrogen rather than at the end of one of the branches of the side chain. The centerpiece of the synthesis is Rigby's [4 + 2] annulation[18] between ketoester **7** and vinyl isocyanate **8** to make the pyridone ring. Working back through two olefinations provides the chiral monoprotected diols **9**. The plan was to prepare both quasi-

(7) Curran, D. P. *Synlett* **2001**, 1488.
(8) (a) Horváth, I. T. *Acc. Chem. Res.* **1998**, *31*, 641. (b) Curran, D. P. *The Cancer J.* **1998**, *4* (Supp. 1), S73. (c) Barthel-Rosa, L. P.; Gladysz, J. A. *Coord. Chem. Rev.* **1999**, *192*, 587. (d) Curran, D. P.; Hadida, S.; Studer, A.; He, M.; Kim, S.-Y.; Luo, Z.; Larhed, M.; Hallberg, M.; Linclau, B. In *Combinatorial Chemistry: A Practical Approach*; Fenniri, H., Ed.; Oxford University Press: Oxford, 2001; Vol. 2, p 327. (e) Curran, D. P. In *Stimulating Concepts in Chemistry*; Vögtle, F., Stoddart, J. F., Shibasaki, M., Eds.; Wiley-VCH: New York, 2000; p 25. (f) Curran, D. P. *Pure Appl. Chem.* **2000**, *72*, 1649. (g) Curran, D. P.; Hadida, S.; Studer, A.; He, M.; Kim, S. Y.; Luo, Z. Y.; Larhed, M.; Hallberg, A.; Linclau, B. In *Combinatorial Chemistry*; Fenniri, H., Ed.; Oxford University Press: New York, 2000; Vol. 233, p 327. (h) Curran, D. P.; Luo, Z. Y. *J. Am. Chem. Soc.* **1999**, *121*, 9069. (i) Luo, Z. Y.; Williams, J.; Read, R. W.; Curran, D. P. *J. Org. Chem.* **2001**, *66*, 4261.
(9) (a) Takahashi, S.; Uchida, K.; Kakinuma, N.; Hashimoto, R.; Yanagisawa, T.; Nakagawa, A. *J. Antibiot.* **1998**, 51, 1051. (b) Takahashi, S.; Kakinuma, N.; Uchida, K.; Hashimoto, R.; Yanagisawa, T.; Nakagawa, A. *J. Antibiot.* **1998**, 51, 596.
(10) (a) Govindachari, T. R.; Ravindranath, K. R.; Viswanathan, N. *J. Chem. Soc., Perkin Trans. 1* **1974**, 1215. (b) Pirillo, A.; Verotta, L.; Gariboldi, P.; Torregiani, E.; Bombardelli, E. *J. Chem. Soc., Perkin Trans. 1* **1995**, 583.
(11) (a) Comins, D. L.; Saha, J. K. *J. Org. Chem.* **1996**, *61*, 9623. (b) Boger, D. L.; Hong, J. Y. *J. Am. Chem. Soc.* **1998**, *120*, 1218. (c) Yadav, J. S.; Sarkar, S.; Chandrasekhar, S. *Tetrahedron* **1999**, *55*, 5449. (d) Mekouar, K.; Genisson, Y.; Leue, S.; Greene, A. E. *J. Org. Chem.* **2000**, *65*, 5212. (e) Ishibashi, H.; Kato, I.; Takeda, Y.; Tamura, O. *Tetrahedron Lett.* **2001**, *42*, 931. (f) Toyota, M.; Komsii, C.; Ihara, M. *J. Org. Chem.* **2000**, *65*, 7110. (g) Das, B.; Madhusudhan, P. *Tetrahedron* **1999**, *55*, 7875.
(12) (a) Curran, D. P.; Liu, H. *J. Am. Chem. Soc.* **1991**, *113*, 2127. (b) Josien, H.; Ko, S. B.; Bom, D.; Curran, D. P. *Chem. Eur. J.* **1998**, *4*, 67.
(13) Josien, H.; Curran, D. P. *Tetrahedron* **1997**, *53*, 8881.
(14) deFrutos, O.; Curran, D. P. *J. Comb. Chem.* **2000**, *2*, 639.
(15) Gabarda, A.; Choi, J.-H.; Curran, D. P. Unpublished observations.
(16) Luo, Z. Y.; Zhang, Q. S.; Oderaotoshi, Y.; Curran, D. P. *Science* **2001**, *291*, 1766.
(17) Rigby, J. H.; Qabar, M. *J. Org. Chem.* **1989**, 54, 5852.
(18) Rigby, J. H. *Synlett* **2000**, 1.

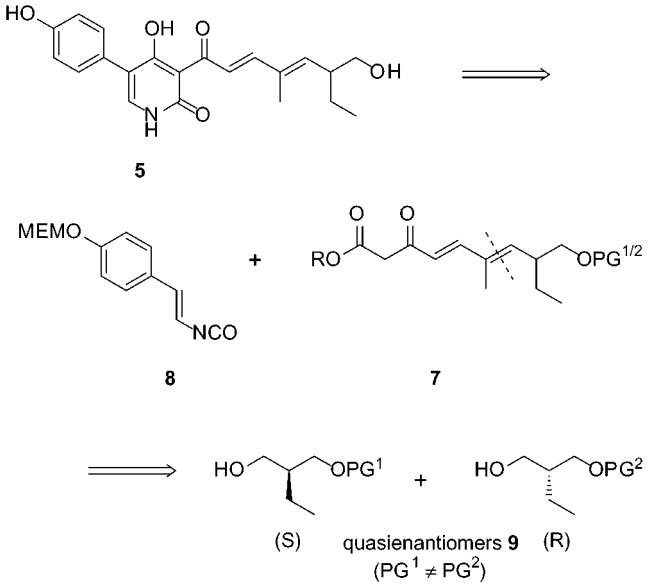


Figure 3. Strategy for quasiracemic synthesis of pyridovericin.

enantiomers of diol **9** protected with different fluorous tags (PG^1, PG^2), and then to mix the quasienantiomers for the quasiracemic synthesis. This plan maximizes the efficiency of the mixture synthesis by mixing early and demixing late.

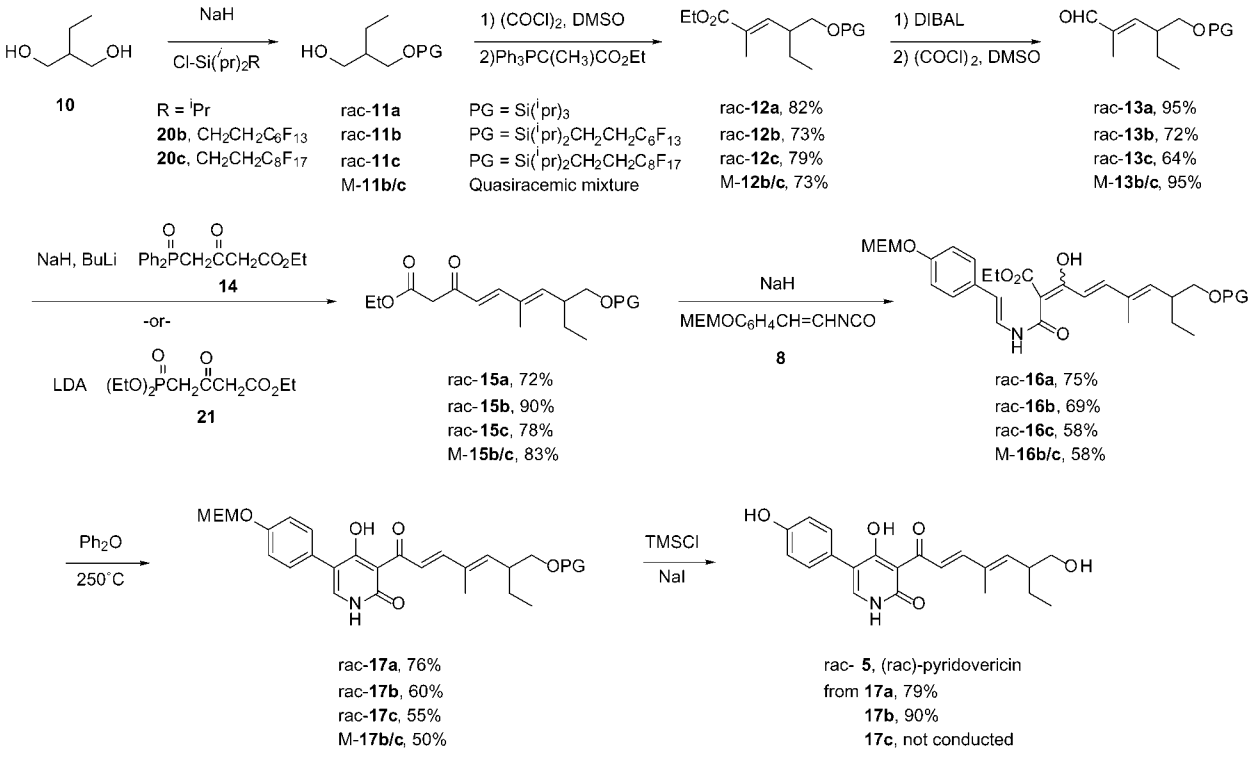
Scheme 1 summarizes all four of the syntheses of pyridovericin conducted in this work. Three of these are traditional racemic syntheses with a nonfluorous and two different fluorous tags (protecting groups), and the fourth is the quasiracemic synthesis. Prefixes before numbers have the following meanings: "*rac*", true racemate; "*R*" or "*S*", enriched in the *R*- or *S*-enantiomer; and "M", quasiracemic mixture made by manual mixing of quasienantiomers. Suffixes designate the alcohol protecting groups: "a" series, triisopropylsilyl; "b" series, perfluorhexylethyl diisopropylsilyl; and "c" series, perfluorooctylethyl diisopropylsilyl.

To validate the synthetic plan and confirm the structure, we first synthesized racemic pyridovericin using a standard (nonfluorous) protecting group. Monosilylation of 2-ethyl-1,3-propane diol **10** with triisopropylsilyl chloride gave *rac*-**11a** in 98% yield.[19] Swern oxidation[20] and Wittig olefination provided unsaturated ester *rac*-**12a** in 82% isolated yield (>97% E). DIBAL reduction[21] and Swern oxidation provided aldehyde *rac*-**13a** in 95% yield. This was olefinated with phosphine oxide **14**[22] to give *β*-keto ester *rac*-**15a** in 72% yield as a mixture of keto and enol forms (enol form not shown).

Keto ester *rac*-**15a** was then condensed with readily available isocyanate **8**[23] according to the two-step protocol of Rigby and Qabar.[24] Deprotonation of *rac*-**15a** with sodium hydride at 0 °C was followed by addition of vinyl isocyanate **7** and warming to room temperature. After 12 h, workup and flash chromatographic purification provided enamide *rac*-**16a**. This existed in the enol form as indicated, although the geometry of the enol double bond was not assigned. Heating of *rac*-**16a** for 5 min at 250 °C in diphenyl ether provided pyridone *rac*-**17a** in 76% yield. Shorter times or lower temperatures gave incomplete conversion, while longer times resulted in lower yields due to product decomposition.

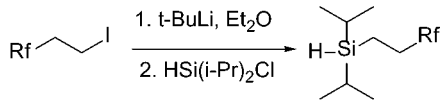
We initially planned to remove the MEM group of *rac*-**17a** first followed by the TIPS group, and later to conduct demixing between these two steps in the quasiracemic synthesis. However, treatment of *rac*-**17a** with TMSCl and sodium iodide at −20 °C for 2 h[25] provided the desilylated product with the MEM group intact (not shown). Prolonged exposure of *rac*-**17a** to a larger excess of reagents then removed the MEM group to provide *rac*-pyridoverin **5** in 79% overall yield. This was identical in all respects (except rotation) with the natural product, so both the structure and the synthesis were validated. The

Scheme 1

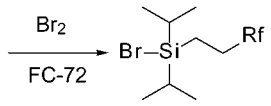


observations in deprotection of *rac*-**17a** led us to reformulate the plan; demixing would be conducted before any deprotection and then both protecting groups would be removed simultaneously by prolonged exposure to TMSCl/NaI.

We chose fluorous diisopropyl silanes bearing $-(CH_2)_2C_6F_{13}$ and $-(CH_2)_2C_8F_{17}$ groups as tags for the quasienantiomers. The requisite known silanes[26] were prepared as shown in eq 1.



18b, Rf = C_6F_{13}
18c, Rf = C_8F_{17} **19b,c**



(1)

20b,c

Addition of diisopropylchlorosilane to the lithium reagents derived from perfluorohexylethyl iodide **18b** and perfluorooctylethyl iodide **18c** gave the silanes **19b**,**c**. These were then brominated in situ to give solutions of silyl bromides **20b**,**c**, which were used immediately for silylations.

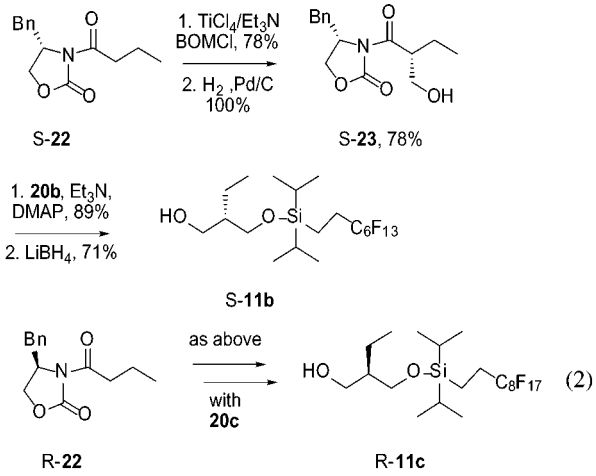
To ensure that the fluorous silyl groups behave identically to each other and to the triisopropylsilyl groups, we then repeated the synthesis two more times with racemic alcohols **11b**,**c** tagged to the corresponding fluorous silyl tags. The results of these syntheses are also summarized in Scheme 1. This proved to be a good decision since one of the steps failed.

Silylation of propane diol **10** with the in situ generated silyl bromides provided *rac*-**11b** and **11c** in 75% and 86% yields. Swern oxidation and olefination then provided *rac*-**12b**,**c** (73%, 79%). Reduction and a second Swern oxidation (72%, 64%) provided aldehydes *rac*-**13b**,**c** without event. However, olefination of *rac*-**13b** with phosphine oxide **14** under the conditions used for the nonfluorous silyl ether *rac*-**13a** and a variety of others failed to provide the keto ester *rac*-**15b**. In general, substantial amounts of the aldehyde were recovered. To solve this problem, we switched to using a phosphonate[27] **21** in place of the phosphine oxide **14**. Generation of the dianion of **21** with 2 equiv of LDA followed by addition of *rac*-**13b**,**c** gave keto esters *rac*-**15b**,**c** in 90% and 78% yields. Keto esters **15b**,**c** again existed as mixtures of keto/enol tautomers (enol not shown).

With the olefination problem solved, the remaining steps proceeded without event. Condensation of *rac*-**15b**,**c** with isocyanate **8** gave *rac*-**16b**,**c** (69%, 58%), and heating of these enols gave pyridones *rac*-**17b**,**c** (60%, 55%). Removal of both the silyl group and the MEM group occurred on exposure of *rac*-**17b** to excess TMSCl/NaI, and *rac*-pyridovericin **5** was produced in 90% yield. The larger homologue *rac*-**17c** was not deprotected.

Throughout these two separate syntheses, the chromatographic behavior of the products bearing shorter (**b** series, C_6F_{13}) and longer (**c** series, C_8F_{17}) tails was compared. In no case could the pairs of products be differentiated by thin-layer chromatography. This suggests that "accidental" separation of the quasienantiomers after mixing will not occur. In contrast, the pairs of products were well differentiated on a fluorous column (over 6 min difference in retention time with gradient elution on a Fluofix 120E column), so preparative demixing by fluorous chromatography was assured.

With the groundwork laid, we then moved ahead to the quasiracemic synthesis of pyridovericin with the double goals of producing both enantiomers in enantiopure form and assigning the absolute configuration of the natural product. Enantiopure monosilylated propane diol derivatives were prepared as shown in eq 2. Evans alkylation[28] of oxazolidinone (*S*)-**22** with



(2)

(benzyloxy)methyl chloride (BOMCl) according to Fukumoto[29] followed by removal of the benzyl group provided (*S*)-**23** as a single diastereomer in 78% yield after chromatography. Silylation of (*S*)-**23** with the shorter fluorous silyl tag (89%) and reductive removal of the auxiliary[30] gave (*S*)-**11b**. In turn, (*R*)-**11c** was prepared by an identical sequence of steps starting from (*R*)-**22**, but silylation was conducted with a longer fluorous silyl tag.

Equimolar amounts of quasienantiomers (*S*)-**11b** and (*R*)-**11c** were then mixed to make the first quasiracemic mixture M-**11b**/**c**, and this was taken forward in the synthesis summarized again in Scheme 1. Swern oxidation and olefination provided M-**12b**/**c** (73%), which was then reduced to give an alcohol followed by oxidation under Swern conditions to aldehyde M-**13b**/**c** (95%). Olefination with phosphonate **21** provided ketoester M-**15b**/**c** (83%, keto/enol mixture). Coupling

(19) McDougal, P. G.; Rico, J. G.; Oh, Y.-I.; Condon, B. D. *J. Org. Chem.* **1986**, *51*, 3388.
(20) Gaucher, A.; Ollivier, J.; Marguerite, J.; Paugam, R.; Salaün, J. *Can. J. Chem.* **1994**, *72*, 1312.
(21) Diez-Martin, D.; Kotecha, N. R.; Ley, S. V.; Mantegani, S.; Menendez, J. C.; Organ, H. M.; White, A. D.; Banks, B. J. *Tetrahedron* **1992**, *48*, 7899.
(22) van der Gen, A.; van den Goorbergh, J. A. M. *Tetrahedron Lett.* **1980**, *21*, 3621.
(23) Rigby, J. H.; Burkhardt, F. J. *J. Org. Chem.* **1986**, *51*, 1374.
(24) (a) Rigby, J. H.; Balasubramanian, N. *J. Org. Chem.* **1984**, *49*, 4569. (b) Rigby, J. H.; Balasubramanian, N. *J. Org. Chem.* **1989**, *54*, 224. (c) Rigby, J. H.; Holsworth, D. D.; James, K. *J. Org. Chem.* **1989**, *54*, 4019.
(25) Rigby, J. H.; Wilson, J. Z. *Tetrahedron Lett.* **1984**, *25*, 1429.
(26) Boutevin, B.; Guida-Pietrasanta, F.; Ratsimihety, A.; Caporiccio, G.; Gornowicz, G. J. *J. Fluorine Chem.* **1993**, *60*, 211. The silanes are now commercially available from Fluorous Technologies, Inc (www.fluorous.com).
(27) (a) Svendsen, A.; Boll, P. M. *Tetrahedron* **1973**, *29*, 4251. (b) Bodalski, R.; Pietrusiewicz, K. M.; Monkiewicz, J.; Koszuk, J. *Tetrahedron Lett.* **1980**, *21*, 2287.
(28) (a) Gage, J. R.; Evans, D. A. *Org. Synth.* **1989**, *68*, 83. (b) Evans, D. A.; Chapman, K. T.; Hung, D. T.; Kawaguchi, A. T. *Angew. Chem., Int. Ed. Engl.* **1987**, *26*, 1184.
(29) Ihara, M.; Katsumata, A.; Setsu, F.; Tokunaga, Y.; Fukumoto, K. *J. Org. Chem.* **1996**, *61*, 677.
(30) Crimmins, M.; King, B. W. *J. Am. Chem. Soc.* **1998**, *120*, 9084.

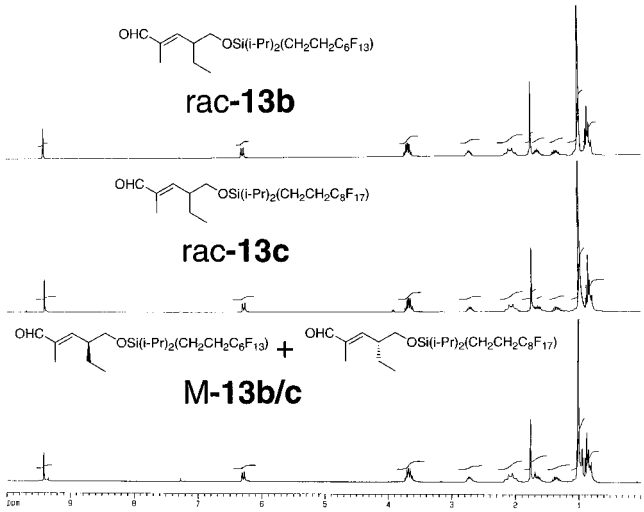


Figure 4. ^{1}H NMR spectra of *rac*-**13b**, *rac*-**13c**, and quasiracemic mixture M-**13b**/**c**.

with isocyanate **8** (58%) followed by heating at 250 °C for 8 min gave protected pyridovericin M-**17b**/**c** in 50% purified yield.

The seven mixture steps of the quasiracemic synthesis were conducted just as in the prior racemic syntheses, and yields were calculated based on the average molecular weight of the quasienantiomers. Most products were purified by flash chromatography on standard silica gel, and there was no evidence of separation of the quasienantiomers. We started with 440 mg of M-**11b**/**c** and finished with 230 mg of M-**17b**/**c**.

Quasiracemic mixtures were characterized in the standard way by ^{1}H NMR and ^{13}C NMR spectroscopy. Like spectra from true racemic mixtures, the spectra from the quasiracemic mixtures appeared to reflect resonances from a single compound. Figure 4 provides as an example the ^{1}H NMR spectra of *rac*-**13b**, *rac*-**13c,** and M-**13b**/**c** recorded at 300 MHz in $CDCl_3$; these spectra are indistinguishable. Broad band decoupled ^{13}C NMR spectra of these same molecules are shown in Figure 5. These spectra are also identical except in the region of 105−125 ppm, where the carbons bearing fluorines resonate. This region has a complicated set of low-intensity peaks (due to C−F coupling) which differ for the C_6F_{13}, C_8F_{17}, and mixture samples. With appropriate experiments and expansions, this complicated region can actually be fully analyzed to provide information about the tags. However, this is unnecessary since there are simpler ways to get this information (see below).

Complementary to ^{1}H and ^{13}C NMR spectroscopy, which provide information about the substrate but not the tag, is ^{19}F NMR spectroscopy, which provides information about the tag and not the substrate. Figure 6 shows the CF_2 region of three ^{19}F NMR spectra corresponding to *rac*-**13b**, *rac*-**13c**, and M-**13b**/**c**. These spectra show characteristic differences. For example, the peak at −120.5 ppm in *rac*-**13b** bearing the C_6F_{13} group integrates to two fluorines (one difluoromethylene group), while the same peak in *rac*-**13c** bearing the C_8F_{17} group integrates to six fluorines (three overlapping difluoromethylene groups). In the quasiracemic mixture (M-**13b**/**c**), this peak integrates to the average value of four fluorines. Also, the CF_2 group in the C_6F_{13} compound at −122.0 ppm is slightly upfield from its counterpart in the C_8F_{17} quasienantiomer at −121.8 ppm. There were no detectable variations in the ^{19}F NMR spectra of any of the intermediates as a function of change of

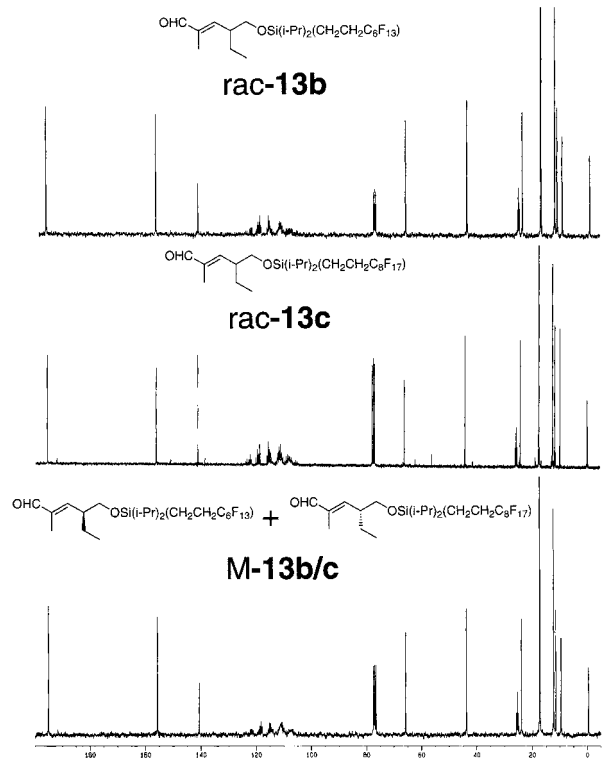


Figure 5. ^{13}C NMR spectra of *rac*-**13b**, *rac*-**13c**, and quasiracemic mixture M-**13b**/**c**.

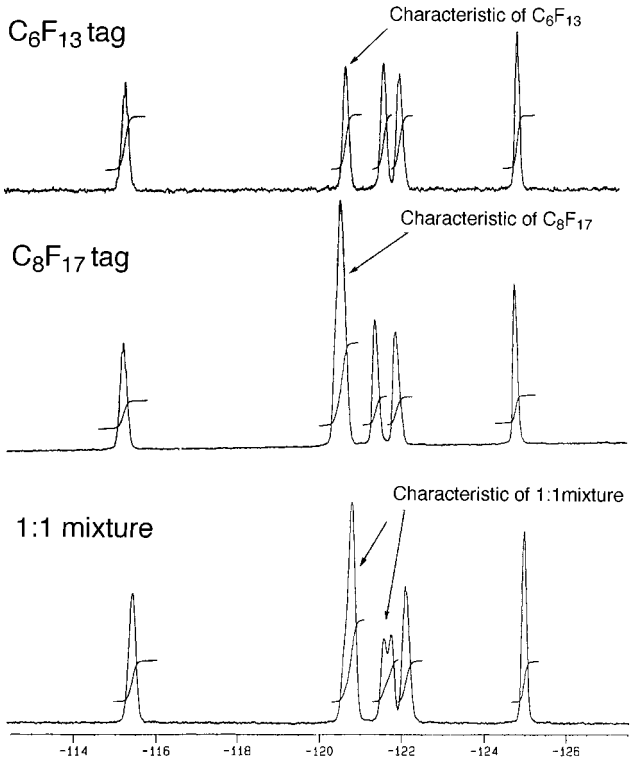


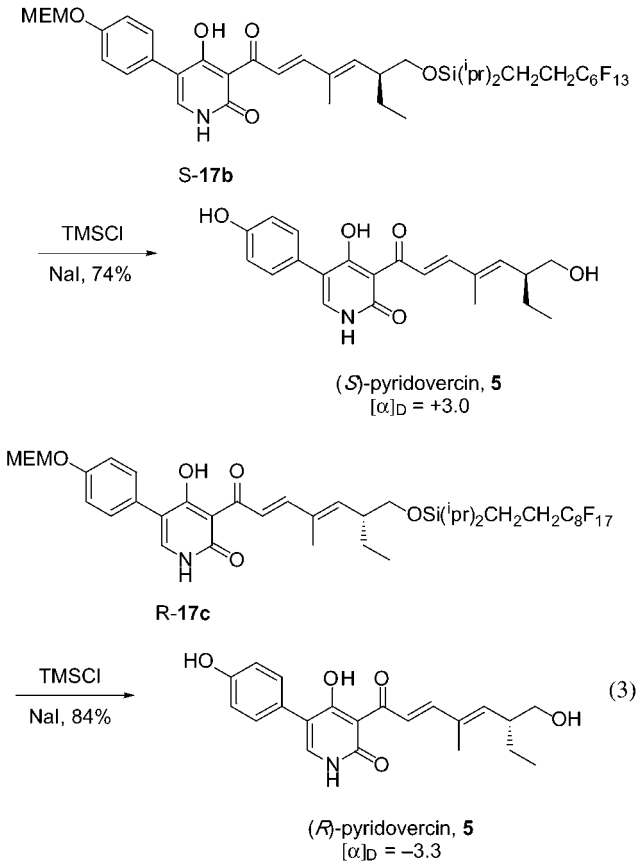
Figure 6. ^{19}F NMR spectra of *rac*-**13b**, *rac*-**13c**, and quasiracemic mixture M-**13b**/**c**.

the substrate part of the molecule. Evidently, the fluorines are too remote to be affected by the structural changes imposed by the synthesis. Accordingly, ^{19}F NMR spectroscopy can be used at any time to ensure (within the limits of integration error) that the quasiracemic mixture does not become deficient in one of the quasienantiomers due to differential reaction or separation.

Mass spectroscopy provides information about both the tag and the substrate, and LCMS with a fluorous hplc column is especially useful. Quasienantiomers (*S*)-**13b** and (*R*)-**13c** were separated by about 7 min under standard conditions (see Supporting Information), and were present in roughly equimolar ratios. Molecular ions corresponding to each quasienantiomer were observed by MS detection. Conveniently, the quasienantiomers differ in molecular weight by 100 mu (C_2F_4).

Prior to detagging, pyridone M-**17b**/**c** was demixed into its individual tagged components by semipreparative fluorous hplc. Considerably larger columns are now available, but at this time we were limited to injections of about 7 mg. A 30 min run included a 10 min linear gradient of 92% MeOH/water up to 100% MeOH followed by 20 min at 100% MeOH. Under these conditions, (*S*)-**17b** and (*R*)-**17c** eluted at about 13 and 18 min, respectively. A series of ten 7 mg injections provided 30 mg of the lighter **17b** and 33 mg of the heavier **17c**; the total recovery was 90%.

Compounds (*S*)-**17b** and (*R*)-**17c** were then individually detagged (deprotected) by using TMSCl/NaI in acetonitrile to give (*S*)- and (*R*)-pyridovericin **5**, respectively (eq 3). These



samples were purified first by flash chromatography and then by preparative reverse phase hplc (Symmetry C18 column). The NMR and mass spectra of each enantiomer were identical with that of natural pyridovericin. The optical rotation of (*S*)-**5** was +3.0 (*c* 0.1, MeOH) while that of (*R*)-**5** was −3.3 (*c* 0.06, MeOH). These rotations are considerably lower than the reported value of −20.3 (*c* 0.1 MeOH) for the natural product. Dr. Nakagawa of Teikyo University, Japan, kindly provided us a

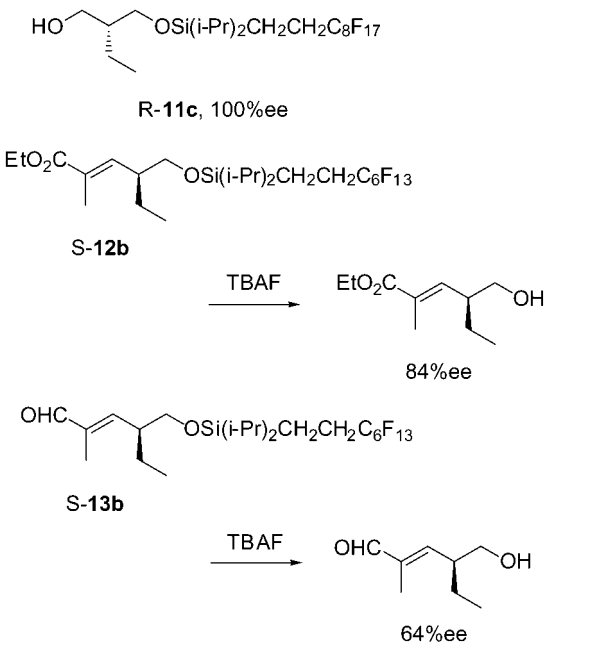
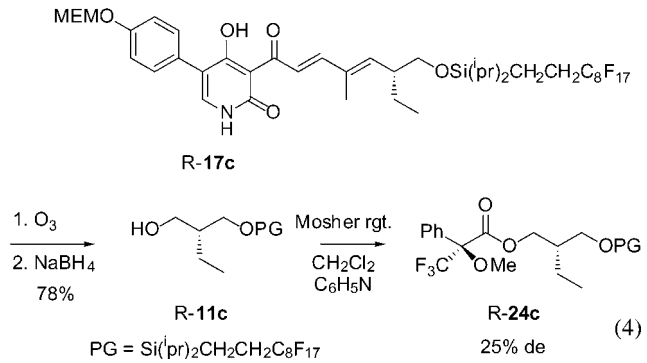


Figure 7. Alcohol precursors for Mosher analysis of pyridovericin intermediates.

sample of about 1 mg of natural pyridovericin, whose rotation was measured in our lab as −17.8 (*c* 0.09, MeOH). With these results, we can confidently assign the absolute configuration of natural pyridovericin as R. Disappointingly, with estimated ee values of about 15%, the samples of **5** prepared from quasiracemic synthesis are a lot closer to racemic than enantiopure.

Unsatisfied with the use of optical rotation to measure enantiopurity, we experimented with several chiral columns to resolve pyridovericin **5** or its immediate protected precursor **17**, but without success. We then resorted to Mosher ester methods (eq 4). The sample of (*R*)-pyridovericin precursor (*R*)-**17c** from

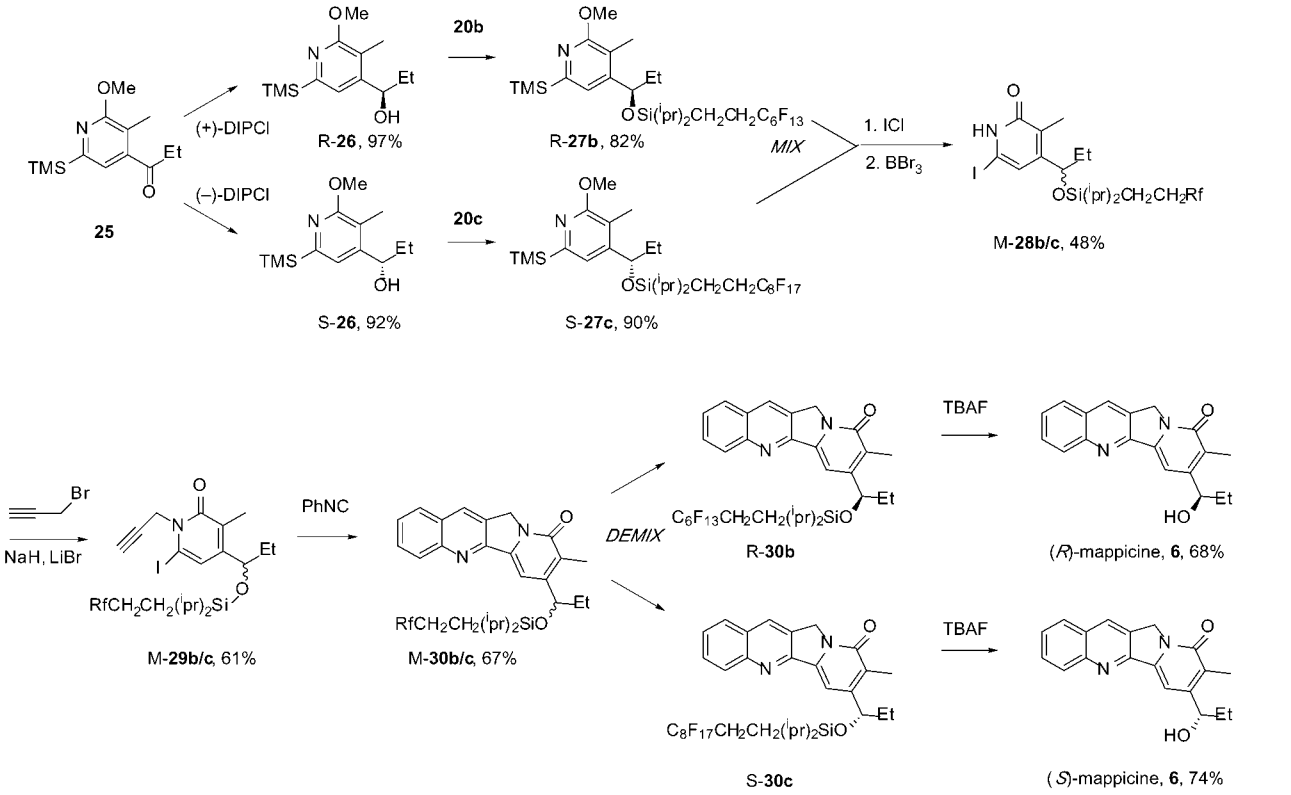


quasiracemic synthesis was degraded by ozonolysis and reduction[31] to regenerate the starting alcohol (*R*)-**11c**. This was converted to its Mosher ester (*R*)-**24c**,[32] whose de value was measured by ^{1}H and ^{19}F NMR to be 25%. The 25% ee determined by derivatization is probably more accurate than the 15% ee from rotation, but the take home message is the same: substantial racemization occurred during the synthesis.

To provide more information on where racemization was occurring, we converted three other intermediates to Mosher

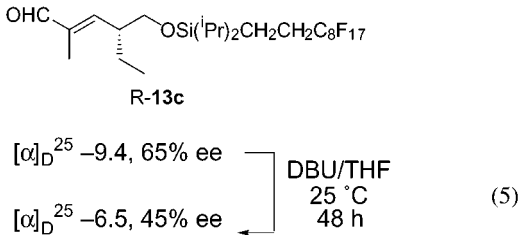
(31) Smith, A. B., III; et al. U.S. Patent 5,789,605.
(32) Takeuchi, Y.; Itoh, N.; Kawahara, S.; Koizumi, T. *Tetrahedron* **1993**, *49*, 1861.

Scheme 2



esters and measured their de values. Racemic samples of each Mosher ester were made to facilitate experiments. The results of these experiments are summarized in Figure 7. Starting alcohol (*R*)-**11c** was derivatized and shown to be enantiopure within the limits of detection. Two steps later, ester (*S*)-**12b** has decreased to 84% ee. An additional two steps provided aldehyde (*S*)-**13b** of only 64% ee. Clearly, the ee loss comes not at one but several steps in the sequence. Additional basic and high-temperature conditions follow in the synthesis, so in retrospect we were fortunate that our final samples were not racemic.

In quasiracemic synthesis, racemization can occur by epimerization as in standard asymmetric synthesis. It can also occur by chemical exchange (scrambling) of the tags from one enantiomer to the other. Thus, both epimerization and tag scrambling must be avoided. We felt that tag scrambling was unlikely and that base-catalyzed (or thermal) epimerization was the likely culprit. To support this hypothesis, we exposed a THF solution of (*R*)-**13c** to DBU for 48 h at 25 °C (eq 5). As



(5)

measured by optical rotation, the ee of the sample dropped from 65% ($[\alpha]_D$ −9.4, *c* 1.0, MeOH) to 45% ($[\alpha]_D$ −6.5, *c* 1.0, MeOH). Thus, epimerization through enolization of **13** and related intermediates probably accounts for the loss of enantiopurity of the tagged quasienantiomers.

The first quasiracemic synthesis only met one of the two initial goals: The absolute configuration of natural pyridovericin **5** was assigned as R by making both enantiomers in a single synthesis, but the final products were not enantiopure (or even highly enriched). To demonstrate that enantiopure products could be made, we turned to our recent asymmetric synthesis of (*S*)-mappicine **6**,[13,14] which was mutated into a quasiracemic synthesis as shown in Scheme 2. Reduction of readily available ketone **25** with (+)-B-chlorodiisopinylcampheylborane ((+)-DIP-chloride)[33] afforded (*R*)-**26** in 97% yield with >98% ee, as reported.[14] This was silylated with the smaller silyl bromide **20b** to give (*R*)-**27b** (82%). Likewise, reduction with (−)-DIP-chloride provided enantiopure (*S*)-**26** (92%), which was silylated with the larger silyl bromide **20c** to give (*S*)-**27c** (90%). These were then mixed in equimolar amounts for the four-step quasiracemic synthesis.

Treatment of M-**27b**/**c** with ICl followed by demethylation with BBr_3 and purification by flash chromatography over standard silica gel gave M-**28b**/**c** in 48% yield. *N*-Propargylation under standard conditions[34] provided the radical precursor M-**29b**/**c** in 61% yield. In turn, this was reacted with phenyl isonitrile under established conditions for the cascade radical annulation to provide the quasiracemic mixture of protected mappicines M-**30b**/**c**.

This mixture was then separated by fluorous chromatography employing the same gradient as above. The two quasi-enantiomers were well separated ((*R*)-**30b**, 18 min; (*S*)-**30c**, 26 min). Six serial injections of about 10 mg each provided 23 mg of (*R*)-**30b** and 25 mg of (*S*)-**30c**. Deprotection of the

(33) Brown, H. C.; Ramachandran, P. V. In *Advances in Asymmetric Synthesis*; Hassner, A., Ed.; Jai Press Inc: Greenwich, CT, 1995; Vol. 1, p 147.
(34) Liu, H.; Ko, S. B.; Josien, H.; Curran, D. P. *Tetrahedron Lett.* **1995**, *36*, 8917.

resolved quasienantiomers provided the natural (−)-mappicine (*S*)-**6** (from (*S*)-**30c**, 74%) and its enantiomer (+)-mappicine (*R*)-**6** (from (*R*)-**30b**, 68%). The final products (*R*)/(*S*)-**6** from the quasiracemic synthesis were analyzed by chiral HPLC on a ChiralCel OD column and found to be enantiopure within the limits of detection (>98% ee).[11b] Unnatural (*R*)-mappicine has been isolated previously by resolution,[11b,11g] and this is its first asymmetric synthesis.

Conclusions

These straightforward syntheses of pyridovericin **5** and mappicine **6** show that quasiracemic synthesis has value as a complement to both racemic and asymmetric synthesis. Like racemic synthesis, both enantiomers are produced in a single set of steps, yet separation and identification are guaranteed by the tags. Like asymmetric synthesis, the preparation of enantiopure precursors is required at some stage, and racemization, either by epimerization or tag exchange, must be avoided. Effort is economized by tagging early in the synthesis and detagging late. A fluorous quasiracemic synthesis should be considered whenever both enantiomers are needed for structure identification, biological testing, or any other end. Fluorous quasiracemic synthesis is readily scaleable to make larger quantities, and the limitation at demixing is quickly receding as larger fluorous columns become available.

Conveniently, fluorous quasienantiomers behave like true enantiomers when subjected to the typical spectroscopic analyses (^{1}H and ^{13}C NMR) and chromatographic techniques (flash chromatography) used in modern synthesis. The spectroscopic similarities were expected since the tag is remote from the portion of the molecule being analyzed. But the chromatographic similarities were a pleasant outcome. Adding CF_2 groups decreases the polarity of molecules, so we were concerned at the outset that quasienantiomers might separate on regular silica gel. Whether they do or not may be a function of structure, sorbent, and conditions, so more experience is needed before the lack of separation can be generalized. At any time, the quasiracemic mixture can be resolved based on the fluorous tag by fluorous chromatography. The large retention time differences in this work show that the incremental change of tags by two CF_2 groups is unneeded; tags differing by one CF_2 group should suffice. But even-numbered perfluoroalkyl chains are currently less expensive, so the incremental change of two may be preferred.

Can other tags besides fluorous tags be used for quasiracemic synthesis? Yes, without doubt. The practice of tagging molecules for strategic separation purposes has become increasingly common in recent years, and a number of practical strategies have emerged.[3] Extending any one of these strategies for use in quasiracemic synthesis requires the development of two different kinds of tags that ideally behave the same under typical synthetic reaction and separation conditions, but have at least one "tag specific" reaction or separation that can be used at will to differentiate tagged molecules.

In the larger picture of solution phase mixture synthesis aided by separation tags,[14] quasiracemic synthesis can be viewed as the simplest technique: only two compounds are mixed and prior to tagging the two compounds are enantiomers. Since enantiomers have identical separation behavior under achiral conditions, it is not difficult to design tags that will dominate when paired with a complementary separation technique. Mixing more compounds involves tagging diastereomers or even nonisomers, so the tag dominance becomes more challenging. We have already communicated that fluorous mixture synthesis can be used to make and separate nonisomeric analogues of mappicine,[16] and we will be reporting in detail on this and other endeavors soon.

The classical synthesis of racemic mixtures beautifully illustrates the problems of mixture synthesis: the individual enantiomeric components of the mixture are not reliably analyzed, separated, or identified on demand. The use of separation tags in quasiracemic synthesis and other more advanced techniques[16] provides a conceptual foundation to approach problems of mixture synthesis[35] when pure final products are desired. And fluorous tagging provides the first practical solution to the experimental problems posed by mixture synthesis.

Acknowledgment. We thank the National Institutes of Health for funding of this work. We also thank Dr. A. Nakagawa for providing samples of pyridovericin and Professor James Rigby for providing experimental details.

Supporting Information Available: Complete experimental data for all new compounds reported in this paper (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

JA025606X

(35) Houghten, R. A.; Pinilla, C.; Appel, J. R.; Blondelle, S. E.; Dooley, C. T.; Eichler, J.; Nefzi, A.; Ostresh, J. M. *J. Med. Chem.* **1999**, *42*, 3743.