Pergamon

Tetrahedron Letters 43 (2002) 8621–8623

TETRAHEDRON LETTERS

syn-Selective additions to Garner aldehyde: synthesis of a potent glucosylceramide synthase inhibitor

Arifa Husain and Bruce Ganem*

Department of Chemistry and Chemical Biology, Baker Laboratory, Cornell University, Ithaca, NY 14853-1301, USA

Received 22 July 2002; accepted 27 August 2002

Abstract—Highly *syn*-selective additions of aryl Grignard reagents to Garner aldehyde **5** are reported, making possible a practical, asymmetric synthesis of the potent glucosylceramide synthase inhibitor **3**.

Fabry disease is an inherited, X-linked inborn error of glycolipid catabolism resulting in a defective lysozomal α-galactosidase A in tissues of affected males.[1] As in other lysozomal storage disorders (Gaucher, Sandhoff, Tay-Sachs diseases), the enzymatic defect leads to an accumulation in most visceral tissues of precursor glycosphingolipids. The current treatment of Gaucher disease involves costly intravenous infusions of a replacement enzyme.[2] A similar strategy for treating Fabry disease using infusions of α-galactosidase A has recently been reported.[3]

An alternative approach to treatment, known as 'substrate deprivation', involves inhibiting an enzyme in the biosynthesis of the accumulating glycosphingolipid, specifically glucosylceramide synthase, which catalyzes the first committed step of glycosphingolipid biosynthesis.[4] Promising results using this strategy have been reported with *N*-butyldeoxynojirimycin (NBDNJ), a compound originally developed as an α-glucosidase inhibitor.[5] However, NBDNJ-treated mice experienced weight loss as well as a decrease in both the size and function of spleen and thymus.[6]

In 1992, Abe et al. identified D-*threo*-1-phenyl-2-decanoylamino-3-morpholino-1-propanol (PDMP; **1** in Fig. 1) as a new type of glucosylceramide synthase inhibitor.[7] Since then, more active sphingosine-like analogs of PDMP have been developed by modifying the cyclic amine,[8] the fatty acid amide[9] and the benzene ring[10] of **1**. Such modifications led to much more potent PDMP analogs **2**–**4**. Compounds **3**–**4** are 2000-fold more active than either NBDNJ or PDMP, and markedly deplete globotriaosylceramide, the glycolipid that accumulates in virally transformed lymphoblasts from a patient with Fabry disease, with no impairment of cell growth.[11] Here we report a method for the *syn*-selective addition of aryl Grignard reagents to the well-known Garner aldehyde, and illustrate its utility with a practical synthesis of inhibitor **3**.

We designed our approach around the stereoselective addition of aryl metal compounds to aldehyde **5** (Scheme 1) hoping to obtain *syn*-adducts **6**, which were envisioned as precursors of **2**–**4** via intermediates **7** (Scheme 1). Our goal was to improve on prior work with **5** and its congeners.

In an earlier synthesis of **1** (eight steps, 14% overall), Mitchell et al. were successful in achieving predominantly *syn*-addition (8:1, 70% yield) of PhMgBr to an

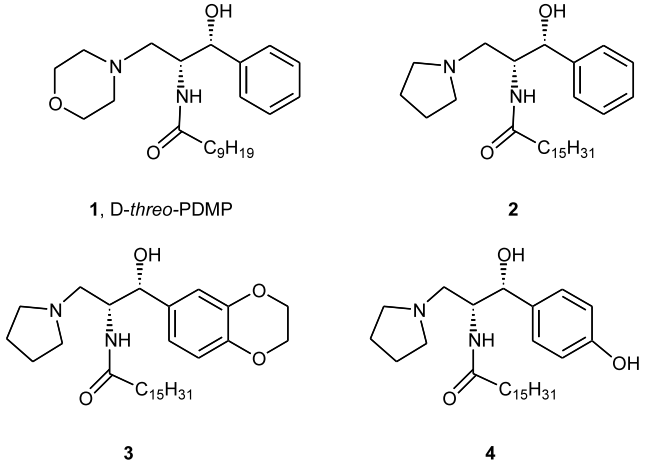
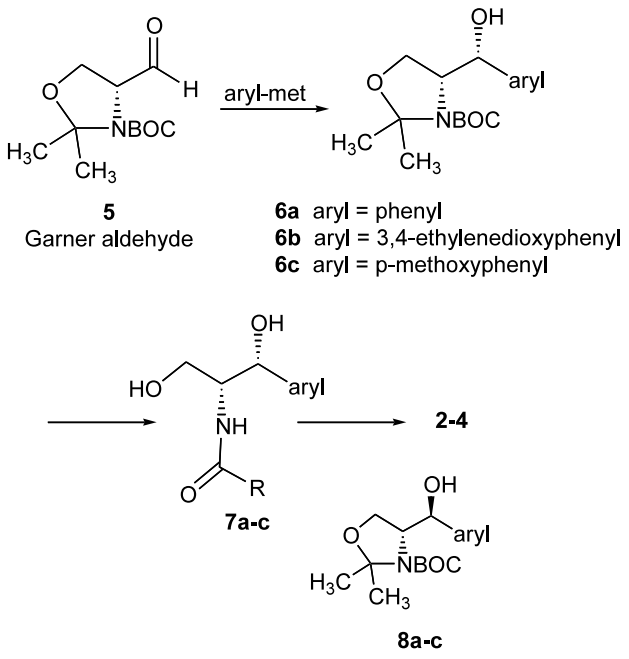


Figure 1.

* Corresponding author.

0040-4039/02/$ - see front matter
PII: S0040-4039(02)02096-8



Scheme 1.

O-protected serine aldehyde as its benzophenone Schiff base.[8c] By contrast, Nishida et al. found that *anti*-addition prevailed in reactions of PhLi and PhMgBr with **5** under various conditions. The *syn*-adduct **6a** was only slightly favored (1.1:1 **6a**:**8a**) in the reaction of PhMgBr with **5** in ether using excess $ZnBr_2$. Moreover, separation of the isomers by chromatography proved difficult.[12,13] Subsequently, an exhaustive review of organometallic additions to **5** summarizing the effects of metal, solvent, and added Lewis acid catalyst was published, confirming that substantial quantities of the undesired *anti*-product **8** were formed in most arylmetal addition reactions.[14] Using C_6H_5Li and C_6H_5MgBr, for example, ratios of **6a**:**8a** ranged from 1:5 to 1:3.[15]

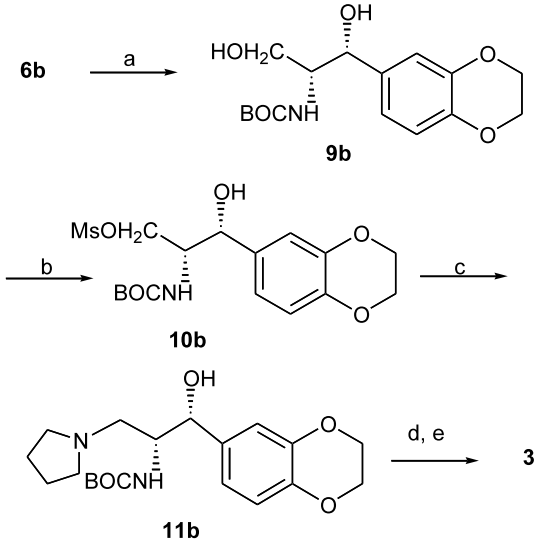
Those results notwithstanding, two promising findings led us to investigate the reaction of **5** with aryl Grignard reagents mediated by CuI–Me_2S. In one report, the reaction of trimethylsilylethynylmagnesium bromide with **5** in the presence of CuI using THF–Me_2S as solvent afforded the expected carbinol in 95% yield, with a *syn*:*anti* ratio of 20:1.[16] In a separate study, predominantly *syn*-addition of allylmagnesium bromide to **5** was achieved under similar conditions (96% yield, 3:1 *syn*:*anti*).[17]

Encouraged by those results, we investigated the additions of phenyl, 3,4-ethylenedioxyphenyl,[18] and *p*-methoxyphenylmagnesium bromides with **5**. Optimal results were obtained by dropwise addition of the appropriate Grignard reagent (2 equiv., based on **5**) to a −78°C mixture of CuI (3 equiv.) in 5:1 THF:Me_2S. The −78°C bath was replaced with a −35°C bath and the mixture was stirred at −40 to −35°C for 30 min and then recooled to −78°C. A solution of **5** (1 equiv.) in THF was then added over 30 min via syringe pump at −78°C. The mixture was warmed to rt with stirring for 12 h. In each case, the desired adduct was obtained in good yield (68% for **6a**, 74% for **6b**; 64% for **6c**) and with 20:1 *syn*:*anti* stereoselectivity.[19] In each case, the identity of the *syn*-product was established by comparison of NMR spectral data with authentic samples of both stereoisomers.

The utility of this *syn*-selective addition reaction was illustrated by an enantioselective synthesis of the potent glucosylceramide synthase inhibitor **3** (Scheme 2). Compound **3** had previously been prepared from ethylenedioxyphenacyl bromide as an unspecified mixture of *syn* and *anti* racemic forms.[10] Besides being enantioselective, the new route also circumvented several protection and deprotection steps reported in earlier syntheses of PDMP and its analogs.[8c,12]

To synthesize **3**, the acetonide protecting group in **6b** was removed using 0.1N HCl,[12] affording **9b** in 82% yield.[20a] The minor amounts of *anti*-stereoisomer **8b** proved resistant to acid hydrolysis, and was readily removed from **9b** by chromatographic purification at this stage. Selective mesylation of **9b** afforded **10b** in 85% yield. The *N*-alkylation of pyrrolidine with **10b** led to **11b** (58%).[20b] Acid-catalyzed deprotection of the BOC group, then *N*-acylation using palmitoyl chloride ($C_{15}H_{31}COCl$) in a modification of the published procedure[12] led to the desired target **3** in 87%.[20c] The transformation of **5** into **3** was accomplished in six steps and 26% overall yield.

The short, enantioselective synthesis of inhibitor **3** reported here should be readily adaptable to scale-up, as well as to the synthesis of other glucosylceramide synthase inhibitors for clinical development. The versatility of the route should also make it possible to construct a wide array of functionalized analogs and derivatives, which may be of interest as glycolipid



Scheme 2. *Reagents and conditions*: (a) 0.1N HCl, THF; (b) MsCl, Et_3N (1 equiv. each), CH_2Cl_2, 0°C; (c) pyrrolidine (5 equiv.) DMF, 45°C; (d) 3N HCl, 0°C to rt; (e) $C_{15}H_{31}COCl$ (0.95 equiv.) Et_3N (2 equiv.), DMAP (cat), CH_2Cl_2, −20°C to rt.

inhibitors find new applications in the fields of insulin sensitization,[21] inhibition of metastasis,[22] multidrug resistance,[23] antigen presentation,[24] and antifungal chemotherapy.[25]

Acknowledgements

This work was supported by the National Institutes of Health (DK 55823). Funding for major instrumentation in the Cornell NMR Facility has been provided by NSF (CHE 7904825; PGM 8018643) and NIH (RR02002).

References

1. Desnick, R. J.; Ioannou, Y. A.; Eng, C. M. In *The Metabolic and Molecular Bases of Inherited Disease*; Scriver, C. R., Ed. α-Galactosidase A Deficiency: Fabry Disease; McGraw-Hill, 1995; pp. 2741–2784.
2. Beutler, E. *Curr. Opin. Hematol.* **1997**, *4*, 19–23.
3. Schiffmann, R.; Murray, G. J.; Treco, D. *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 365–370.
4. Radin, N. *Prog. Clin. Biol.* **1982**, *95*, 357–383.
5. Ratner, L.; vander Heyden, N.; Dedera, D. *Virology* **1991**, *181*, 180–192.
6. Platt, F. M.; Reinkensmeier, G.; Dwek, R. A.; Butters, T. D. *J. Biol. Chem.* **1997**, *272*, 19365–19372.
7. Abe, A.; Inokuchi, J.; Jimbo, M.; Shimeno, H.; Nagamatsu, A.; Shayman, J. A.; Shukla, G. S.; Radin, N. S. *J. Biochem. (Tokyo)* **1992**, *111*, 191–196.
8. (a) Carson, K. G.; Ganem, B.; Radin, N. S.; Abe, A.; Shayman, J. A. *Tetrahedron Lett.* **1994**, *35*, 2659–2663; (b) Abe, A.; Radin, N. S.; Shayman, J. A.; Wotring, L. L.; Zipkin, R. E.; Sivakumar, R.; Ruggieri, J. M.; Carson, K. G.; Ganem, B. *J. Lipid Res.* **1995**, *36*, 611–621; (c) Mitchell, S. A.; Oates, B. D.; Razavi, H.; Polt, R. *J. Org. Chem.* **1998**, *63*, 8837–8842.
9. Shayman, J. A.; Lee, L.; Abe, A.; Shu, L. *Methods Enzymol.* **2000**, *311*, 373–387.
10. Lee, L.; Abe, A.; Shayman, J. A. *J. Biol. Chem.* **1999**, *274*, 14662–14669.
11. (a) Abe, A.; Arend, L. J.; Lee, L.; Lingwood, C.; Brady, R. O.; Shayman, J. A. *Kidney Int.* **2000**, *57*, 446–454; (b) Abe, A.; Gregory, S.; Lee, L.; Killen, P. D.; Brady, R. O.; Kulkarni, A.; Shayman, J. A. *J. Clin. Invest.* **2000**, *105*, 1563–1571.
12. Nishida, A.; Sorimachi, H.; Iwaida, M.; Matsumizu, M.; Kawate, T.; Nakagawa, M. *Synthesis* **1998**, 389–390.
13. An alternative route to D-*threo*-**1** was recently achieved by stereoselective reduction of the corresponding arylketone: Jimbo, M.; Yamagishi, K.; Yamaki, T.; Nunomura, K.; Kabayama, K.; Igarashi, Y.; Inokuchi, J. *J. Biochem.* **2000**, *127*, 485–491.
14. Mengel, A.; Reiser, O. *Chem. Rev.* **1999**, *99*, 1191–1223.
15. Radunz, H.-E.; Devant, R. M.; Eiermann, V. *Liebigs Ann. Chem.* **1988**, 1103–1105.
16. Herold, P. *Helv. Chim. Acta* **1988**, *71*, 354–362.
17. Bayquen, A. V.; Read, R. W. *Tetrahedron* **1996**, *52*, 13467–13482.
18. This Grignard reagent was prepared in standard fashion from the corresponding bromide: Mattson, R. J.; Sloan, C. P.; Lockhart, C. C.; Catt, J. D.; Gao, Q.; Huang, S. *J. Org. Chem.* **1999**, *64*, 8004–8007.
19. For **6a**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.45–7.26 (m, 5H), 4.83–4.63 (d, $J=9.1$ Hz, 1H), 4.33–4.07 (m, 1H), 3.81–3.49 (m, 2H), 1.60–1.44 (m, 15H); for **6b**: δ 6.95–6.74 (m, 3H), 4.70–4.48 (d, $J=8.6$ Hz, 1H), 4.21 (s, 4H), 4.14–4.07 (m, 1H), 3.79–3.46 (m, 2H), 1.63–1.40 (m, 15H); for **6c**: δ 7.34–7.23 (d, $J=8.0$ Hz, 2H), 6.91–6.78 (d, $J=8.6$ Hz, 2H), 4.75–4.59 (d, $J=9.1$ Hz, 1H), 4.22–4.09 (m, 1H), 3.79 (s, 3H), 3.74–3.51 (m, 2H), 1.57–1.45 (m, 15H).
20. (a) For **8b**: $[\alpha]_D^{20}$ −11.4° (*c* 0.27, $CHCl_3$); ^{13}C NMR δ 156.9, 143.4, 143.1, 135.2, 119.4, 117.2, 115.4, 80.0, 73.0, 64.5, 63.3, 57.4, 28.5; (b) for **11b**: $[\alpha]_D^{20}$ +8.0° (*c* 0.39, $CHCl_3$); ^{13}C NMR δ 156.0, 143.5, 142.9, 135.0, 119.4, 117.0, 115.5, 79.5, 74.5, 64.5, 57.7, 55.0, 53.3, 46.1, 28.5, 23.7; (c) for **3**: $[\alpha]_D^{20}$ +3.0° (*c* 0.13, $CHCl_3$); ^{13}C NMR δ 174.1, 143.6, 143.1, 134.5, 119.2, 117.3, 115.3, 76.9, 74.3, 64.6, 57.3, 55.1, 52.5, 46.2, 36.9, 32.2, 29.9, 29.8, 29.7, 29.6, 29.5, 29.4, 25.9, 23.8, 22.9, 14.4.
21. Grigsby, R. J.; Dobrowsky, R. T. *Biochem. Biophys. Res. Commun.* **2001**, *287*, 1121–1124.
22. Inokuchi, J.; Jimbo, M.; Momosaki, K.; Shimeno, H.; Nagamatsu, A.; Radin, N. S. *Cancer Res.* **1990**, *50*, 6731–6737.
23. Sietsma, H.; Veldman, R. J.; Kok, J. W. *J. Membrane Biol.* **2001**, *181*, 153–162.
24. Cheng, P. C.; Dykstra, M. L.; Mitchell, R. N.; Pierce, S. K. *J. Exp. Med.* **1999**, *190*, 1549–1560.
25. (a) Mandala, S. M.; Harris, G. H. *Methods Enzymol.* **2000**, *311*, 335–348; (b) Georgopapadakou, N. H. *Exp. Opin. Invest. Drugs* **2000**, *9*, 1787–1796.