JOC The Journal of Organic Chemistry

Article

pubs.acs.org/joc

Synthesis and Evaluation of Eight- and Four-Membered Iminosugar Analogues as Inhibitors of Testicular Ceramide-Specific Glucosyltransferase, Testicular β-Glucosidase 2, and Other Glycosidases

Jae Chul Lee,[†,§] Subhashree Francis,[†,§] Dinah Dutta,[+] Vijayalaxmi Gupta,[‡,§] Yan Yang,[#] Jin-Yi Zhu,[#] Joseph S. Tash,[‡,§] Ernst Schönbrunn,[#] and Gunda I. Georg*[,†,§]

[†]Department of Medicinal Chemistry, Institute for Therapeutics Discovery and Development, University of Minnesota, Minneapolis, Minnesota 55455, United States

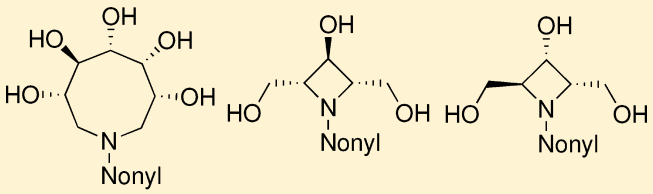
[+]Department of Medicinal Chemistry, University of Kansas, Lawrence, Kansas 66045, United States

[‡]Department of Molecular & Integrative Physiology and [§]Interdisciplinary Center For Male Contraceptive Research and Drug Development, University of Kansas Medical Center, Kansas City, Kansas 66160, United States

[#]Drug Discovery Department, Moffitt Cancer Center, 12902 Magnolia Drive, Tampa, Florida 33612, United States

Supporting Information

ABSTRACT: Eight- and four-membered analogues of *N*-butyldeoxynojirimycin (NB-DNJ), a reversible male contraceptive in mice, were prepared and tested. A chiral pool approach was used for the synthesis of the target compounds. Key steps for the synthesis of the eight-membered analogues involve ring-closing metathesis and Sharpless asymmetric dihydroxylation and for the four-membered analogues Sharpless epoxidation, epoxide ring-opening (azide), and Mitsunobu reaction to form the four-membered ring. (3*S*,4*R*,5*S*,6*R*,7*R*)-1-Nonylazocane-3,4,5,6,7-pentaol (**6**) was moderately active against rat-derived ceramide-specific glucosyltransferase, and four of the other eight-membered analogues were weakly active against rat-derived β-glucosidase 2. Among the four-membered analogues, ((2*R*,3*S*,4*S*)-3-hydroxy-1-nonylazetidine-2,4-diyl)dimethanol (**25**) displayed selective inhibitory activity against mouse-derived ceramide-specific glucosyltransferase and was about half as potent as NB-DNJ against the rat-derived enzyme. ((2*S*,4*S*)-3-Hydroxy-1-nonylazetidine-2,4-diyl)dimethanol (**27**) was found to be a selective inhibitor of β-glucosidase 2, with potency similar to NB-DNJ. Additional glycosidase assays were performed to identify potential other therapeutic applications. The eight-membered iminosugars exhibited specificity for almond-derived β-glucosidase, and the 1-nonylazetidine **25** inhibited α-glucosidase (*Saccharomyces cerevisiae*) with an IC_{50} of 600 nM and β-glucosidase (almond) with an IC_{50} of 20 μM. Only *N*-nonyl derivatives were active, emphasizing the importance of a long lipophilic side chain for inhibitory activity of the analogues studied.



INTRODUCTION

Hormonal male contraceptive agents are currently in clinical trials, but have not yet reached the market due to side effects and pharmacokinetic issues.[1] The discovery and development of nonhormonal contraceptive agents is another approach toward male contraception.[2] Nonhormonal experimental agents such as gossypol[3] and α-chlorohydrin[4] have been studied, but they are neither safe nor effective enough for human use. Among newer nonhormonal contraceptive lead compounds,[5] the alkylated iminosugar *N*-butyldeoxynojirimycin (NB-DNJ, zavesca) has been reported to be an effective, reversible, and nontoxic oral male contraceptive agent in mice (Figure 1).[6] NB-DNJ is in clinical use for the treatment of mild-to-moderate type 1 Gaucher's disease in adult patients who cannot be treated with enzyme replacement therapy (ERT).[7]

The iminosugar NB-DNJ (Figure 1) is an inhibitor of ceramide-specific glucosyl transferase[8] and β-glucosidase 2, which are key enzymes (Figure 1 and Table 1) in the biosynthesis of glycosphingolipids.[9,10] Inhibition of these enzymes leads to an imbalance of testicular glucosylceramide levels, which is believed to impair spermatogenesis. The effect of NB-DNJ on spermatogenesis was found to be species- and strain-specific.[11] NB-DNJ is active in C57B1/6J-related mouse strains but not in other mouse strains or in rabbits. Although NB-DNJ also showed no discernible effects on human spermatogenesis,[12] we hypothesized that analogues of NB-DNJ with higher potency and or differential enzyme selective inhibitory activity could be discovered that would affect spermatogenesis in mammalian species other than C57B1/6J-related mouse strains, including man.

Received: October 9, 2011
Published: March 20, 2012

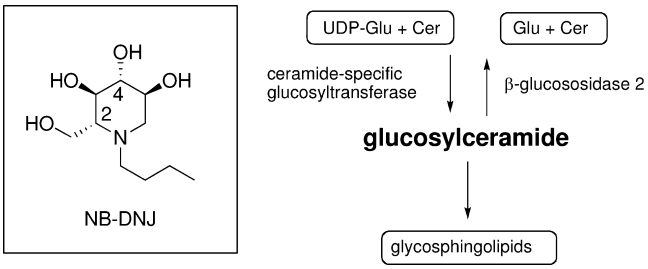
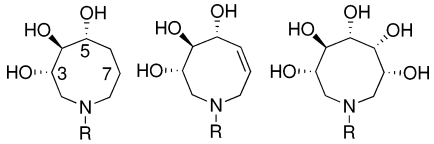


Figure 1. Structure of *N*-butyldeoxynojirimycin (NB-DNJ) and metabolism of glucosylceramide.

Five-, six-, and seven-membered NB-DNJ analogues[13] have been reported as inhibitors for ceramide-specific glucosyltransferase, but none of them are significantly better inhibitors than NB-DNJ except *N*-alkyloxyalkyl DNJ analogues[14] including adamantane-DNJ conjugates.[15] Six- and seven-membered ring DNJ analogues have also been investigated for pharmacological chaperoning to improve protein folding and trafficking defects in Gaucher's disease.[16] More potent analogues and derivatives effective in all mammalian species could possibly be obtained by structural changes of the parent compound, which includes ring-enlarged or ring-contracted analogues related to NB-DNJ such as azocane analogues (eight-membered ring) and azetidine analogues (four-membered ring). A series of azetidine analogues were recently reported as inhibitors of various glycosidases,[17] but they were not tested against ceramide-specific glucosyltransferase. We report herein the design and synthesis of novel eight-membered and four-membered iminosugar analogues and the evaluation of their inhibitory potencies for testicular ceramide-specific glucosyltransferase, testicular β-glucosidase 2, and other glycosidase enzymes. The key synthetic steps for the synthesis of the eight-membered analogues involved a ring-closing metathesis and a Sharpless asymmetric dihydroxylation. The four-membered iminosugars were prepared from L-gulono-1,4-lactone or 1,2:5,6-diisopropylidene-D-mannitol, employing as the key steps a Sharpless epoxidation and a Mitsunobu reaction for ring formation.

RESULTS AND DISCUSSION

Design and Synthesis of Eight-Membered Iminosugars. Molecular models of eight-membered iminosugars **2**, **4**, and **6** (Figure 2) were generated using Maestro (Schrödinger).



1 R = butyl
2 R = nonyl
3 R = butyl
4 R = nonyl
5 R = butyl
6 R = nonyl

Figure 2. Structures of designed eight-membered iminosugar analogues **1**–**6**.

The ceramide structure was prepared on the basis of the crystal structure of galactosylceramide.[13a,18]

Figure 3a shows the overlay of NB-DNJ and ceramide.[13a] Overlay of compound **2** with NB-DNJ (Figure 3b) demonstrates that the 4-hydroxyl, 3-hydroxyl, and 2-hydroxymethyl groups of NB-DNJ have a similar orientation as the 5-hydroxyl, 4-hydroxyl, and 3-hydroxyl groups of compound **2**, respectively. The introduction of a double bond into the 8-membered ring slightly changes the ring conformation (compounds **3** and **4**). Nevertheless, overlay of compound **4** with NB-DNJ (Figure 3c) was very similar to that of compound **2**. Next, two additional hydroxyl groups were added to the ring (compounds **5** and **6**) and then assessed for structural similarity to NB-DNJ. In the overlay of compound **6** with NB-DNJ (Figure 3d), the 7-hydroxyl group of compound **6** is close to the 5-hydroxyl group of NB-DNJ. The 6-hydroxyl group of **6**, however, does not overlay with any of the hydroxyl groups in NB-DNJ. The modeling study reveals that the six designed eight-membered iminosugars possess structural similarities with NB-DNJ and therefore could be expected to be inhibitors of the targeted enzymes.

The retrosynthetic approach for the synthesis of the eight-membered iminosugars is outlined in Scheme 1. The six target compounds would be prepared from common intermediate **7**, which could be formed through a ring-closing metathesis of diene **8**. Diene **8** would be generated by a reductive amination of aldehyde **9** with allylamine.

Compound **9** was prepared as shown in Scheme 2, utilizing known procedures.[19,20] The primary hydroxyl group of methyl-α-D-glucopyranoside (**10**) was protected as its TBS ether **11**. Benzylation of intermediate **11** provided the fully protected compound **12**, which was desilylated with TBAF to afford the primary alcohol **13**. Iodination of alcohol **13** was performed with iodine and triphenylphosphine to provide the iodo intermediate **14**, which underwent reductive ring-opening with activated zinc under sonication conditions[21] to furnish aldehyde **9** in good yield.

The synthesis of the eight-membered ring was performed as shown in Scheme 3. Reductive amination[22] of aldehyde **9** with allylamine using $NaBH(OAc)_3$ afforded diene **15**. The secondary amine of the diene was protected with a tosyl group to yield compound **8a**. Ring closing metathesis (RCM)[23] was then performed using the Grubbs II catalyst to obtain the eight-membered ring **7a**.

The amino group of intermediate **15** was also protected with a Boc group to provide compound **8b** (Scheme 4) although in a slightly lower yield than the *N*-tosylation reaction of **15**. RCM reaction of **8b** yielded compound **7b** as a mixture of two Boc rotamers in a ratio of 1:1.4. This reaction provided the targeted compound **7b** in slightly better yield than the reaction of *N*-tosyl derivative **8a** to form RCM product **7a**.

With key intermediates **7a** and **7b** in hand, the trihydroxy compounds **1**–**4** were obtained as shown in Scheme 5. The tosyl group of compound **7a** was cleaved using Na and naphthalene to provide secondary amine **16**.[24] Amine **16** was also prepared from intermediate **7b** by removal of the Boc group. Removal of the Boc group provided a slightly higher yield than the deprotection of the tosyl group. Comparing the two protecting groups in this reaction sequence reveals that the *N*-tosyl and *N*-Boc protecting groups lead to the same overall yield for the synthesis of intermediate **16** from **15**. Reductive alkylation of the secondary amine[22] **16** was carried out next, using butyraldehyde and nonyl aldehyde to afford compounds **17a** and **17b**, respectively. Debenzylation and double bond reduction were achieved with hydrogen gas, using palladium(II) chloride as the catalyst, to obtain target compounds **1** and **2**. Reductive debenzylation[25] of compounds **17a** and **17b** was performed using Li and naphthalene to provide the trihydroxy derivatives **3** and **4**.

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082–3098

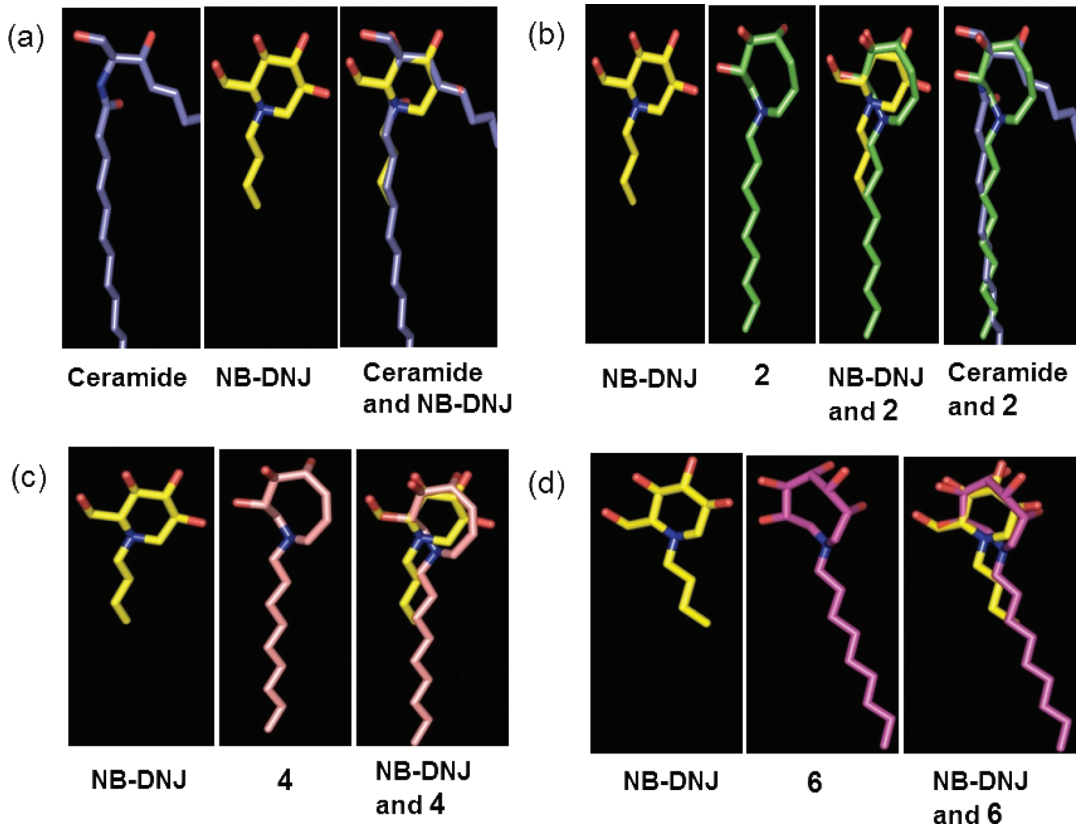
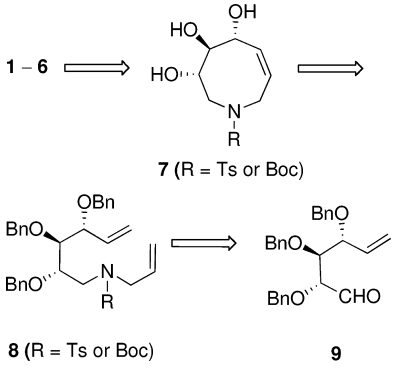


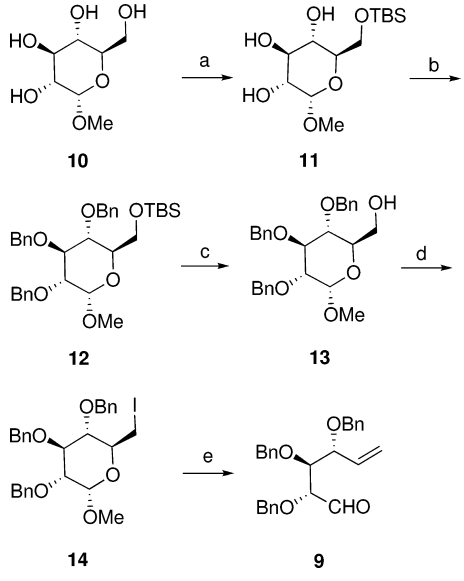
Figure 3. Molecular modeling of ceramide, NB-DNJ, and designed compounds **2**, **4**, and **6**.

Scheme 1



The synthesis of pentahydroxy derivatives **5** and **6** was also achieved from common intermediate **7a** (Scheme 6). First, the *cis*-diol moiety was introduced into compound **7a** by a catalytic Sharpless *cis*-asymmetric dihydroxylation[26] to provide diol **18**. A nonasymmetric dihydroxylation was also performed using OsO_4, but the reaction provided an inseparable mixture of reaction products. Diol **18** was then benzylated to form pentabenzyl ether **19**. Benzylation of **18** provided advantages such as clean reactions and ease of purification in the following reaction steps. Reductive detosylation of **19** gave secondary amine **20**. Reductive alkylation of the secondary amine was performed using butyraldehyde and nonyl aldehyde to furnish compounds **21a** and **21b** respectively. Finally, hydrogenolysis of compounds **21a** and **21b** afforded the corresponding pentahydroxyazocanes **5** and **6**. The structure of pentahydroxyazocane **6** was confirmed by single-crystal X-ray crystallography (Figure 4).

Scheme 2[a]



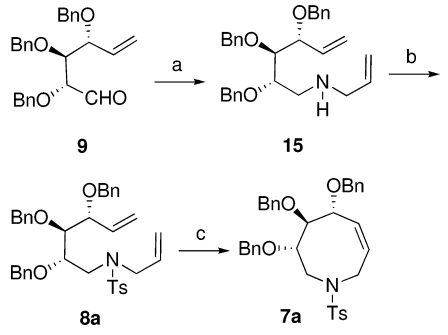
[a]Reagents and conditions: (a) TBSCl, imidazole, DMF, 0 °C to rt, 16 h, 72%; (b) NaH, benzyl bromide, DMF, rt, 17 h, 72%; (c) TBAF, THF, rt, 16 h, 99%; (d) I_2, PPh_3, imidazole, toluene, 70 °C, 3 h, 94%; (e) Zn, THF/H_2O, sonication at 40 °C, 2 h, 93%.

Design and Synthesis of Four-Membered Iminosugar Analogues. Six four-membered iminosugars, **22**–**27**, were designed and synthesized (Figure 5).

As shown in Figure 6, using Maestro (Schrödinger), NB-DNJ was overlaid with compounds **23**, **25**, and **27** (parts a-c, respectively, of Figure 6). Compound **25** was also overlaid with

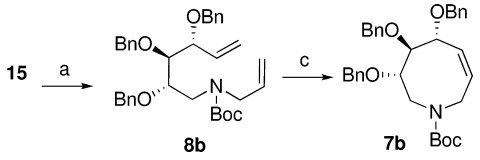
dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082–3098

Scheme 3[a]



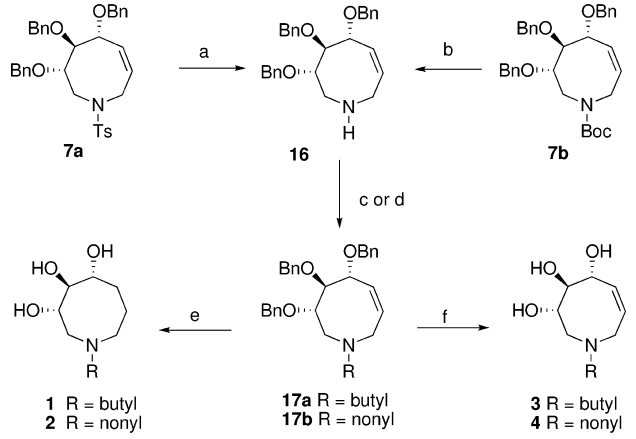
[a]Reagents and conditions: (a) allylamine, $NaBH(OAc)_3$, $ClCH_2CH_2Cl$, rt, 16 h, 86%; (b) TsCl, TEA, DMAP, CH_2Cl_2, rt, 3 h, 84%; (c) Grubbs II catalyst, CH_2Cl_2, reflux, 1 h, 84%.

Scheme 4[a]



[a]Reagents and conditions: (a) Boc anhydride, DMAP, CH_2Cl_2, rt, 16 h, 80%; (b) Grubbs II catalyst, CH_2Cl_2, reflux, 4 h, 92%.

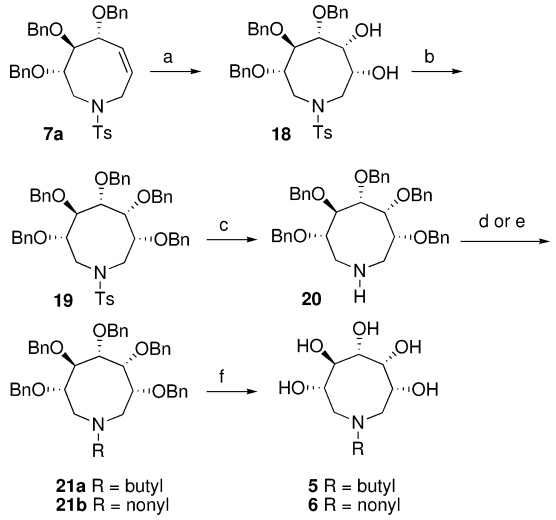
Scheme 5[a]



[a]Reagents and conditions: (a) Na, naphthalene, DME, −78 °C, 30 min, 75%; (b) 4 N HCl in dioxane, rt, 1 h, 85%; (c) butyraldehyde, $NaBH(OAc)_3$, $ClCH_2CH_2Cl$, rt, 16 h, 75%; (d) nonyl aldehyde, $NaBH(OAc)_3$, $ClCH_2CH_2Cl$, rt, 16 h, 76%; (e) $PdCl_2$, MeOH, H_2, rt, 16 h, 76% for **1**, 78% for **2**; (f) Li, liquid NH_3, THF, −78 °C, 1 h, 59% for **3**, 66% for **4**.

ceramide (Figure 6d). The 2-hydroxymethyl, 3-hydroxyl, and 4-hydroxymethyl groups of compound **23** align well with the 4-hydroxyl, 3-hydroxyl, and 2-hydroxymethyl groups of NB-DNJ (Figure 6a). As for compound **25**, only the 4-hydroxymethyl group was not aligned with NB-DNJ. Overlay of NB-DNJ with compound **27** shows that the 2-hydroxymethyl and the 3-hydroxyl groups are close to the 4-hydroxyl and 3-hydroxyl groups of NB-DNJ, respectively. Compound **25** aligned very well with ceramide (Figure 6d, *N*-acyl chain, N, C2, C3, and the 3-hydroxyl group). Compound **23** aligned well with ceramide similar to compound **25** (picture not shown). The only difference between compounds **23** and **25** is the stereochemistry at

Scheme 6[a]



[a]Reagents and conditions: (a) $(DHQ)_2$-PHAL, $K_2OsO_2(OH)_4$, K_2CO_3, $K_3(FeCN)_6$, $CH_3SO_2NH_2$,THF/*t*-BuOH/H_2O, rt, 40 h, 82%; (b) NaH, BnBr, DMF, rt, 15 h, 87%; (c) Na, naphthalene, DME, −78 °C, 30 min, 72%; (d) butyraldehyde, $NaBH(OAc)_3$, $ClCH_2CH_2Cl$, rt, 16 h, 79%; (e) nonyl aldehyde, $NaBH(OAc)_3$, $ClCH_2CH_2Cl$, rt, 16 h, 84%; (f) $PdCl_2$, MeOH, H_2, rt, 18 h, 80% for **5**, 74% for **6**.

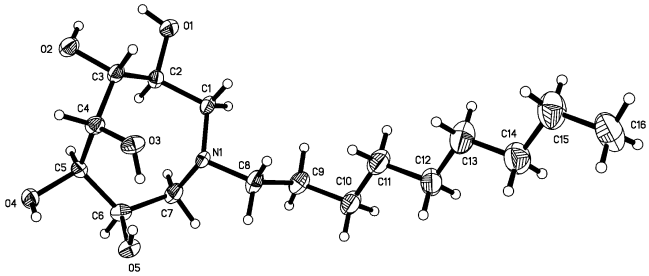


Figure 4. X-ray structure of pentahydroxyazocane **6**.

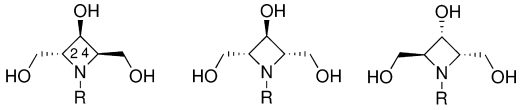


Figure 5. Structures of designed four-membered iminosugars **22**–**27**.

C4. As a result of the modeling study, we concluded that the designed six four-membered iminosugars are good structural mimics of NB-DNJ and ceramide.

The synthesis of the four-membered iminosugars (Scheme 7) began by subjecting L-glyceraldehyde acetonide (**28**) to a Wittig reaction, followed by DIBAL-H reduction to furnish allylic alcohol **29**. Sharpless epoxidation and protection of the hydroxyl group provided epoxide **30**. Next, the epoxide was opened with sodium azide, and then the secondary hydroxyl group was benzylated to yield benzyl ether **31**. The azide group was reduced with $LiAlH_4$, and the resulting amino group was reacted subsequently with tosyl chloride to form tosylate **32**. The acetonide protecting group of intermediate **32** was removed and the primary alcohol was next converted to silyl ether **33**. Ring closure to the four-membered ring was

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082–3098

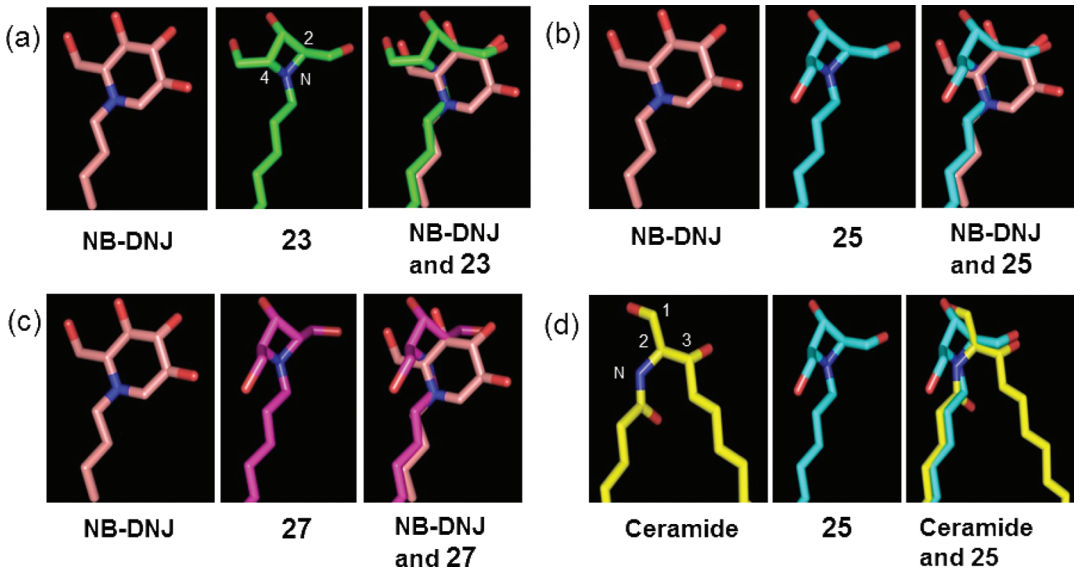
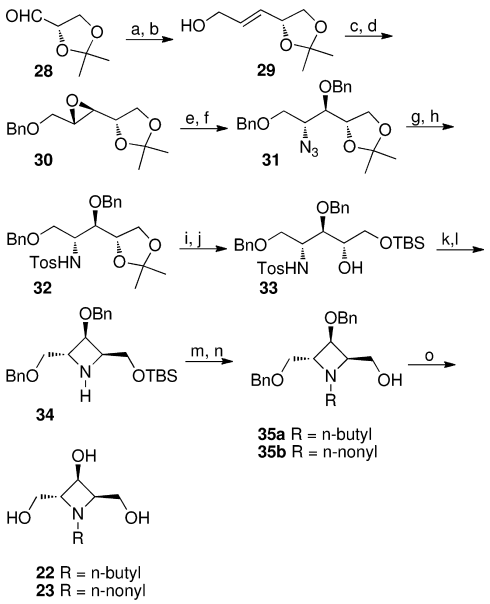


Figure 6. Overlay of ceramide, NB-DNJ, and designed compounds **23**, **25**, and **27**.

Scheme 7[a]

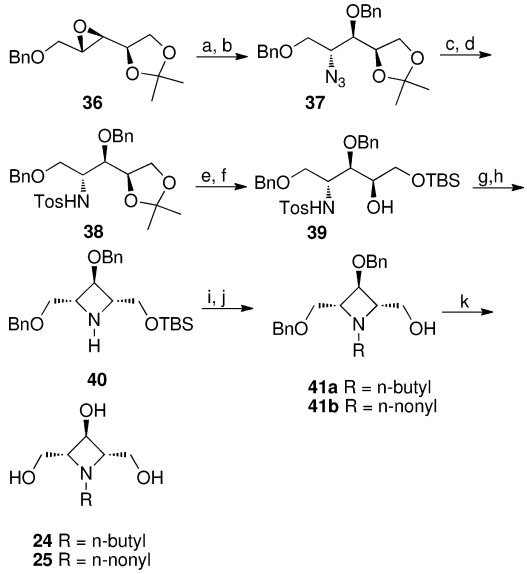


[a]Reagents and conditions: (a) (carbethoxymethylene)-triphenylphosphorane, benzene, reflux, 74%; (b) DIBAL-H, CH_2Cl_2, −78 to 0 °C, 93%; (c) cumene hydroperoxide, (+)-DIPT, $Ti(OiPr)_4$, 3 Å molecular sieves, CH_2Cl_2, −40 °C, 79%; (d) NaH, benzyl bromide, TBAI, THF, rt, 1 h, 96%; (e) NaN_3, NH_4Cl, 2-methoxyethanol−water 9:1, reflux; (f) NaH, benzyl bromide, TBAI, THF, rt, 1 h, 78% over two steps; (g) $LiAlH_4$, THF; (h) tosyl chloride, triethylamine, CH_2Cl_2, rt, 90% over two steps. (i) 2N HCl: methanol, 40 °C; (j) TBSCl, triethylamine, DMAP, CH_2Cl_2, 85% over two steps; (k) triphenylphosphine, DIAD, CH_2Cl_2, rt; (l) Na, naphthalene, DME, −60 °C, 60% over two steps; (m) aldehyde (butyraldehyde or nonyl aldehyde), sodium triacetoxyborohydride, $ClCH_2CH_2Cl$, rt, 70% and 72% respectively over two steps; (n) TBAF, THF, rt; (o) $PdCl_2$, H_2, methanol, 65% and 70%, respectively.

accomplished by a Mitsunobu reaction, which was followed by reductive removal of the *N*-tosyl group to furnish azetidine **34**. Azetidine **34** was subjected to reductive amination with butyraldehyde and nonyl aldehyde followed by desilylation to afford intermediates **35a** and **35b**. Hydrogenolysis of the benzyl protecting groups yielded the targeted four-membered (2*R*,4*R*)-3-hydroxyazetidines **22** and **23**.

Starting with known epoxyacetonide **36** and following the same synthesis procedures as shown in Scheme 7 furnished the (2*R*,3*S*,4*S*)-3-hydroxyazetidines **24** and **25** (Scheme 8).

Scheme 8[a]

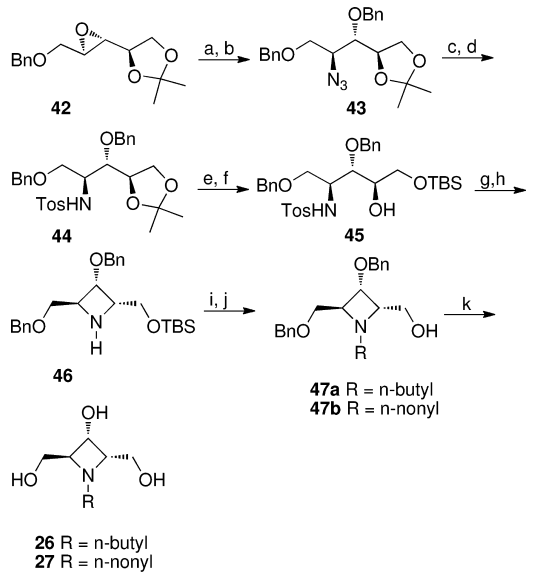


[a]Reagents and conditions: (a) NaN_3, NH_4Cl, 2-methoxyethanol, water 9:1, reflux; (b) NaH, benzyl bromide, TBAI, THF, rt, 1 h 70% over two steps; (c) $LiAlH_4$, THF; (d) tosyl chloride, triethylamine, CH_2Cl_2, rt, 90% over two steps (e) 2N HCl: methanol, 40 °C; (f) TBSCl, triethylamine, DMAP, CH_2Cl_2, 89% over two steps; (g) PPh_3, DIAD, CH_2Cl_2, rt; (h) Na, naphthalene, DME, −60 °C, 53% over two steps; (i) aldehyde (butyraldehyde or nonyl aldehyde), sodium triacetoxyborohydride, $ClCH_2CH_2Cl$, rt; (j) TBAF, THF, rt 73 and 80%, respectively, over two steps; (k) $PdCl_2$, H_2, methanol, 74 and 80%, respectively.

Similarly, starting from epoxyacetonide **42** and following the same procedures shown above in Schemes 7 and 8, the (2*S*,4*S*)-3-hydroxyazetidines **26** and **27** were prepared (Scheme 9).

dx.doi.org/10.1021/jo202054g | *J. Org. Chem.* 2012, 77, 3082−3098

Scheme 9[a]



[a]Reagents and conditions: (a) NaN_3, NH_4Cl, 2-methoxyethanol, water 9:1, reflux; (b) NaH, benzyl bromide, TBAI, THF, rt, 1 h, 70% over two steps; (c) $LiAlH_4$, THF; (d) tosyl chloride, triethylamine, CH_2Cl_2, rt, 88% over two steps (e) 2N HCl: methanol, 40 °C; (f) TBSCl, triethylamine, DMAP, CH_2Cl_2, 87% over two steps; (g) PPh_3, DIAD, CH_2Cl_2, rt; (h) Na, naphthalene, DME, −60 °C, 55% over two steps; (i) aldehyde (butyraldehyde or nonyl aldehyde), sodium triacetoxyborohydride, $ClCH_2CH_2Cl$, rt; (j) TBAF, THF, rt 72% and 76%, respectively, over two steps; (k) $PdCl_2$, H_2, methanol 68% and 74%, respectively.

Enzyme Inhibition Studies. Compounds **1**−**6** and **22**−**27** were tested (Table 1) for inhibition of ceramide-specific glucosyltransferase derived from C57BL/6 mouse and Long Evans (LE) rat testicular microsomes and LE rat-derived testicular β-glucosidase 2. NB-DNJ was used as the positive control. Compounds **1**−**5**, **22**−**24**, **26**, and **27** did not inhibit rat or mouse ceramide-specific glucosyltransferase; however, *N*-nonylazocane derivative **6** showed moderate inhibition (IC_{50} = 127 μM) of the rat-derived ceramide-specific glucosyltransferase (Table 1). *N*-Nonylazetidine derivative **25** was as active as NB-DNJ against mouse-derived ceramide-specific glucosyltransferase (IC_{50} = 44 μM) and moderately active against rat-derived ceramide-specific glucosyltransferase (IC_{50} = 91 μM). The result suggests that the C6 and C7 hydroxyl groups and the longer alkyl chain in the eight-membered analogue **6** are important for activity. As shown with the overlays in Figure 3, the 7-hydroxyl group of compound **6** matches the 5-hydroxyl group of NB-DNJ. This structural similarity with NB-DNJ could be the reason why compound **6** is moderately active. Our results also show that a longer *N*-alkyl group is important for inhibition of this enzyme by eight-membered analogue *N*-nonyl-**6** and four-membered analogue *N*-nonyl-**25** because the corresponding *N*-butyl analogues **5** and **24** are inactive compounds. In the overlay shown in Figure 6d between compound **25** and ceramide, the *N*-alkyl chain and the N−C2−CH_2OH bond of **25** align well with the *N*-acyl chain and the N−C2−C3−OH bond of ceramide, which could be the reason why this is the most active compound in the series. In case of compound **23**, even though the *N*-alkyl chain and the N−C2−CH_2OH bond align with ceramide (picture not shown), the stereochemistry of the 4-hydroxymethyl group seems to prevent inhibition.

When tested for inhibitory activity against β-glucosidase 2, compounds **1**, **5**, and **22**−**26** did not show inhibitory activity, whereas compounds **2**, **3**, **4**, and **6** exhibited weak activities. The IC_{50} values for these compounds are 803, 1123, 904, and 766 μM, respectively. The three *N*-nonyl analogues **2**, **4**, and **6** were more potent then the corresponding *N*-butyl derivatives **1**, **3**, and **5**. *N*-Nonylazetidine **27** inhibited rat testicular β-glucosidase 2 at 70 μM, which is similar to the inhibitory activity of the positive control NB-NDJ. This result suggests that the azetidine stereochemistry and a long alkyl chain, such as the nonyl group, are important for inhibitory activity. Interestingly, the *N*-nonylazocane **6** exhibited activities in both the ceramide-specific glucosyltransferase assay and the β-glucosidase assay.

Iminosugars are already used and also hold promise as modulators of carbohydrate-processing enzymes for various therapeutic applications such as Gaucher's disease, cystic fibrosis, Niemann Pick disease, diabetes, viral disease, Pompe's disease, Fabry's disease, and Parkinson's disease.[8,27] In addition, so-called immucillins are in clinical trials for the treatment of T- and B-cell cancers and autoimmune diseases.[28] Therefore, we further evaluated the inhibitory properties of the new iminosugars toward other readily available glycosidases (Table 2). The following enzymes were investigated: α-glucosidase (*Saccharomyces cerevisiae*), β-glucosidase (almond), α-galactosidase (green coffee beans), β-galactosidase (*Escherichia coli*), α-mannosidase (jack bean), and β-mannosidase (Roman snail). As positive controls for the glycosidase inhibition assays, the following standard compounds were used:[29] DNJ (1-deoxynojirimycin) and NB-DNJ (*N*-butyldeoxynojirimycin) for the α-glucosidase assay; castanospermine for the β-glucosidase assay; DGJ (1-deoxygalactonojirimycin) and NB-DGJ (*N*-butyldeoxygalactonojirimycin) for the α-galactosidase and β-galactosidase assays; and DMJ (1-deoxymannojirimycin) for the α-mannosidase and β-mannosidase assays. The results are summarized in Table 2. In these assays we determined the percent remaining activity of the enzymes in the presence of 100 μM of the iminosugars and also their IC_{50} values (μM).

We found that eight-membered compounds **1**−**4** were modest inhibitors of β-glucosidase (IC_{50} = 87−134 μM). Compounds **5** and **6** showed weak inhibition against β-glucosidase (87 and 85% remaining activity at 100 μM). All of the eight-membered compounds showed weak inhibition against α-glucosidase and β-galactosidase (81−98% remaining activity at 100 μM) and little or no inhibitory activity toward α-galactosidase and mannosidases. Even though the activities were moderate or weak, eight-membered iminosugars exhibited specificity for β-glucosidase. When the activities of compounds **1**−**4** toward β-glucosidase were compared, the length of the *N*-alkyl group did not show much difference in the activity (IC_{50} values, 87 versus 92 μM, and 105 versus 134 μM). The four-membered analogues showed specificity toward α-glucosidase and β-glucosidase. Compound **25** was the most potent compound tested. At 100 μM concentration α-glucosidase activity was inhibited completely and only 26% activity of β-glucosidase remained. The IC_{50} values were 0.6 and 20 μM, respectively. A similar trend was observed for the inhibition of α- and β-galactosidase by **25** but with greatly reduced inhibitory potency. Compound **24** was a moderate inhibitor of α-glucosidase (65% remaining activity) and compound **27** a moderate inhibitor of α-galactosidase (57% remaining activity). Compound **26** had weak β-glucosidase inhibitory activity (82% remaining activity at 100 μM). Compounds **22** and **23**

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082−3098

Table 1. Inhibition of Ceramide-Specific Glucosyltransferase and β-Glucosidase 2 by Iminosugar Analogues 1−6 and 22−27[a]

	ceramide-specific glucosyltransferase		β-glucosidase 2
inhibitor	IC_{50} (C57BL/6, mouse) (μM)	IC_{50} (LE rat) (μM)	IC_{50} (LE rat) (μM)
***NB-DNJ**	51[b]	32	81
1	ni	ni	ni
2	ni	ni	803
3	ni	ni	1123
4	ni	ni	904
5	ni	ni	ni
6	ni	127	766
22	>300	>300	>300
23	>300	>300	>300
24	>300	>300	>300
25	44	91	>300
26	>300	>300	>300
27	>300	>300	70

[a]The details of the enzyme inhibition studies are described in the Experimental Section ni, no inhibition at 1000 μM concentration. *NB-DNJ inhibits HL60 cell-derived ceramide-specific glucosyltransferase with an IC_{50} of 20.4 μM[13d] and a K_i of 7.4 μM.[13a] [b]Mouse-derived testicular ceramide-specific glucosyltransferase was inhibited with an IC_{50} = 23 μM and testicular mouse-derived β-glucosidase 2 with an IC_{50} = 0.14.[10]

displayed no inhibitory activities. Of note is the observation that *N*-nonyl-**25** was active in inhibiting α-glucosidase and β-glucosidase activity, whereas the corresponding *N*-butyl analogue **24** did not show significant activity in these assays.

In conclusion, we have designed and synthesized novel eight- and four-membered iminosugars as potential male contraceptive agents. The *N*-alkyl iminosugar analogues were tested for inhibitory activities against testicular ceramide-specific glucosyltransferase, testicular β-glucosidase 2, and other glycosidases. Among the eight-membered analogues, only the *N*-nonylpentanol derivative **6** was moderately active against rat-derived ceramide-specific glucosyltransferase. *N*-Nonylazetidine **27** was the most potent inhibitor of testicular β-glucosidase 2, on par with the positive control NB-DNJ. Unlike NB-DNJ, azetidine **27** is a selective inhibitor of β-glucosidase 2 since this derivative does not inhibit ceramide-specific glucosyltransferase. Compounds **1**−**4** exhibited modest activity against β-glucosidase from almonds. *N*-Nonylazetidine **25** was found to be a specific inhibitor of mouse- and rat-derived ceramide-specific glucosyltransferase that did not inhibit testicular β-glucosidase 2. Compound **25** was also an effective inhibitor of α-glucosidase and a moderately active inhibitor of almond β-glucosidase. The studies revealed that pentahydroxy substitution and the *N*-nonyl group are important for the activity of the eight-membered analogue **6** for the testis-specific enzymes. In the series of four-membered analogues a stereochemical bias for the *meso*-**25** compound for inhibition of the testis-derived ceramide-specific glucosyltransferases was observed. The *N*-nonyl group was important for the activity for the two most potent compounds, **6** and **25**, because their corresponding *N*-butyl derivatives **5** and **24** were inactive in all assays.

EXPERIMENTAL SECTION

General Procedures. Commercially available chemicals were used as purchased without further purification. All solvents were dried over an activated alumina column before use except commercially available anhydrous 1,2-dimethoxyethane and 1,2-dichloroethane. All reactions with air- or moisture-sensitive reagents were carried out under a nitrogen atmosphere. The ^{1}H NMR spectra were obtained on a 400 MHz spectrometer. For ^{1}H NMR spectra, the chemical shifts are referenced to the tetramethylsilane (TMS) peak as an internal standard or the residual solvent peak. The ^{13}C NMR spectra were recorded at 100 MHz. Chemical shifts are reported in ppm and were referenced to the appropriate residual solvent peak. High-resolution mass spectra (HRMS) were recorded with electron-spray ionization.

Table 2. Glycosidase Inhibitory Activity of Compounds 1−6 and 22−27[a]

	glycosidase % remaining activity at 100 μM/IC_{50} values (μM)											
	α-Glc		β-Glc		α-Gal		β-Gal		α-Man		β-Man	
inhibitor	% Ra	IC_{50}	% Ra	IC_{50}	% Ra	IC_{50}	% Ra	IC_{50}	% Ra	IC_{50}	% Ra	IC_{50}
DNJ	34	79	–	–	–	–	–	–	–	–	–	–
NB-DNJ	–	1030	–	–	–	–	–	–	–	–	–	–
Castano.	–	–	13.5	119	–	–	–	–	–	–	–	–
DGJ	–	–	–	–	0.05	0.018	23	40	–	–	–	–
NB-DGJ	–	–	–	–	–	10	9.6	6.0	–	–	–	–
DMJ	–	–	–	–	–	–	–	–	75	–	77	–
1	93	–	37	87	100	–	98	–	100	–	100	–
2	84	–	43	92	100	–	92	–	100	–	93	–
3	81	–	52	105	100	–	94	–	100	–	99	–
4	90	–	44	134	100	–	90	–	100	–	99	–
5	89	–	87	–	100	–	95	–	100	–	100	–
6	84	–	85	–	100	–	92	–	100	–	99	–
22	100	–	93	–	100	–	99	–	99	–	99	–
23	100	–	95	–	99	–	88	–	100	–	115	–
24	65	–	101	–	106	–	109	–	99	–	113	
25	0	0.6	26	20	66	–	107	–	92	–	76	–
26	90	–	82	–	98	–	96	–	98	–	95	–
27	100	–	95	–	57	–	99	–	99	–	106	–

[a]The details of the enzyme inhibition study are described in the Experimental Section −, not measured; % Ra, % remaining activity at 100 μM; Castano., Castanospermine.

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082−3098

IR spectra were taken on a FT-IR spectrometer. Optical rotations were measured on a polarimeter. Flash column chromatography was performed on silica gel (230−400 mesh).

Methyl 6-O-(tert-Butyldimethylsilyl)-α-D-glucopyranoside (11). To a solution of methyl-α-D-glucopyranoside **10** (20.0 g, 102 mmol) in DMF (160 mL) at 0 °C was added imidazole (17.4 g, 256 mmol) followed by *tert*-butyldimethylsilyl chloride (18.5 g, 123 mmol). The reaction mixture was stirred for 16 h at room temperature. DMF was removed through vacuum distillation, and the residue was taken up in EtOAc (1500 mL). The solution was washed with water (3 × 500 mL) and brine (3 × 500 mL), dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The residue was purified by silica column chromatography (100% EtOAc) to afford **11** (22.6 g, 72%) as a white solid: ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.65 (d, J = 3.8 Hz, 1H), 3.78 (dd, J = 10.6, 4.9 Hz, 1H), 3.72 (dd, J = 10.6, 4.9 Hz, 1H), 3.63 (td, J = 9.1, 2.6 Hz, 1H), 3.50 (dt, J = 9.8, 4.9 Hz, 1H), 3.42 (m, 2H), 3.32 (s, 3H), 3.19 (d, J = 2.2 Hz, 1H), 3.04 (d, J = 2.6 Hz, 1H), 2.34 (d, J = 9.2 Hz, 1H), 0.81 (s, 9H), 0.00 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 99.1, 74.6, 72.3, 72.1, 70.4, 64.2, 55.3, 25.9, 18.3, −5.4; HRMS (ESI) calcd for $[C_{13}H_{28}O_6Si + Na]^+$ 331.1547, found 331.1540. The data are in accordance with reported values.[30]

Methyl 2,3,4-Tri-O-benzyl-6-O-(tert-butyldimethylsilyl)-α-D-glucopyranoside (12). To a solution of compound **11** (8.00 g, 25.9 mmol) in DMF (90 mL) at 0 °C was added sodium hydride (60% dispersion in mineral oil, 3.84 g, 96.0 mmol), and the mixture was stirred at 0 °C for 30 min. Benzyl bromide (20.0 g, 117 mmol) was added at 0 °C, and the mixture was stirred at room temperature for 17 h. The reaction mixture was cooled to 0 °C, and MeOH (10 mL) was added dropwise in order to quench excess sodium hydride. The reaction mixture was poured into water (630 mL) and extracted with Et_2O (5 × 130 mL). The combined organic layers were washed with water (2 × 250 mL) and brine (2 × 250 mL), dried over anhydrous Na_2SO_4, and evaporated under reduced pressure. The residue was purified by silica column chromatography (EtOAc/hexanes 1:12) to afford **12** as a white solid (11 g, 72%): mp 77−78 °C; $[\alpha]^{22}_D$ +24.1 (c 1.00 $CHCl_3$); IR (neat) 3064, 3031, 2928, 2856,, 1949, 1873, 1808, 1748, 1606, 1454, 1360, 1252, 1160, 1092, 1072, 835, 736 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.35−7.20 (m, 15H), 4.93 (d, J = 10.7 Hz, 1H), 4.84 (d, J = 11.2 Hz, 1H), 4.78 (d, J = 10.7 Hz, 1H), 4.75 (d, J = 12.2 Hz, 1H), 4.63 (d, J = 12.2 Hz, 1H), 4.60 (d, J = 11.2 Hz, 1H), 4.57 (d, J = 3.6 Hz, 1H), 3.95 (t, J = 9.2 Hz, 1H), 3.74 (d, J = 3.1 Hz, 2H), 3.58 (dt, J = 9.9, 3.1 Hz, 1H), 3.48 (m, 2H), 3.32 (s, 3H), 0.84 (s, 9H), 0.00 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.8, 138.5, 138.3, 128.4, 128.1, 127.8, 127.7, 127.6, 97.9, 82.2, 80.2, 77.8, 75.9, 75.0, 73.4, 71.5, 62.3, 54.9, 25.9, 18.3, −5.2, −5.4; HRMS (ESI) calcd for $[C_{34}H_{46}O_6Si + Na]^+$ 601.2956, found 601.2975. The data are in agreement with reported values.[31]

Methyl 2,3,4-Tri-O-benzyl-α-D-glucopyranoside (13). To a solution of compound **12** (6.62 g, 11.4 mmol) in THF (33 mL) was added tetrabutylammonium fluoride (1 M solution in THF, 23 mL, 23 mmol), and the mixture was stirred at room temperature for 16 h. The reaction mixture was quenched with water (10 mL) and then extracted with EtOAc (300 mL). The organic layer was washed with brine (2 × 150 mL), dried over anhydrous Na_2SO_4, and evaporated under reduced pressure. The crude product was purified by silica column chromatography (EtOAc/hexanes 1:9 and 1:1) to afford compound **13** as a white solid (5.3 g, 99%): ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.38−7.25 (m, 15H), 4.99 (d, J = 10.9 Hz, 1H), 4.88 (d, J = 11.0 Hz, 1H), 4.83 (d, J = 10.9 Hz, 1H), 4.80 (d, J = 12.1 Hz, 1H), 4.66 (d, J = 12.1 Hz, 1H), 4.64 (d, J = 11.0 Hz, 1H), 4.57 (d, J = 3.6 Hz, 1H), 4.00 (t, J = 9.3 Hz, 1H), 3.76 (dd, J = 11.6, 2.5 Hz, 1H), 3.68 (dd, J = 15.8, 4.1 Hz, 1H), 3.65 (m, 1H), 3.51 (m, 2H), 3.36 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.7, 138.2, 138.1, 128.5, 128.4, 128.1, 128.0, 127.9, 127.6, 98.2, 82.0, 80.0, 77.4, 75.8, 75.0, 73.4, 70.7, 61.9, 55.2; HRMS (ESI) calcd for $[C_{28}H_{32}O_6 + Na]^+$ 487.2091, found 487.2101. The data are in accordance with reported values.[20]

Methyl 6-Deoxy-6-iodo-2,3,4-tri-O-benzyl-α-D-glucopyranoside (14). To a solution of compound **13** (4.6 g, 9.9 mmol), triphenylphosphine (5.2 g, 20 mmol), and imidazole (3.4 g, 50 mmol) in toluene (70 mL) was added iodine (5.0 g, 20 mmol). The reaction mixture was stirred at 70 °C for 3 h and then cooled to room temperature. The toluene layer was decanted from the resulting solid, which was then washed with EtOAc (100 mL). The combined organic solution was concentrated under reduced pressure. Purification by silica column chromatography (EtOAc/hexanes 1:10 and 1:1) furnished **14** as a white solid (5.4 g, 94%): ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.38−7.26 (m, 15H), 4.99 (d, J = 10.8 Hz, 1H), 4.94 (d, J = 10.9 Hz, 1H), 4.80 (d, J = 10.8 Hz, 1H), 4.79 (d, J = 12.1 Hz, 1H), 4.68 (d, J = 10.9 Hz, 1H), 4.65 (d, J = 12.1 Hz, 1H), 4.61 (d, J = 3.6 Hz, 1H), 4.01 (t, J = 9.3 Hz, 1H), 3.53 (dd, J = 9.6, 3.6 Hz, 1H), 3.45 (m, 2H), 3.42 (s, 3H), 3.33 (t, J = 9.1 Hz, 1H), 3.28 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.5, 138.0, 128.5, 128.1, 128.0, 127.9, 127.7, 98.1, 81.6, 81.5, 80.1, 75.8, 75.4, 73.5, 69.3, 55.5, 7.7; HRMS (ESI) calcd for $[C_{28}H_{31}IO_5 + Na]^+$ 597.1108, found 597.1127. The data are in accordance with reported values.[32]

2,3,4-Tri-O-benzyl-5,6-dideoxy-D-xylo-hex-5-enose (9). To a solution of **14** (5.0 g, 8.7 mmol) in THF/H_2O (200 mL/22 mL) was added preactivated Zn (5.7 g, 87 mmol). The reaction mixture was sonicated at 40 °C until full conversion was observed by TLC. After the reaction mixture was cooled to room temperature, Et_2O (340 mL) and H_2O (130 mL) were added. The resulting mixture was filtered, and the organic layer was separated. The organic layer was washed with H_2O (150 mL) and brine (150 mL), dried over anhydrous K_2CO_3, and evaporated under reduced pressure. The resulting yellow syrup was purified by silica column chromatography (EtOAc/hexanes 1:9 and 1:5) to afford **9** as a colorless oil (3.4 g, 93%): ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.65 (s, 1H), 7.36−7.23 (m, 15H), 5.83 (ddd, J = 16.8, 10.8, 7.7 Hz, 1H), 5.28 (d, J = 10.2 Hz, 1H), 5.27 (d, J = 17.6 Hz, 1H), 4.71 (d, J = 11.8 Hz, 2H), 4.58 (d, J = 11.7 Hz, 1H), 4.54 (d, J = 11.5 Hz, 1H), 4.49 (d, J = 11.8 Hz, 1H), 4.36 (d, J = 11.5 Hz, 1H), 4.15 (dd, J = 7.5, 5.1 Hz, 1H), 3.87 (d, J = 4.4 Hz, 1H), 3.80 (t, J = 4.7 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 201.6, 137.8, 137.7, 137.2, 134.8, 128.5, 128.4, 128.3, 128.2, 128.1, 127.9, 127.6, 119.4, 82.4, 81.8, 79.9, 74.5, 73.3, 70.9; HRMS (ESI) calcd for $[C_{27}H_{28}O_4 + K]^+$ 455.1619, found 455.1837. The data are in accordance with reported values.[33]

(2S,3S,4R)-N-Allyl-2,3,4-tris(benzyloxy)hex-5-en-1-amine (15). To a solution of **9** (3.9 g, 9.3 mmol) and allylamine (0.53 g, 9.3 mmol) in 1,2-dichloroethane (40 mL) was added $NaBH(OAc)_3$ (2.7 g, 13 mmol). The reaction mixture was stirred at room temperature for 16 h and quenched by addition of aqueous saturated $NaHCO_3$ (100 mL). The mixture was extracted with EtOAc (300 mL), and the organic layer was washed with aqueous saturated $NaHCO_3$ (100 mL). The organic layer was dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. The crude product was purified by silica column chromatography ($MeOH/CH_2Cl_2$ 1:19) to give **15** as a pale yellow oil (3.6 g, 86%): $[\alpha]^{23}_D$ −11.8 (c 1.00 $CHCl_3$); IR (neat) 3329, 3064, 3030, 2866, 1642, 1497, 1454, 1351, 1208, 1088, 1068, 995, 922, 735, 687 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.37−7.21 (m, 15H), 5.88 (m, 1H), 5.77 (m, 1H), 5.29 (d, J = 15.4 Hz, 1H), 5.26 (d, J = 12.4 Hz, 1H), 5.07 (dd, J = 17.2, 1.7 Hz, 1H), 5.02 (dd, J = 10.2, 1.4 Hz, 1H), 4.74 (s, 2H), 4.64 (d, J = 11.5 Hz, 1H), 4.63 (d, J = 11.8 Hz, 1H), 4.58 (d, J = 11.5 Hz, 1H), 4.36 (d, J = 11.8 Hz, 1H), 4.03 (dd, J = 7.4, 5.0 Hz, 1H), 3.77 (q, J = 5.6 Hz, 1H), 3.61 (t, J = 5.3 Hz, 1H), 3.05 (m, 2H), 2.70 (dd, J = 12.3, 4.7 Hz, 1H), 2.58 (dd, J = 12.3, 6.5 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.8, 138.6, 138.3, 136.9, 135.6, 128.4, 128.3, 128.2, 128.1,127.9, 127.6, 127.5, 118.6, 115.7, 82.0, 80.6, 79.3, 74.9, 73.1, 70.5, 52.4, 49.0; HRMS (ESI) calcd for $[C_{30}H_{35}NO_3 + H]^+$ 458.2690, found 458.2690.

N-Allyl-4-methyl-N-((2S,3S,4R)-2,3,4-tris(benzyloxy)hex-5-enyl)-benzenesulfonamide (8a). A solution of amine **15** (3.5 g, 7.7 mmol), tosyl chloride (1.8 g, 9.3 mmol), DMAP (0.094 g, 0.77 mmol), and triethylamine (1.6 g, 15 mmol) in CH_2Cl_2 (35 mL) was stirred at room temperature for 3 h. The reaction mixture was then washed with H_2O (3 × 35 mL), and the combined water layer was back-extracted with CH_2Cl_2 (2 × 35 mL). The combined organic layer was dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. The residue was purified by silica column chromatography (EtOAc/hexanes 1:9) to give sulfonamide **8a** as a colorless oil (4.0 g, 84%). $[\alpha]^{24}_D$ −25.1 (c 1.00 $CHCl_3$); IR (neat) 3064, 3030, 2868, 1598, 1496,

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082−3098

1454, 1345, 1160, 1090, 930, 736, 698 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.59 (d, J = 8.1 Hz, 2H), 7.35−7.23 (m, 15H), 7.20 (d, J = 8.1 Hz, 2H), 5.84 (ddd, J = 17.3, 10.4, 7.7 Hz, 1H), 5.45 (m, 1H), 5.27 (dd, J = 10.4, 1.6 Hz, 1H), 5.22 (ddd, J = 17.3, 1.6, 0.9 Hz, 1H), 4.99 (dd, J = 3.8, 1.4 Hz, 1H), 4.96 (dd, J = 10.4, 1.4 Hz, 1H), 4.76 (d, J = 11.6 Hz, 1H), 4.69 (d, J = 11.6 Hz, 1H), 4.61 (d, J = 11.6 Hz, 1H), 4.59 (d, J = 11.7 Hz, 1H), 4.55 (d, J = 11.6 Hz, 1H), 4.36 (d, J = 11.7 Hz, 1H), 4.14 (dd, J = 7.7, 5.3 Hz, 1H), 3.99 (dt, J = 7.7, 4.3 Hz, 1H), 3.81(dd, J = 6.4, 1.3 Hz, 2H), 3.56 (t, J = 4.9 Hz, 1H), 3.43 (dd, J = 14.9, 4.2 Hz, 1H), 3.22 (dd, J = 14.9, 7.6 Hz, 1H), 2.40 (s, 3H)); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.1, 138.5, 138.3, 136.8,135.5, 132.9, 129.6, 128.3, 128.2, 128.0, 127.9, 127.6, 127.5, 127.4, 118.8, 80.7, 80.5, 78.5, 74.1, 73.3, 70.6, 52.5, 48.4, 21.5; HRMS (ESI) calcd for $[C_{37}H_{41}NO_5S + Na]^+$ 634.2598, found 634.2600.

*(3S,4S,5R,Z)-3,4,5-Tris(benzyloxy)-1-tosyl-1,2,3,4,5,8-hexahydroazocine (**7a**).* A solution of **8a** (3.9 g, 6.3 mmol) and Grubbs catalyst II (0.53 g, 0.63 mmol, 10 mol %) in CH_2Cl_2 (1600 mL) was refluxed for 1 h. The reaction mixture was concentrated under reduced pressure, and the resulting crude product was purified by silica column chromatography (EtOAc/hexanes 1:5) to afford **7a** as a viscous semisolid (3.1 g, 84%): $[\alpha]^{24}_D$ +86.5 (c 1.00 $CHCl_3$); IR (neat) 3063, 3029, 2865, 1453, 1347, 1162, 1092, 1070, 738, 698 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.58 (d, J = 8.3 Hz, 2H), 7.35−7.22 (m, 17H), 5.67 (ddd, J = 11.8, 6.5, 1.7 Hz, 1H), 5.45 (br d, J = 11.8 Hz, 1H), 5.00 (t, J = 7.6 Hz, 1H), 4.91 (d, J = 11.1 Hz, 1H), 4.70 (d, J = 11.7 Hz, 1H), 4.66 (d, J = 11.1 Hz, 1H), 4.62 (s, 2H), 4.58 (d, J = 11.7 Hz, 1H), 4.22 (br d, J = 16.4 Hz, 1H), 3,96 (ddd, J = 8.5, 6.8, 3.3 Hz, 1H), 3.60 (dd, J = 14.5, 3.3 Hz, 1H), 3.57 (dd, J = 9.1, 6.8 Hz, 1H), 3.24 (dd, J = 16.4, 4.9 Hz, 1H), 2.87 (dd, J = 14.5, 8.5 Hz, 1H), 2.4 (s, 3H)); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.7, 138.9, 138.7, 138.6, 134.2, 134.0, 129.8, 128.4, 128.3, 128.0, 127.8, 127.6, 127.5, 127.4, 125.0, 83.5, 81.4, 77.6, 75.3, 73.2, 72.5, 48.9, 48.0, 21.6; HRMS (ESI) calcd for $[C_{35}H_{37}NO_5S + Na]^+$ 606.2285, found 606.2277.

*tert-Butyl Allyl ((2S,3S,4R)-2,3,4-tris(benzyloxy)hex-5-en-1-yl)carbamate (**8b**).* To a solution of **15** (1.6 g, 3.5 mmol) and Boc anhydride (916 mg, 4.20 mmol) in CH_2Cl_2 (14 mL) was added DMAP (43 mg, 0.35 mmol). The mixture was stirred at room temperature for 16 h and then concentrated under reduced pressure. The residue was purified by column chromatography (EtOAc/hexanes 1:8) to give **8b** (1.56 g, 80%) as a colorless oil: $[\alpha]^{22}_D$ −29.1 (c 1.00 $CHCl_3$); IR (neat) 3065, 3031, 2977, 1694, 1644, 1455, 1405, 1247, 925, 735 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.35−7.22 (m, 15H), 5.81 (m, 1H), 5.68 (br s, 1H), 5.27 (d, J = 10.8 Hz, 1H), 5.22 (d, J = 17.8 Hz, 1H), 5.04 (d, J = 9.6 Hz, 1H), 4.96 (d, J = 17.1 Hz, 1H), 4.82 (d, J = 11.5 Hz, 1H), 4.67 (d, J = 11.5 Hz, 1H), 4.60 (m, 2H), 4.38 (d, J = 11.8 Hz, 1H), 4.14 (t, J = 6.9 Hz, 1H), 3.91 (dd, J = 9.4, 5.2 Hz, 1H), 3.71 (br s, 2H), 3.48 (dd, J = 6.0, 4.3 Hz, 1H), 3.37 (br s, 2H), 1.44 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 155.6, 138.5, 135.5, 134.0, 128.3, 128.2, 128.0, 127.5, 118.9, 115.9, 81.2, 79.6, 74.2, 73.3, 70.7, 50.9, 47.8, 28.5; HRMS (ESI) calcd for $[C_{35}H_{43}NO_5 + H]^+$ 558.3219, found 558.3229.

*(5R,6S,7S)-tert-Butyl 5,6,7-Tris(benzyloxy)-5,6,7,8-tetrahydroazocine-1(2H)-carboxylate (**7b**).* A solution of **8b** (100 mg, 0.179 mmol) and Grubbs catalyst II (15 mg, 0.018 mmol, 10 mol %) in CH_2Cl_2 (45 mL) was refluxed for 4 h. The reaction mixture was concentrated under reduced pressure, and the resulting crude product was purified by silica column chromatography (EtOAc/hexaness 1:8) affording 88 mg (92%) of a rotameric mixture of **7b** (1:1.4) as a colorless oil. *Rotamer A*: ^1H NMR (400 MHz, $CDCl_3$) δ 7.36−7.23 (m, 15H), 5.66 (m, 2H), 4.84 (d, J = 12.0 Hz, 1H), 4.71−4.50 (m, 5H), 4.47 (m, 1H), 4.17 (d, J = 16.1 Hz, 1H), 3.90 (dd, J = 14.6, 3.0 Hz, 1H), 3.80 (m, 1H), 3.66 (dd, J = 16.6, 4.0 Hz, 1H), 3.60 (dd, J = 9.0, 6.5 Hz, 1H), 3.25 (dd, J = 14.3, 8.3 Hz, 1H), 1.42 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 155.3, 138.7, 138.5, 133.0, 132.8, 128.4, 128.3, 128.2, 127.7, 127.6, 127.5, 127.4, 127.3, 83.8, 79.9, 79.8, 77.5, 75.1, 72.7, 71.9, 46.6, 45.8, 28.4. *Rotamer B*: ^1H NMR (400 MHz, $CDCl_3$) δ 7.36−7.23 (m, 15H), 5.66 (m, 2H), 4.89 (d, J = 11.0 Hz, 1H), 4.71−4.50 (m, 5H), 4.47 (m, 2H), 3.90 (dd, J = 14.6, 3.0 Hz, 1H), 3.80 (m, 1H), 3.60 (dd, J = 9.0, 6.5 Hz, 1H), 3.57 (dd, J = 18.7, 2.6 Hz, 1H), 3.16 (dd, J = 14.7, 8.4 Hz, 1H), 1.46 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 155.5, 138.9, 138.6, 133.0, 132.8, 128.4, 128.3, 128.2, 127.7, 127.6, 127.5, 127.4, 127.3, 83.6, 80.7, 80.5, 77.5, 75.4, 72.9, 71.9, 46.6, 46.5, 28.5; HRMS (ESI) calcd for $[C_{33}H_{39}NO_5 + Na]^+$ 552.2720, found 552.2711.

*(3S,4S,5R,Z)-3,4,5-Tris(benzyloxy)-1,2,3,4,5,8-hexahydroazocine (**16**).* A solution of Na metal (405 mg, 17.6 mmol) and naphthalene (2.48 g, 19.4 mmol) in 1,2-dimethoxyethane (18 mL) was stirred at room temperature for 2 h. To a solution of **7a** (664 mg, 1.14 mmol) in 1,2-dimethoxyethane (13 mL) at −78 °C was added the Na-naphthalene solution (11.4 mL) dropwise for 30 min. The reaction mixture was stirred at −78 °C for 5 min and then H_2O (2.1 mL) was slowly added to the mixture at −78 °C to quench the reaction. The reaction mixture was diluted with Et_2O (120 mL), dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. The residue was purified by silica column chromatography (MeOH:CH_2Cl_2 1: 9) to furnish **16** as a yellowish oil (366 mg, 75%): $[\alpha]^{24}_D$ −13.6 (c 1.00 $CHCl_3$); IR (neat) 3364, 3063, 3029, 2863, 1496, 1454, 1355, 1207, 1090, 1069, 735, 697 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.37−7.22 (m, 15H), 5.76 (m, 1H), 5.69 (dd, J = 11.7, 6.7 Hz, 1H), 4.77 (m, 1H), 4.74 (d, J = 11.5 Hz, 1H), 4.65 (d, J = 11.5 Hz, 1H), 4.63 (d, J = 11.8 Hz, 1H), 4.59 (d, J = 11.8 Hz, 1H), 4.53 (d, J = 11.8 Hz, 1H), 4.49 (d, J = 11.8 Hz, 1H), 3.71 (dd, J = 7.4, 5.4 Hz, 1H), 3.57 (m, 1H), 3.44 (dd, J = 16.8, 4.8 Hz, 1H), 3.29 (ddd, J = 16.8, 5.2, 1.5 Hz, 1H), 3.06 (m, 2H)); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.7,138.6, 138.5, 132.9, 130.0, 128.4, 128.3, 128.1, 127.7, 127.6, 127.5, 84.0, 79.9, 78.2, 74.2, 72.3, 71.7, 48.1, 47.8; HRMS (ESI) calcd for $[C_{28}H_{31}NO_3 + Na]^+$ 452.2196, found 452.2209. Compound **16** was also obtained from **7b**. Compound **7b** (20 mg, 0.038 mmol) was dissolved with 4 N HCl in dioxane (118 μL, 0.47 mmol). The solution was stirred at room temperature for 1 h. Excess HCl and dioxane were removed by evaporation. Concentrated NH_4OH (60 μL) and H_2O (300 μL) were added to the concentrated mixture. The mixture was extracted with CH_2Cl_2 (3 × 2 mL), dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The crude product was purified by silica column chromatography (MeOH/CH_2Cl_2 1:9) to give **16** as a yellowish oil (14 mg, 85%). All the spectra are identical with the ones of compound **16** obtained from compound **7a**.

*(3S,4S,5R,Z)-3,4,5-Tris(benzyloxy)-1-butyl-1,2,3,4,5,8-hexahydroazocine (**17a**).* To a solution of **16** (200 mg, 0.47 mmol) and butyraldehyde (40 μL, 0.45 mmol) in 1,2-dichloroethane (2 mL) at room temperature was added $NaBH(OAc)_3$ (140 mg, 0.65 mmol). The reaction mixture was stirred at room temperature for 16 h, diluted with EtOAc (16 mL), washed with saturated $NaHCO_3$ (2 × 4 mL), dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The crude product was purified by silica column chromatography (EtOAc/hexanes 1:3) to give **17a** as a yellowish oil (170 mg, 75%): $[\alpha]^{24}_D$ +1.8 (c 1.0 $CHCl_3$); IR (neat) 3063, 3029, 2930, 2861, 1496, 1454, 1358, 1207, 1091, 1068, 734, 697 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.36−7.22 (m, 15H), 5.57 (ddd, J = 11.9, 5.7, 1.6 Hz, 1H), 5.50 (br d, J = 11.7 Hz, 1H), 5.18 (br s, 1H), 4.94 (d, J = 10.7 Hz, 1H), 4.69 (d, J = 11.3 Hz, 1H), 4.68 (d, J = 10.7 Hz, 1H), 4.66 (d, J = 11.3 Hz, 1H), 4.59 (d, J = 11.6 Hz, 1H), 4.58 (d, J = 11.6 Hz, 1H), 3.54 (m, 2H), 3.25 (br d, J = 16.4 Hz, 1H), 2.97 (dd, J = 16.4, 3.6 Hz, 1H), 2.76 (dd, J = 13.8, 8.9 Hz, 1H), 2.57 (d, J = 13.8 Hz, 1H), 2.37 (m, 2H), 1.35 (m, 2H), 1.24 (sex, J = 7.2 Hz, 2H), 0.86 (t, J = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 139.3, 139.2, 138.9, 133.1, 128.8, 128.3, 128.2, 127.9, 127.7, 127.5, 127.4, 127.2, 85.1, 81.4, 78.2, 75.7, 72.9, 71.9, 57.7, 55.1, 55.0, 30.0, 20.5, 14.0; HRMS (ESI) calcd for $[C_{32}H_{39}NO_3 + H]^+$ 486.3003, found 486.3020.

*(3S,4S,5R,Z)-3,4,5-Tris(benzyloxy)-1-nonyl-1,2,3,4,5,8-hexahydroazocine (**17b**).* To a solution of **16** (220 mg, 0.51 mmol) and nonyl aldehyde (85 μL, 0.49 mmol) in 1,2-dichloroethane (2 mL) at room temperature was added $NaBH(OAc)_3$ (152 mg, 0.717 mmol). The reaction mixture was stirred at room temperature for 16 h, diluted with EtOAc (18 mL), washed with saturated $NaHCO_3$ (2 × 5 mL), dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The crude product was purified by silica column chromatography (EtOAc/hexanes 1:4) to give **17b** as a yellowish oil (217 mg, 76%). $[\alpha]^{24}_D$ −3.5 (c 1.0 $CHCl_3$); IR (neat) 3063, 3030, 2926, 2855, 1496, 1454, 1357, 1207, 1092, 1068, 733, 697 cm^{-1};

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082−3098

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.38−7.22 (m, 15H), 5.57 (dd, J = 11.5, 5.5 Hz, 1H), 5.50 (br d, J = 11.8 Hz, 1H), 5.18 (br s, 1H), 4.94 (d, J = 10.7 Hz, 1H), 4.69 (d, J = 11.4 Hz, 1H), 4.68 (d, J = 10.7 Hz, 1H), 4.65 (d, J = 11.4 Hz, 1H), 4.59 (d, J = 11.6 Hz, 1H), 4.58 (d, J = 11.6 Hz, 1H), 3.53 (m, 2H), 3.25 (br d, J = 16.4 Hz, 1H), 2.97 (dd, J = 16.5, 3.8 Hz, 1H), 2.76 (dd, J = 13.9, 8.9 Hz, 1H), 2.58 (d, J = 13.9 Hz, 1H), 2.37 (m, 2H), 1.42−1.15 (br m, 14H), 0.87 (t, J = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 139.2, 138.8, 133.1, 128.7, 128.3, 128.2, 127.8, 127.7, 127.5, 127.4, 127.2, 85.1, 81.4, 78.3, 75.6, 72.9, 72.0, 58.0, 55.1, 54.9, 31.9, 29.6, 29.3, 27.8, 27.4, 22.7, 14.1; HRMS (ESI) calcd for $[C_{37}H_{49}NO_3 + H]^+$ 556.3785, found 556.3804.

*(3S,4S,5R)-1-Butylazocane-3,4,5-triol (**1**).* To a solution of **17a** (80 mg, 0.17 mmol) in MeOH (5 mL) was added $PdCl_2$ (20 mg, 0.12 mmol). The reaction mixture was stirred under H_2 atmosphere at room temperature for 16 h. The reaction mixture was filtered through Celite and concentrated under reduced pressure. The residue was purified by SPE-amine column chromatography ($MeOH/CH_2Cl_2$ 1:100 to 1:50 gradient) to furnish **1** as a colorless thick oil (27 mg, 76%): $[\alpha]^{24}_D$ +40.9 (c 1.04 MeOH); IR (neat) 3363, 2929, 2863, 1653, 1456, 1364, 1102, 1035, 943 cm^{-1}; ^{1}H NMR (400 MHz, CD_3OD) δ 3.89 (dd, J = 8.4, 3.3 Hz, 1H), 3.72 (td, J = 9.6, 5.8 Hz, 1H), 3.44 (dd, J = 9.1, 3.3 Hz, 1H), 2.66−2.45 (m, 6H), 1.92−1.74 (m, 3H), 1.59 (m, 1H), 1.50 (quin, J = 7.4 Hz, 2H), 1.36 (m, 2H), 0.95 (t, J = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 80.6, 71.9, 71.8, 60.1, 60.0, 53.7, 30.8, 27.8, 25.9, 21.6, 14.3; HRMS (ESI) calcd for $[C_{11}H_{23}NO_3 + H]^+$ 218.1751, found 218.1753.

*(3S,4S,5R)-1-Nonylazocane-3,4,5-triol (**2**).* To a solution of **17b** (70 mg, 0.13 mmol) in MeOH (4 mL) was added $PdCl_2$ (16 mg, 0.088 mmol). The reaction mixture was stirred under H_2 atmosphere at room temperature for 16 h. The reaction mixture was filtered through Celite and concentrated under reduced pressure. The residue was purified by SPE-amine column chromatography ($MeOH/CH_2Cl_2$ 1:100 to 1:50 gradient) to furnish **2** as a colorless thick oil (28 mg, 78%). $[\alpha]^{24}_D$ +33.4 (c 1.03 MeOH); IR (neat) 3392, 2923, 2854, 1647, 1468, 1364, 1105, 1039, 950 cm^{-1} ; ^{1}H NMR (400 MHz, CD_3OD) δ 3.79 (dd, J = 8.2, 3.3 Hz, 1H), 3.62 (td, J = 9.6, 5.7 Hz, 1H), 3.34 (dd, J = 9.0, 3.3 Hz, 1H), 2.56−2.35 (m, 6H), 1.82−1.64 (m, 3H), 1.49 (m, 1H), 1.42 (m, 2H), 1.23 (br s, 12H), 0.81 (t, J = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 80.6, 72.0, 71.8, 60.5, 60.0, 53.8, 33.0, 30.6, 30.4, 28.6, 28.4, 27.9, 25.9, 23.7, 14.4; HRMS (ESI) calcd for $[C_{16}H_{33}NO_3 + H]^+$ 288.2533, found 288.2535.

*(3S,4S,5R,Z)-1-Butyl-1,2,3,4,5,8-hexahydroazocine-3,4,5-triol (**3**).* To liquid ammonia (24 mL) at −78 °C was added granular Li metal (91 mg, 13 mmol). The solution was stirred at −78 °C for 20 min. A solution of **17a** (80 mg, 0.16 mmol) in THF (4 mL) was added slowly to the Li−ammonia solution at −78 °C, and then the reaction mixture was stirred at −78 °C for 1 h. Liquid ammonia was removed by nitrogen purge at −78 °C. When most ammonia was removed, MeOH (30 mL) containing 4 drops of H_2O was added to the residue at −78 °C to quench the reaction. After being stirred at −78 °C for 20 min, the mixture was evaporated under reduced pressure. The residue was dissolved in $MeOH/CH_2Cl_2$ (2:8, 5 mL) and filtered through Celite. The filtered solution was concentrated in vacuo and purified by SPE-amine column chromatography ($MeOH/CH_2Cl_2$ 0.5:100 to 4:100 gradient) to furnish **3** as a colorless thick oil (21 mg, 59%): $[\alpha]^{25}_D$ +73.2 (c 1.04 MeOH); IR (neat) 3370, 3020, 2957, 2932, 2871, 1655, 1458, 1377, 1039, 968, 725 cm^{-1}; ^{1}H NMR (400 MHz, CD_3OD) δ 5.86 (m, 1H), 5.71 (dt, J = 12.2, 3.5 Hz, 1H), 4.18 (br s, 1H), 3.71 (m, 1H), 3.46 (dd, J = 8.5, 3.7 Hz, 1H), 3.38 (br d, J = 17.5 Hz, 1H), 3.04 (br d, J = 18.5 Hz, 1H), 2.81 (t, J = 11.6 Hz, 1H), 2.54 (m, 3H), 1.52 (quin, J = 7.5 Hz, 2H), 1.36 (sex, J = 7.4 Hz, 2H), 0.95 (t, J = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 132.9, 130.3, 80.7, 72.3, 70.9, 60.9, 59.3, 56.5, 30.3, 21.6, 14.2; HRMS (ESI) calcd for $[C_{11}H_{21}NO_3 + Na]^+$ 238.1414, found 238.1423.

*(3S,4S,5R,Z)-1-Nonyl-1,2,3,4,5,8-hexahydroazocine-3,4,5-triol (**4**).* To liquid ammonia (21 mL) at −78 °C was added granular Li metal (80 mg, 12 mmol). The solution was stirred at −78 °C for 20 min. A solution of **17b** (80 mg, 0.14 mmol) in THF (4 mL) was added slowly to the Li−ammonia solution at −78 °C, and then the reaction mixture was stirred at −78 °C for 1 h. Liquid ammonia was removed by nitrogen purge at −78 °C. When most ammonia was removed, MeOH (30 mL) containing 5 drops of H_2O was added to the residue at −78 °C to quench the reaction. After being stirred at −78 °C for 20 min, the mixture was evaporated under reduced pressure. The residue was dissolved in $MeOH/CH_2Cl_2$ (1:9, 5 mL) and filtered through Celite. The filtered solution was concentrated in vacuo and purified by SPE-amine column chromatography ($MeOH/CH_2Cl_2$ 0.5:100 to 4:100 gradient) to furnish **4** as a semisolid (27 mg, 66%): $[\alpha]^{25}_D$ +62.2 (c 1.05 MeOH); IR (neat) 3408, 3226, 2921, 2853, 1660, 1486, 1459, 1360, 1294, 1074, 1039, 974, 715 cm^{-1}; ^{1}H NMR (400 MHz, CD_3OD) δ 5.77 (m, 1H), 5.61 (dt, J = 12.3, 3.5 Hz, 1H), 4.08 (br s, 1H), 3.61 (m, 1H), 3.36 (dd, J = 8.4, 3.7 Hz, 1H), 3.28 (br d, J = 18.1 Hz, 1H), 2.94 (br d, J = 18.5 Hz, 1H), 2.71 (t, J = 11.6 Hz, 1H), 2.44 (m, 3H), 1.44 (br t, J = 6.9 Hz, 2H), 1.23 (br s,12H), 0.81 (t, J = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 133.0, 130.3, 80.7, 72.3, 70.9, 60.9, 59.7, 56.6, 33.0, 30.6, 30.4, 28.4, 28.1, 23.7, 14.4; HRMS (ESI) calcd for $[C_{16}H_{31}NO_3 + Na]^+$ 308.2196, found 308.2194.

*(3R,4R,5R,6R,7S)-5,6,7-Tris(benzyloxy)-1-tosylazocane-3,4-diol (**18**).* To a solution of **7a** (300 mg, 0.514 mmol) in THF/*t*-BuOH/H_2O (3 mL/9 mL/9 mL) were added K_2CO_3 (213 mg, 1.54 mmol), $K_3(FeCN)_6$ (508 mg, 1.54 mmol), and $(DHQ)_2$-PHAL (40 mg, 0.051 mmol). The mixture was stirred at 0 °C for 5 min, and then to the solution were added $CH_3SO_2NH_2$ (98 mg, 1.0 mmol) and $K_2OsO_2(OH)_4$ (4 mg, 0.01 mmol). After the reaction mixture had been stirred at room temperature for 40 h, Na_2SO_3 (780 mg, 6.2 mmol) was added for quenching and the mixture was stirred for 40 min. The mixture was diluted with H_2O (25 mL) and extracted with EtOAc (4 × 75 mL). The organic layer was washed with 2 N KOH (120 mL), dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The residue was purified by silica column chromatography (EtOAc/hexanes 1:1) to give **18** as a white foam (260 mg, 82%): $[\alpha]^{24}_D$ +41.4 (c 1.00 MeOH); IR (neat) 3392, 3063, 3030, 2924, 1598, 1496, 1454, 1344, 1160, 1089, 1072, 908, 816, 726, 699 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.66 (d, J = 8.3 Hz, 2H), 7.37−7.23 (m, 15H), 7.16 (m, 2H), 4.78 (d, J = 11.8 Hz, 1H), 4.68 (d, J = 11.1 Hz, 1H), 4.62 (d, J = 11.1 Hz, 1H), 4.56 (d, J = 11.8 Hz, 1H), 4.44 (d, J = 11.1 Hz, 1H), 4.38 (d, J = 11.1 Hz, 1H), 4.38 (d, J = 8.0 Hz, 1H), 4.16−4.01 (m, 3H), 3.99−3.91 (m, 2H), 3.68 (q, J = 4.0 Hz, 1H), 3.39 (dd, J = 14.8, 5.0 Hz, 1H), 2.96 (dd, J = 14.8, 10.7 Hz, 1H), 2.82 (dd, J = 13.8, 3.0 Hz, 1H), 2.67 (d, J = 9.0 Hz, 1H), 2.43 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.8, 138.3, 138.0, 135.7, 134.1, 129.8, 128.8, 128.6, 128.4, 128.3, 128.0, 127.8, 127.7, 127.5, 83.8, 80.7, 79.4, 74.9, 74.6, 73.8, 73.6, 71.8, 48.2, 47.4, 21.6; HRMS (ESI) calcd for $[C_{35}H_{39}NO_7S + Na]^+$ 640.2345, found 640.2348.

*(3S,4R,5S,6R,7R)-3,4,5,6,7-Pentakis(benzyloxy)-1-tosylazocane (**19**).* To a solution of **18** (700 mg, 1.1 mmol) in dry DMF (4.2 mL) at 0 °C were added sodium hydride (60% dispersion in mineral oil, 159 mg, 3.97 mmol) and benzyl bromide (582 mg, 3.40 mmol). The reaction mixture was stirred at room temperature for 15 h and then quenched with MeOH (20 drops). The reaction mixture was diluted with H_2O (42 mL) and extracted with EtOAc (4 × 80 mL). The organic layer was washed with H_2O (160 mL) and brine (2 × 160 mL), dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The crude product was purified by silica column chromatography (EtOAc/hexanes 1:5) to afford **19** as a colorless thick oil (787 mg, 87%): $[\alpha]^{24}_D$ +30.8 (c 1.00 $CHCl_3$); IR (neat) 3063, 3030, 2930, 2869, 1598, 1496, 1454, 1346, 1161, 1089, 912, 816, 737, 698 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.52 (d, J = 8.3 Hz, 2H), 7.37−7.19 (m, 27H), 4.91 (d, J = 10.6 Hz, 1H), 4.82 (d, J = 11.5 Hz, 1H), 4.81 (d, J = 11.8 Hz, 1H), 4.79 (d, J = 11.9 Hz, 1H), 4.75 (d, J = 10.6 Hz, 1H), 4.74 (d, J = 11.5 Hz, 1H), 4.71 (d, J = 11.8 Hz, 1H), 4.47 (d, J = 11.9 Hz, 1H), 4.33 (d, J = 11.8 Hz, 1H), 4.28 (d, J = 11.8 Hz, 1H), 4.22 (s, 1H), 4.01 (m, 2H), 3.82 (d, J = 8.1 Hz, 1H), 3.60 (dd, J = 13.8, 5.0 Hz, 1H), 3.49−3.41 (m, 2H), 3.11 (dd, J = 15.1, 10.1 Hz, 1H), 2.82 (dd, J = 13.8, 10.7 Hz, 1H), 2.40 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.7, 139.2, 139.0, 138.9, 138.2, 134.8, 129.8, 128.4, 128.3, 128.2, 128.0, 127.9, 127.7, 127.6, 127.5, 127.3, 127.2, 83.9, 83.8, 83.2, 81.8, 81.6, 76.2, 74.7, 74.2, 73.8, 71.9, 51.8, 50.2, 21.5; HRMS (ESI) calcd for $[C_{49}H_{51}NO_7S + Na]^+$ 820.3278, found 820.3307.

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082−3098

*(3S,4R,5S,6R,7R)-3,4,5,6,7-Pentakis(benzyloxy)azocane (**20**).* A solution of Na metal (583 mg, 25.4 mmol) and naphthalene (3.58 g, 27.9 mmol) in 1,2-dimethoxyethane (25 mL) was stirred at room temperature for 2 h. To a solution of **19** (763 mg, 0.956 mmol) in 1,2-dimethoxyethane (11 mL) at −78 °C was added the Na−naphthalene solution (3.8 mL) dropwise for 20 min. After the reaction mixture was stirred at −78 °C for 10 min, H_2O (1.5 mL) with Et_2O (10 mL) and 1,2-dimethoxyethane (2 mL) were slowly added to the mixture at −78 °C to quench the reaction. The slurry was stirred at −78 °C until the green color disappeared. The reaction mixture was diluted with Et_2O (120 mL), dried over anhydrous $MgSO_4$, and concentrated under reduced pressure. The residue was purified by silica column chromatography ($MeOH/CH_2Cl_2$ 1: 13) to furnish **20** as a yellowish oil (440 mg, 72%): $[\alpha]^{24}_D$ −3.0 (*c* 1.0 $CHCl_3$); IR (neat) 3384, 3063, 3030, 2868, 1496, 1454, 1362, 1208, 1092, 1072, 736 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.42−7.20 (m, 25H), 4.85−4.67 (m, 6H), 4.60 (d, *J* = 11.6 Hz, 1H), 4.58 (d, *J* = 11.6 Hz, 1H), 4.39 (d, *J* = 12.0 Hz, 1H), 4.34 (d, *J* = 12.0 Hz, 1H), 4.09 (br s, 1H), 3.96 (t, *J* = 8.1 Hz, 1H), 3.84 (d, *J* = 7.8 Hz, 1H), 3.44−3.36 (m, 2H), 3.09−2.99 (m, 2H), 2.92 (dd, *J* = 14.6, 10.1 Hz, 1H), 2.85 (dd, *J* = 13.9, 4.4 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 139.1, 139.0, 138.9, 138.7, 138.6, 128.3, 128.2, 128.0, 127.8, 127.6, 127.5, 127.4, 83.5, 83.3, 82.7, 82.2, 81.6, 75.5, 74.3, 73.5, 73.4, 71.5, 50.0, 48.3; HRMS (ESI) calcd for $[C_{42}H_{45}NO_5 + H]^+$ 644.3370, found 644.3386.

*(3S,4R,5S,6R,7R)-3,4,5,6,7-Pentakis(benzyloxy)-1-butylazocane (**21a**).* To a solution of **20** (204 mg, 0.317 mmol) and butyraldehyde (27 μL, 0.30 mmol) in 1,2-dichloroethane (1.5 mL) at room temperature was added $NaBH(OAc)_3$ (94 mg, 0.44 mmol). The reaction mixture was stirred at room temperature for 16 h, diluted with EtOAc (30 mL), and washed with saturated $NaHCO_3$ (2 × 10 mL). The organic layer was dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. The crude product was purified by silica column chromatography (first purification EtOAc/hexanes 1:1, second purification $MeOH/CH_2Cl_2$ 1:18) to give **21a** as a yellowish oil (175 mg, 79%): $[\alpha]^{26}_D$ +20.2 (*c* 1.00 $CHCl_3$); IR (neat) 3063, 3030, 2930, 2862, 1679, 1496, 1454, 1360, 1206, 1072, 735, 698 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.43−7.20 (m, 25H), 4.94 (d, *J* = 10.3 Hz, 1H), 4.88 (d, *J* = 12.1 Hz, 1H), 4.81 (d, *J* = 12.1 Hz, 1H), 4.79 (d, *J* = 12.1 Hz, 1H), 4.74 (d, *J* = 10.3 Hz, 1H), 4.70 (d, *J* = 11.6 Hz, 1H), 4.57 (d, *J* = 11.6 Hz, 1H), 4.49 (d, *J* = 12.1 Hz, 1H), 4.30 (d, *J* = 12.1 Hz, 1H), 4.22 (s, 1H), 4.19 (d, *J* = 12.1 Hz, 1H), 4.09 (t, *J* = 8.8 Hz, 1H), 3.88 (d, *J* = 9.4 Hz, 1H), 3.33 (ddd, *J* = 10.6, 8.0, 2.8 Hz, 1H), 3.12−3.01 (m, 2H), 2.79 (dd, *J* = 13.2, 10.8 Hz, 1H), 2.64 (dd, *J* = 14.7, 3.2 Hz, 1H), 2.59 (dd, *J* = 13.6, 5.7 Hz, 1H), 2.42 (m, 1H), 2.25 (m, 1H), 1.17 (br m, 4H), 0.83 (t, *J* = 7.0 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 139.4, 139.1, 138.9, 138.6, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.5, 127.4, 127.3, 84.0, 83.4, 82.3, 81.9, 76.4, 74.2, 74.0, 73.1, 71.1, 57.8, 57.7, 55.1, 30.5, 20.4, 14.1; HRMS (ESI) calcd for $[C_{46}H_{53}NO_5 + H]^+$ 700.3996, found 700.4004.

*(3S,4R,5S,6R,7R)-3,4,5,6,7-Pentakis(benzyloxy)-1-nonylazocane (**21b**).* To a solution of **20** (198 mg, 0.308 mmol) and nonyl aldehyde (51 μL, 0.30 mmol) in 1,2-dichloroethane (1.5 mL) at room temperature was added $NaBH(OAc)_3$ (91 mg, 0.43 mmol). The reaction mixture was stirred at room temperature for 16 h, diluted with EtOAc (30 mL), and washed with saturated $NaHCO_3$ (2 × 10 mL). The organic layer was dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. The crude product was purified by silica column chromatography (first purification EtOAc/hexanes 1:1, second purification $MeOH/CH_2Cl_2$ 1:16) to give **21b** as a yellowish oil (200 mg, 84%): $[\alpha]^{25}_D$ +20.6 (*c* 1.00 $CHCl_3$); IR (neat) 3063, 3030, 2925, 2854, 1681, 1496, 1454, 1359, 1206, 1067, 912, 733, 696 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.44−7.13 (m, 25H), 4.93 (d, *J* = 10.3 Hz, 1H), 4.88 (d, *J* = 12.1 Hz, 1H), 4.81 (d, *J* = 12.1 Hz, 1H), 4.79 (d, *J* = 12.0 Hz, 1H), 4.74 (d, *J* = 10.3 Hz, 1H), 4.70 (d, *J* = 11.6 Hz, 1H), 4.58 (d, *J* = 11.6 Hz, 1H), 4.48 (d, *J* = 12.0 Hz, 1H), 4.29 (d, *J* = 12.1 Hz, 1H), 4.22 (s, 1H), 4.19 (d, *J* = 12.1 Hz, 1H), 4.08 (t, *J* = 8.8 Hz, 1H), 3.88 (d, *J* = 9.4 Hz, 1H), 3.33 (ddd, *J* = 10.5, 8.1, 2.6 Hz,1H), 3.14−3.00 (m, 2H), 2.79 (dd, *J* = 13.2, 10.8 Hz, 1H), 2.64 (dd, *J* = 14.7, 2.7 Hz, 1H), 2.60 (dd, *J* = 13.2, 5.3 Hz, 1H), 2.41 (m, 1H), 2.25 (m, 1H), 1.36−1.07 (br m, 14H), 0.89 (t, *J* = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 139.4, 139.3, 139.0, 138.8, 138.6, 128.4, 128.3, 128.2, 128.0, 127.9, 127.5, 127.4, 127.3, 83.9, 83.4, 82.3, 82.1, 74.3, 73.9, 73.1, 71.1, 58.1, 57.6, 55.1, 31.9, 29.7, 29.6, 29.4, 28.3, 27.3, 22.7, 14.2; HRMS (ESI) calcd for $[C_{51}H_{63}NO_5 + H]^+$ 770.4779, found 770.4770.

*(3S,4R,5S,6R,7R)-1-Butylazocane-3,4,5,6,7-pentaol (**5**).* To a solution of **21a** (164 mg, 0.234 mmol) in MeOH (6 mL) was added $PdCl_2$ (33 mg, 0.19 mmol). The reaction mixture was stirred under H_2 atmosphere at room temperature for 18 h. The reaction mixture was filtered through Celite and concentrated under reduced pressure. The residue was purified by SPE-amine column chromatography ($MeOH/CH_2Cl_2$ 1:9 and 1:5) to afford **5** as a semisolid (47 mg, 80%): $[\alpha]^{25}_D$ +19.9 (*c* 0.932 MeOH); IR (neat) 3403, 3356, 2947, 2872, 2807, 1683, 1469, 1398, 1105, 1040 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 3.89 (m, 3H), 3.63 (td, *J* = 8.3, 6.1 Hz, 1H), 3.57 (dd, *J* = 8.5, 4.2 Hz, 1H), 2.84 (dd, *J* = 14.8, 5.2 Hz, 1H), 2.73 (dd, *J* = 14.8, 2.7 Hz, 1H), 2.67 (d, *J* = 6.1 Hz, 1H), 2.67 (d, *J* = 8.3 Hz, 1H), 2.59 (t, *J* = 7.7 Hz, 2H), 1.54 (m, 2H), 1.34 (sex, *J* = 7.4 Hz, 2H), 0.95 (t, *J* = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 79.9, 78.6, 74.3, 73.4, 72.6, 60.9, 59.3, 57.1, 30.4, 21.6, 14.3; HRMS (ESI) calcd for $[C_{11}H_{23}NO_5 + H]^+$ 250.1649, found 250.1644.

*(3S,4R,5S,6R,7R)-1-Nonylazocane-3,4,5,6,7-pentaol (**6**).* To a solution of **21b** (19 mg, 0.025 mmol) in MeOH (0.8 mL) was added $PdCl_2$ (4 mg, 0.02 mmol). The reaction mixture was stirred under H_2 atmosphere at room temperature for 18 h. The reaction mixture was filtered through Celite and concentrated under reduced pressure. The residue was purified by silica column chromatography ($MeOH/CH_2Cl_2$ 1:5 and 1:3) followed by SPE-C18 column chromatography ($MeOH/H_2O$ 2:8 to 9:1 gradient) to afford **6** as a semisolid (5.3 mg, 74%): $[\alpha]^{24}_D$ +15.0 (*c* 1.04 MeOH); IR (neat) 3370, 3311, 2921, 2852, 1680, 1467, 1106, 1035 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 3.90 (m, 3H), 3.62 (td, *J* = 8.2, 6.0 Hz, 1H), 3.58 (dd, *J* = 8.4, 4.2 Hz, 1H), 2.84 (dd, *J* = 14.8, 5.3 Hz, 1H), 2.73 (dd, *J* = 14.8, 2.7 Hz, 1H), 2.68 (d, *J* = 6.0 Hz, 1H), 2.67 (d, *J* = 8.2 Hz, 1H), 2.59 (t, *J* = 7.7 Hz, 2H), 1.55 (m, 2H), 1.32 (brs, 12H), 0.91 (t, *J* = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, CD_3OD) δ 79.7, 78.3, 73.8, 73.2, 72.4, 61.2, 59.1, 57.0, 33.1, 30.7, 30.6, 30.5, 28.3, 28.0, 23.8, 14.5; HRMS (ESI) calcd for $[C_{16}H_{33}NO_5 + H]^+$ 320.2431, found 320.2428.

*(R,E)-3-(2,2-Dimethyl-1,3-dioxolan-4-yl)prop-2-en-1-ol (**29**).* L-Glyceraldehyde acetonide (**28**) was obtained from of L-gulono-1,4-lactone as described in the literature.[34,35] The aldehyde **28** (3.07 g, 23.6 mmol) in benzene was added to a refluxing solution of (carbethoxymethylene)triphenylphosphorane (12.3 g, 35.4 mmol) in benzene (35 mL) via cannula. The reaction mixture was refluxed overnight and cooled to room temperature. Benzene was evaporated under reduced pressure, and the resulting residue was triturated with Et_2O to separate the insoluble triphenylphosphine oxide. The ether portions were combined and concentrated. The crude product was then purified by flash silica gel column chromatography on silica gel using hexanes/EtOAc (9:1) to afford 3.5 g (74%) of the *E*-isomer and 0.4 g (8%) of the *Z*-isomer. To a solution of the *E*-ester (3.5 g, 17.5 mmol) in anhydrous CH_2Cl_2 (100 mL) was added dropwise DIBAL-H (1 M solution in hexanes, 38.5 mmol, 38.5 mL) at −78 °C. The solution was stirred for 1 h at the same temperature and allowed to warm to 0 °C. After completion of the reaction (monitored by TLC), methanol was added slowly (about 2 mL) followed by addition of a cold aqueous saturated potassium tartrate solution. The biphasic mixture was stirred for 2 h and extracted with EtOAc. The combined organic extracts were dried over anhydrous sodium sulfate and purified by column chromatography to give 2.5 g (93%) in 69% overall yield of the allylic alcohol **29** as a colorless oil: $[\alpha]^{22}_D$ −24.2 (*c* 1.01, MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 5.90 (td, *J* = 5.0, 15.5 Hz, 1H), 5.67 (dd, *J* = 7.4, 15.5 Hz, 1H), 4.52 (q, *J* = 7.1 Hz, 1H), 4.07 (d, *J* = 6.1 Hz, 3H), 3.85 (s, 1H), 3.57 (t, *J* = 7.9 Hz, 1H), 1.41 (s, 3H), 1.37 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 133.7, 127.8, 109.3, 76.5, 69.3, 61.9, 26.6, 25.8; HRMS (ESI^+, M + Na) *m*/*z* calcd for $[C_8H_{14}NaO_3^+]$ 181.0841, found 181.0839.

*(S)-4-((2R,3S)-3-(Benzyloxymethyl)oxiran-2-yl)-2,2-dimethyl-1,3-dioxolane (**30**).* To a −40 °C suspension of titanium isopropoxide (0.38 mL, 1.30 mmol) and powdered, activated 3 Å molecular sieves

dx.doi.org/10.1021/jo202054g | *J. Org. Chem.* 2012, 77, 3082−3098

(1 g) in CH_2Cl_2 (5 mL) was added a solution of (+)-diisopropyl tartrate (0.303 mL, 1.45 mmol) in CH_2Cl_2 (2 mL). The mixture was stirred for 40 min at −40 °C, and then a solution of **29** (2.3 g, 15 mmol) in CH_2Cl_2 (2 mL) was added. After 1.5 h, cumene hydroperoxide (6.5 mL, 44 mmol) was added dropwise over 3 min. The resulting solution was stirred for 89 h at −40 °C, cooled to −78 °C, and stirred for 10 min. Bu_3P (7.27 mL, 28.0 mmol) was added dropwise over 10 min to quench the reaction. The mixture was stirred for 30 min and was then treated with citric acid monohydrate (302 mg, 1.46 mmol) dissolved in acetone−ether (1:9, 21 mL). The cooling bath was removed, and the resulting mixture was stirred for an additional 40 min. After filtration through a pad of Celite, the filtrate was dried over $MgSO_4$, concentrated and purified by silica gel flash column chromatography (33% EtOAc/hexanes) to furnish 2 g (79%) of the epoxide. To a 0 °C suspension of sodium hydride (0.46 g, 60% in oil, 12 mmol) in THF (30 mL) was added a solution of the epoxide (2 g, 11.5 mmol) in THF (40 mL), followed by benzyl bromide (1.64 mL, 13.8 mmol) and tetrabutylammonium iodide (46 mg, 56.8 m mol). The mixture was stirred for 8 h at room temperature, and then water (50 mL) was added over 15 min. The phases were separated, and the aqueous phase was further extracted with CH_2Cl_2. The combined organic extracts were washed with brine (50 mL), dried over sodium sulfate, and concentrated under reduced pressure. Purification of the crude product by flash silica gel column chromatography (10% EtOAc/hexanes) provided 2.9 g (96% and 76% yield over two steps) of benzyl ether **30** as a colorless oil: $[\alpha]^{22}_D$ −27.6 (*c* 1.00, MeOH). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26−7.31 (m, 5H), 1.33 (s, 3H), 4.53 (q, *J* = 6.4 Hz, 2H), 4.04 (t, *J* = 13.3 Hz, 1H), 3.82−3.89 (m, 2H), 3.77 (dd, *J* = 2.00, 11.6 Hz, 1H), 3.42 (dd, *J* = 5.6, 11.6 Hz, 1H), 3.07 (d, *J* = 2.5 Hz, 1H), 2.93 (d, *J* = 5.0 Hz, 1H,), 1.41 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.8, 128.4, 127.7, 109.9, 75.5, 73.1, 69.4, 66.7, 55.8, 55.2, 26.5, 25.3; HRMS (ESI) calcd for $[C_{15}H_{20}O_4 + H]^+$ 265.1440, found 265.1446.

*(S)-4-((1R,2R)-2-Azido-1,3-bis(benzyloxy)propyl)-2,2-dimethyl-1,3-dioxolane (**31**).* A solution of the epoxy alcohol **30** (2.2 g, 8.3 mmol) in a 2-methoxyethanol/water mixture (8:l, 94 mL) was refluxed for 5 h with sodium azide (0.0289 g, 41.6 mmol) and ammonium chloride (0.019 g, 33 mmol). The reaction mixture was cooled to room temperature and concentrated under reduced pressure, and the crude product was purified by flash silica gel column chromatography. To a suspension of NaH (0.30 g, 60% in oil, 7.8 mmol) in THF (180 mL) was added a solution of the azide derivative (2.26 g, 7.4 mmol) in THF (40 mL), followed by benzyl bromide (1.00 mL, 0.0944 mmol). The mixture was stirred for 1.5 h at rt, and then H_2O (50 mL) was added over 15 min. The phases were separated and the aqueous phase was further extracted with Et_2O (2 × 100 mL). The combined organic extracts were washed with brine (50 mL), dried ($MgSO_4$) and concentrated under reduced pressure. Purification of the crude product by silica gel flash column chromatography (6% EtOAc/hexanes) provided 2.6 g (78% yield over two steps) of compound **31** as colorless oil: $[\alpha]^{23}_D$ −19.6 (*c* 1.00, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.33−7.37 (m, 10H), 4.70 (d t, *J* = 12.7 Hz, 6.04 Hz, 2H), 4.60 (d, *J* = 6.0 Hz, 6.4 Hz 2H), 4.20 (q, *J* = 6.3 Hz, 1H), 4.05 (dd, *J* = 6.4, 8.3 Hz, 1H), 3.90 (d, *J* = 2.2 Hz, 2H), 3.68−3.76 (m, 3H), 1.42 (s, 3H), 1.37 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.7, 128.5, 128.5, 127.9, 127.7, 127.7, 109.3, 79.1, 75.2, 73.9, 73.4, 69.6, 66.5, 62.5, 26.6, 25.3; HRMS (ESI) calcd for $[C_{22}H_{27}N_3O_4 + H]^+$ 398.2080, found 398.2076.

*N-((1R,2R)-1,3-Bis(benzyloxy)-1-((S)-2,2-dimethyl-1,3-dioxolan-4-yl)propan-2-yl)-4-methylbenzenesulfonamide (**32**).* A solution of the azide **31** (2.4 g, 6.0 mmol) in THF (18 mL) was added dropwise to a stirred suspension of $LiAlH_4$ in THF (2 M solution in THF, 13 mL, 13 mmol) at −78 °C under argon. The reaction mixture was stirred for 20 min, warmed up to 0 °C and then a 10% aqueous sodium solution was added dropwise and diluted with CH_2Cl_2. The biphasic system was stirred for 5 min and then separated. The aqueous phase was further extracted with chloroform and the combined organic phases were washed with brine and dried over sodium sulfate. Concentration of the organic layer under reduced pressure afforded the amine. The amine (2.12 g, 5.74 mmol) was taken into anhydrous CH_2Cl_2. Triethylamine (1.00 mL, 7.46 mmol) was added followed by tosyl chloride (1.42 g, 7.46 mmol) (as a solid) and stirred at room temperature for 4 h. Water was used to quench the reaction. The phases were separated and the aqueous phase was further extracted with CH_2Cl_2, dried over sodium sulfate and concentrated under reduced pressure. Purification of the crude product by silica gel flash column chromatography (30% EtOAc/hexanes provided 2.87 g (90%) of compound **32** as a colorless oil over two steps: $[\alpha]^{24}_D$ +3.74 (*c* 1.02, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.64−7.67 (m, 2H), 7.13−7.28 (m, 12H), 4.72 (d, *J* = 11.2 Hz, 1H), 4.56 (d, *J* = 11.3 Hz, 1H), 4.20−4.28 (m, 3H), 3.97 (dd, *J* = 6.7, 8.0 Hz, 1H), 3.81−3.87 (m, 2H), 3.52 (dd, *J* = 4.5, 9.5 Hz, 1H), 3.35−3.38 (m, 1H), 3.14 (dd, *J* = 4.4, 9.5 Hz, 1H), 2.33 (s, 3H), 1.42 (s, 3H), 1.37 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.4, 138.1, 137.6, 137.2, 129.5, 128.4, 127.9, 127.7, 127.1, 109.0, 78.1, 76.1, 74.7, 73.1, 67.4, 65.5, 54.6, 26.5, 25.1, 21.5; HRMS (ESI) calcd for $[C_{29}H_{35}NO_6S + H]^+$ 526.2263, found 526.2270.

*N-((2R,3R,4S)-1,3-Bis(benzyloxy)-5-(tert-butyldimethylsilyloxy)-4-hydroxypentan-2-yl)-4-methylbenzenesulfonamide (**33**).* A solution of acetonide **32** (2.5 g, 4.8 mmol) was taken into a 1:1 solution of methanol and 2 N HCl and stirred at 40 °C. The reaction was monitored by TLC and following completion was quenched by saturated solution of sodium bicarbonate, extracted with CH_2Cl_2, washed with brine, dried over $MgSO_4$, and concentrated in vacuum to give the crude product. The crude diol (2.4 g, 4.5 mmol) was taken into dry CH_2Cl_2. The solution was cooled to 0 °C, and triethylamine (0.75 mL, 5.4 mmol) was added followed by *tert*-butyldimethylsilyl chloride (1 M solution in CH_2Cl_2, 5.4 mL, 5.4 mmol) and a catalytic amount of DMAP. The reaction was stirred at 0 °C, and after 1 h, water was added to quench the reaction. The reaction mixture was extracted with CH_2Cl_2, dried over sodium sulfate and concentrated under reduced pressure. Purification of the crude product was accomplished by silica gel flash column chromatography using 25% EtOAc/hexanes to provide compound **33** as a colorless oil in 85% yield (2.42 g): $[\alpha]^{23}_D$ −5.2 (*c* 0.67, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.66 (d, *J* = 8.2 Hz, 2H), 7.13−7.28 (m, 12H), 4.57 (d, *J* = 4.8 Hz, 2H), 4.26 (d, *J* = 4.5 Hz, 2H), 3.65−3.77 (m, 4H), 3.55 (ddd, *J* = 5.3, 9.6, 18.9 Hz, 2H), 3.38 (dd, *J* = 5.2, 9.8 Hz, 1H), 2.31 (s, 3H), 0.88 (s, 9H), 0.01 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.1, 138.3, 137.6, 137.5, 129.6, 128.9, 128.5, 128.1, 127.9, 127.3, 78.6, 73.8, 72.9, 71.6, 68.3, 63.6, 54.2, 25.9, 25.9, 21.5, 18.2, −5.4,-5.4; HRMS (ESI$^+$) *m*/*z* calcd for $[C_{32}H_{45}NO_6SSi + H]^+$ 600.2815, found 600.2815.

*(2R,3R,4R)-3-(Benzyloxy)-2-(benzyloxymethyl)-4-((tert-butyldimethylsilyloxy)methyl)azetidine (**34**).* Sulfonamide **33** (2.2 g, 3.67 mmol) and triphenylphosphine (1.45 g, 5.5 mmol) were dissolved in dry CH_2Cl_2 under argon/nitrogen. Diisopropyl azodicarboxylate (0.90 mL, 5.5 mmol) was added dropwise at 0 °C with stirring. The solution was warmed to room temperature and stirred for 16 h. The reaction mixture was then filtered through a pad of silica and concentrated in vacuo. The crude mixture was purified by flash silica gel column chromatography to yield the corresponding *N*-tosylazetidine as a colorless oil. A 1.5 g (2.6 mmol) portion of the *N*-tosylazetidine was dissolved in dry DME (26 mL) and the resulting solution cooled to −60 °C. To this solution was added dropwise a dark-green solution of Na/naphthalene in a dry DME (0.25 M solution prepared by the addition of 0.89 g of Na in a 0.25 M solution of naphthalene (5 g) in DME until the dark-green color persisted. After 30 min, brine was added to the solution, and the aqueous phase was extracted with EtOAc. The organic phase was dried (Na_2SO_4) and concentrated in vacuum and the crude product filtered through a small pad of silica (5% EtOAc/hexanes to remove naphthalene and then pure EtOAc to elute azetidine **34** as a pale yellow oil in 70% yield over two steps (0.96 g): $[\alpha]^{23}_D$ −22 (*c* 0.67, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27−7.33 (m, 10H), 4.46−4.52 (m, 4H), 4.18−4.21 (m, 1H), 3.87−3.96 (m, 3H), 3.75 (d, *J* = 6.6 Hz, 1H), 3.47−3.49 (d, *J* = 4.1 Hz, 2H), 0.88 (s, 9H), 0.01 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.2, 128.4, 127.7, 74.7, 73.3, 72.4, 71.8, 64.0, 62.3, 61.6, 25.9, 18.3, −5.4; HRMS (ESI) calcd for $[C_{25}H_{37}NO_3Si + H]^+$ 428.2621, found 428.2615.

dx.doi.org/10.1021/jo202054g | *J. Org. Chem.* 2012, 77, 3082−3098

*((2R,3S,4R)-3-(Benzyloxy)-4-(benzyloxymethyl)-1-butylazetidin-2-yl)methanol (**35a**) and ((2R,3S,4R)-3-(Benzyloxy)-4-(benzyloxymethyl)-1-nonylazetidin-2-yl)methanol (**35b**).* The azetidine **34** (0.8 g, 1.9 mmol) and the aldehyde (butyraldehyde or nonyl aldehyde, 1.8 mmol ((0.16 mL for butyraldehyde and 0.30 mL for nonaldehyde) were mixed in $ClCH_2CH_2Cl$ and then treated with solid sodium triacetoxyborohydride (0.57 g, 2.7 mmol). The mixture was stirred at rt under a nitrogen atmosphere for 16 h. The reaction mixture was quenched by adding aqueous saturated sodium bicarbonate. The product was extracted with CH_2Cl_2, dried over sodium sulfate, concentrated under reduced pressure, and purified by silica gel flash column chromatography. A mixture of the alkylated product, butyl derivative (0.69 g, 1.4 mmol) or nonyl derivative (0.81 g, 1.5 mmol) and TBAF (1 M in THF, 1.5 equiv, 2.25 mL) was stirred at room temperature for 4 h. The solvent was removed under reduced pressure and the crude product was purified by flash silica gel column chromatography using 100% EtOAc to provide compound **35a** (0.5 g, 70% or **35b** (0.6 g, 72%))) as a pale brown oil. **35a**: $[\alpha]^{25}_D$ −18 (*c* 0.67, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.24−7.34 (m, 10H), 4.44−4.61 (m, 4H), 4.22 (dd, *J* = 5.2, 6.8 Hz, 1H), 4.06 (dd, *J* = 5.1, 12.4 Hz, 1H), 3.90 (dd, *J* = 3.0, 12.4 Hz. 1H), 3.72−3.76 (m, 2H), 3.46−3.48 (m, 2H), 2.76−2.80 (m, 1H), 2.62−2.65 (m, 1H), 1.30−1.33 (m, 4H), 0.88 (t, *J* = 7.1 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.2, 137.6, 128.5, 128.4, 127.9, 127.7, 127.6, 74.9, 73.4, 71.8, 71.6, 71.1, 64.8, 60.4, 49.7, 31.5, 20.6, 14.1; HRMS (ESI) calcd for $[C_{23}H_{31}NO_3 + H]^+$ 370.2382, observed 370.2378.

35b: $[\alpha]^{23}_D$ −10 (*c* 0.40, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.24−7.34 (m, 10 H), 4.45−4.60 (m, 4H), 4.24 (dd, *J* = 5.5, 6.3 Hz, 1H), 4.06 (dd, *J* = 5.1, 12.4 Hz, 1H), 3.92 (dd, *J* = 3.1, 12.5 Hz, 1H), 3.70−3.76 (m, 2H), 3.47−3.49 (m, 2H), 2.77−2.80 (m, 1H), 2.64−2.67 (m, 1H), 1.24−1.36 (m, 14H), 0.86 (t, *J* = 7.1 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.1, 137.6, 128.5, 127.9, 127.7, 74.6, 73.4, 71.9, 71.2, 71.0, 65.1, 60.2, 50.1, 31.8, 29.5, 29.3, 29.0, 27.4, 22.6,14.1; HRMS (ESI) calcd for $[C_{28}H_{41}NO_3 + H]^+$ 440.3165, found 440.3169.

*((2R,4R)-1-Butyl-3-hydroxyazetidine-2,4-diyl)dimethanol (**22**) and ((2R,4R)-3-Hydroxy-1-nonylazetidine-2,4-diyl)dimethanol (**23**).* The dibenzyl compound (200 mg of **35a** or **35b,** 0.5 and 0.45 mmol) was dissolved in methanol, and palladium chloride (10 mol %) was added and the reaction mixture was subjected to hydrogenation (balloon) at room temperature for 4 h. The reaction mixture was filtered through Celite and the solvent removed under reduced pressure. The crude product was then washed with hexanes and dried over sodium sulfate to furnish compounds **22** and **23** as colorless oils in 65% (60 mg) and 70% yield (81 mg) respectively. **22**: $[\alpha]^{24}_D$ −5.0 (*c* 0.20, MeOH); ^{1}H NMR (400 MHz, DMSO-d_6) δ 4.49 (t, *J* = 6.2 Hz, 1H), 4.06−4.23 (m, 3H), 3.80−3.91 (m, 3H), 3.15−3.23 (m, 2H), 1.49−1.59 (m, 2H), 1.3−1.32 (m, 2H), 0.88 (t, *J* = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 77.1, 71.9, 64.3, 58.8, 57.6, 50.6, 28.4, 20.9, 13.9; HRMS (ESI) calcd for $[C_9H_{19}NO_3 + H]^+$ 190.1443, found 190.1445. **23:** $[\alpha]_D{}^{25}$-6.6 (*c* 0.60, MeOH); ^{1}H NMR (400 MHz, DMSO-d_6) δ 4.48 (t, *J* = 6.3 Hz, 1H), 4.06−4.18 (m, 3H), 3.77−3.91 (m, 3H), 3.12−3.19 (m, 2H), 1.50−1.57 (m, 2H), 1.19−1.22 (m, 12H), 0.78 −0.80 (t, 7.2 Hz, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 77.1, 71.7, 64.5, 59.1, 57.7, 50.1, 33.1, 30.5, 30.3, 30.2, 27.8, 26.6, 23.7, 14.4; HRMS (ESI) calcd for $[C_{14}H_{29}NO_3 + H]^+$ 260.2226, found 260.2225.

General Procedures for Compounds 37−41a, 41b, 24, and 25. The synthesis of compounds **37**−**41a**, **41b**, **24**, and **25** (Scheme 8) follows the procedures for the synthesis of compounds **31**−**35a**, **35b**, **22**, and **23** (Scheme 7).

*(R)-4-((1R,2R)-2-Azido-1,3-bis(benzyloxy)propyl)-2,2-dimethyl-1,3-dioxolane (**37**).* Compound **36** was synthesized as described previously in the literature.[36] Compound **37** was obtained as a colorless oil in 70% yield: $[\alpha]^{22}_D$ −3.4 (*c* 1.0, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.29−7.37 (m, 10H), 4.55−4.73 (m, 4H), 4.25−4.28 (m, 1H), 3.98−4.02 (m, 1H), 3.70−3.82 (m, 3H), 3.54−3.60 (m, 2H), 1.42 (s, 3H), 1.37 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) 137.8, 137.6, 129.5, 128.4, 128.3, 127.8, 127.1, 109.3, 78.3, 74.5, 73.4, 69.1, 65.9, 62.1, 26.4, 25.6; HRMS (ESI) calcd for $[C_{22}H_{27}N_3O_4 + H]^+$ 398.2080, found 398.2087.

*N-((1R,2R)-1,3-Bis(benzyloxy)-1-((R)-2,2-dimethyl-1,3-dioxolan-4-yl)propan-2-yl)-4-methylbenzenesulfonamide (**38**).* Compound **38** was obtained as a colorless oil in 90%: $[\alpha]^{22}_D$ +7.8 (*c* 0.72, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.65−7.68 (m, 2H), 7.17−7.32 (m, 12H), 4.24−4.46 (m, 4H), 4.10 (q, *J* = 7.15 Hz, 1H), 3.94 (dd, *J* = 6.6, 8.1 Hz, 1H), 3.53−3.65 (m, 3H), 3.43 (t, *J* = 4.88 Hz, 1H), 3.34 (dd, *J* = 7.02, 9.46 Hz, 1H), 2.31 (s, 3H), 1.39 (s, 3H), 1.30 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.2, 137.8, 137.4, 129.5, 128.4, 128.3, 127.8, 127.1, 109.4, 76.5, 73.2, 73.1, 68.8, 66.2, 60.3, 53.9, 26.2, 25.6, 21.4, 20.1, 14.2; HRMS (ESI) calcd for $[C_{29}H_{35}NO_6S + H]^+$ 526.2263, found 526.2267.

*N-((2R,3R,4R)-1,3-Bis(benzyloxy)-5-(tert-butyldimethylsilyloxy)-4-hydroxypentan-2-yl)-4-methylbenzenesulfonamide (**39**).* Compound **39** was obtained as a colorless oil in 89% yield: $[\alpha]^{23}_D$ −18 (*c* 0.51, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.66 (d, *J* = 8.2 Hz, 2H), 7.13−7.28 (m, 12H), 4.57 (d, *J* = 11.1 Hz, 2H), 4.25 (d, *J* = 4.5 Hz, 2H), 3.65−3.77 (m, 4H), 3.56 (ddd, *J* = 5.3, 9.6, 18.9 Hz, 2H), 3.38 (dd, *J* = 5.2, 9.8 Hz, 1H), 2.31 (s, 3H) 0.88 (s, 9H) 0.01 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.3, 137.9, 137.5, 137.4, 129.6, 128.4, 128.3, 127.9, 127.8, 127.7, 127.1, 75.6, 73.5, 73.1, 70.3, 68.1, 63.1, 63.5, 60.3, 53.3, 25.9, 21.5, 21.1, 18.2, 14.2, −5.4; HRMS (ESI) calcd for $[C_{32}H_{45}NO_6SSi + H]^+$ 600.2815, found 600.2819.

*(2R,3R,4S)-3-(Benzyloxy)-2-(benzyloxymethyl)-4-((tert-butyldimethylsilyloxy)methyl)azetidine (**40**).* Compound **40** was obtained as a colorless oil in 53% yield: $[\alpha]^{23}_D$ +21 (*c* 0.67, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.20−7.29 (m, 10H), 4.41−4.52 (m, 4H), 3.79−3.86 (m, 3H), 3.76 (d, *J* = 6.6 Hz, 1H), 3.54−3.56 (m, 2H), 3.41−3.44 (m, 2H), 0.84 (s, 9H), 0.01 (6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.2, 128.4, 127.7, 74.7, 73.3, 72.4, 71.8, 64.0, 62.3, 61.6, 25.9, 18.3, −5.4; HRMS (ESI) calcd for $[C_{25}H_{37}NO_3Si + H]^+$ 428.2621, found 428.2621.

*((2R,3S,4S)-3-(Benzyloxy)-4-(benzyloxymethyl)-1-butylazetidin-2-yl)methanol (**41a**).* Compound **41a** was obtained as a pale yellow oil in 73% yield: $[\alpha]^{23}_D$ +19 (*c* 1.0, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.28- 7.34 (m, 10H), 4.42−4.56 (m, 4H), 4.23 (dd, *J* = 5.2, 6.8 Hz, 1H), 4.06 (dd, *J* = 5.1, 12.4 Hz, 1H), 3.91 (t, *J* = 5.3 Hz, 1H), 3.41−3.57 (m, 2H), 3.11−3.17 (m, 2H), 2.56−2.65 (m, 2H), 1.26−1.38 (m, 4H), 0.87 (t, *J* = 7.2 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.2, 138.0, 128.4, 128.4, 127.8, 127.6, 127.5, 72.6, 71.7, 71.5, 71.2, 69.9, 61.1, 57.7, 30.5, 20.5, 13.9; HRMS (ESI) calcd for $[C_{23}H_{31}NO_3 + H]^+$ 370.2382, found 370.2387.

*((2R,3S,4S)-3-(Benzyloxy)-4-(benzyloxymethyl)-1-nonylazetidin-2-yl)methanol (**41b**).* Compound **41b** was obtained as a pale yellow oil in 80% yield: $[\alpha]^{23}_D$ +20 (*c* 1.1, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26−7.34 (m, 10H), 4.46−4.53 (m, 4H), 3.98 (d, *J* = 5.4 Hz, 1H), 3.37−3.64 (m, 6H), 2.78−2.82 (m, 2H), 1.24−1.34 (m, 14H), 0.87 (t, *J* = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.8, 137.4, 128.5, 128.4, 128.1, 127.9, 127.7, 73.4, 72.9, 71.9, 71.4, 70.7, 69.9, 60.3, 58.2, 31.8, 30.1, 29.4, 29.3, 29.2, 27.1, 26.5, 22.6, 14.1; HRMS (ESI) calcd for $[C_{28}H_{41}NO_3 + H]^+$ 440.3165, found 440.3169.

*((2R,3S,4S)-1-Butyl-3-hydroxyazetidine-2,4-diyl)dimethanol (**24**).* Compound **24** was obtained as a pale yellow oil in 74% yield: ^{1}H NMR (400 MHz, MeOD) δ 4.26 (t, *J* = 6.6 Hz, 1H), 4.00−4.04 (m, 2H), 3.81 (d, *J* = 4.2 Hz, 4H), 3.20−3.23 (m, 2H), 1.61−1.65 (m, 2H), 1.29−1.35 (m, 2H), 0.88 (t, *J* = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, MeOD) δ 77.5, 63.2, 59.2, 57.4, 49.9, 27.7, 20.8, 13.9; HRMS (ESI) calcd for $[C_9H_{19}NO_3 + H]^+$ 190.1443, found 190.1448.

*((2R,3S,4S)-3-Hydroxy-1-nonylazetidine-2,4-diyl)dimethanol (**25**).* Compound **25** was obtained as a pale yellow oil in 80% yield: ^{1}H NMR (400 MHz, MeOD) 4.25 (t, *J* = 6.6, 1H), 4.01−4.05 (m, 2H,), 3.79−3.82 (m, 4H), 3.20−3.24 (m, 2H), 1.63−1.66 (m, 2H), 1.20−1.28 (m, 12H), 0.80 (t, *J* = 6.8, 3H); ^{13}C NMR (100 MHz, MeOD) δ 77.5, 63.2, 59.3, 57.7, 49.9, 33.0, 30.5, 30.3, 30.2, 27.6, 25.8, 23.7, 14.5; HRMS (ESI) calcd for $[C_{14}H_{29}NO_3 + H]^+$ 260.2226, found 260.2223.

General Procedures for Compounds 43−47a, 47b, 26, and 27. The synthesis of compounds **43**−**47a**, **47b**, **26**, and **27** (Scheme 9) follows the procedures for the synthesis of compounds **31**−**35a**, **35b**, **22**, and **23** (Scheme 7).

dx.doi.org/10.1021/jo202054g | *J. Org. Chem.* 2012, 77, 3082−3098

*(R)-4-((1S,2S)-2-Azido-1,3-bis(benzyloxy)propyl)-2,2-dimethyl-1,3-dioxolane (**43**).* Compound **42** was synthesized as described previously in the literature.[36] Compound **43** was obtained from compound **42** as a colorless oil in 70% yield: $[\alpha]^{22}_{D}$ +20 (*c* 0.67, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.33−7.38 (m, 10H), 4.70 (d,d, *J* = 12.7 Hz, 6.1 Hz, 2H), 4.61 (d,d *J* = 6.0 Hz, 6.4 Hz, 2H), 4.20 (q, *J* = 6.3 Hz, 1H), 4.05 (dd, *J* = 6.4, 8.3 Hz, 1H), 3.90 (d, *J* = 2.2 Hz, 2H), 3.68−3.76 (m, 3H), 1.42 (s, 3H), 1.37(s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 137.8, 128.4, 127.8, 109.3, 79.1, 75.2, 73.9, 73.4, 69.6, 66.5, 62.5, 26.6, 25.2; $[\alpha]^{24}_{D}$ 20 (*c* 0.67, MeOH). HRMS (ESI) calcd for $[C_{22}H_{27}N_3O_4 + H]^+$ 398.2080, found 398.2082.

*N-((1S,2S)-1,3-Bis(benzyloxy)-1-((R)-2,2-dimethyl-1,3-dioxolan-4-yl)propan-2-yl)-4-methylbenzenesulfonamide (**44**).* Compound **44** was obtained as a colorless oil in 88% yield: $[\alpha]^{23}_{D}$ −3.9 (*c* 0.80, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.65−7.68 (m, 2H), 7.17−7.32 (m, 12H), 4.75 (d, *J* = 11.2 Hz, 1H), 4.57 (d, *J* = 11.3 Hz, 1H), 4.23−4.30 (m, 3H), 3.98 (dd, *J* = 6.7, 8.0 Hz, 1H), 3.82−3.88 (m, 2H), 3.54 (dd, *J* = 4.5, 9.5 Hz, 1H), 3.35−3.38 (m, 1H), 3.15 (dd, *J* = 4.4, 9.5 Hz, 1H), 1.42 (s, 3H), 2.39 (s, 3H), 1.31 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.5, 138.1, 137.5, 137.4, 129.6, 128.4, 127.9, 127.7, 127.2, 109.1, 75.9, 74.7, 73.1, 67.3, 65.5, 54.6, 26.4, 24.9, 21.5; HRMS (ESI) calcd for $[C_{29}H_{35}NO_6S + H]^+$ 526.2263, found 526.2265.

*N-((2S,3S,4R)-1,3-Bis(benzyloxy)-5-(tert-butyldimethylsilyloxy)-4-hydroxypentan-2-yl)-4-methylbenzenesulfonamide (**45**).* Compound **45** was obtained as a colorless oil in 87% yield: $[\alpha]^{23}_{D}$ +5.80 (*c* 1.02, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.65 (d, *J* = 8.2 Hz, 2H), 7.13−7.28 (m, 12H), 4.57 (d, *J* = 11.2 Hz, 2H), 4.25 (d, *J* = 4.5 Hz, 2H), 3.65−3.77 (m, 4H), 3.56 (ddd, *J* = 5.3, 9.6, 18.9 Hz, 2H), 3.38 (dd, *J* = 5.2, 9.8 Hz, 1H), 2.31 (s, 3H), 0.88 (s, 9H), 0.01 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 143.1, 138.1, 137.6, 137.5, 129.5, 128.3, 127.9, 127.7, 127.6, 127.2, 78.6, 73.7, 72.9, 71.5, 68.3, 63.6, 54.1, 25.9, 21.4, 18.2, −5.4; HRMS (ESI$^+$) calcd for $[C_{32}H_{45}NO_6SSi + H]^+$ 600.2815, found 600.2819.

*(2S,3S,4S)-3-(Benzyloxy)-2-(benzyloxymethyl)-4-((tert-butyldimethylsilyloxy)methyl)azetidine (**46**).* Compound **46** was obtained as a colorless oil in 55% yield: $[\alpha]^{22}_{D}$ −24.6 (*c* 2.02, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27−7.33 (m, 10H), 4.46−4.52 (m, 4H), 4.18−4.21 (m, 1H), 3.87−3.96 (m, 3H), 3.75 (d, *J* = 6.6 Hz, 1H), 3.48 (d, *J* = 4.1 Hz, 2H), 0.88 (s, 9H), 0.01 (6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.1, 128.4, 127.7, 73.3, 72.3, 71.8, 63.9, 62.3, 61.5, 25.9, 18.2, −5.3; HRMS (ESI) calcd for $[C_{25}H_{37}NO_3Si + H]^+$ 428.2621, found 428.2618.

*((2S,3R,4S)-3-(Benzyloxy)-4-(benzyloxymethyl)-1-butylazetidin-2-yl)methanol (**47a**).* Compound **47a** was obtained as a pale brown oil in 72% yield: $[\alpha]^{24}_{D}$ +18.6 (*c* 1.20, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.25- 7.34 (m, 10H), 4.45−4.61 (m, 4H), 4.23 (dd, *J* = 5.2, 6.8 Hz, 1H), 4.06 (dd, *J* = 5.1, 12.4 Hz, 1H), 3.91 (dd, *J* = 3.0, 12.4 Hz, 1H), 3.70−3.74 (m, 2H), 3.47−3.49 (m, 2H), 2.76−2.82 (m, 1H), 2.63−2.66 (m, 1H), 1.30−1.32 (m, 4H), 0.88 (t, *J* = 7.1 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.2, 137.6, 128.4, 127.9, 127.6, 74.9, 73.4, 71.9, 71.5, 71.1. 64.9, 60.4, 49.8, 31.4, 20.6, 14.1; HRMS (ESI) calcd for $[C_{23}H_{31}NO_3 + H]^+$ 370.2382, found 370.2383.

*((2S,3R,4S)-3-(Benzyloxy)-4-(benzyloxymethyl)-1-nonylazetidin-2-yl)methanol (**47b**).* Compound **47b** was obtained as a pale brown oil in 76% yield: $[\alpha]^{24}_{D}$ +17.5 (*c* 1.30, MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.24−7.34 (m, 10H), 4.45−4.61 (m, 4H), 4.22 (dd, *J* = 5.5, 6.3 Hz, 1H), 4.06 (dd, *J* = 5.1, 12.4 Hz, 1H), 3.91 (dd, *J* = 3.1, 12.5 Hz, 1H), 3.68−3.76 (m, 2H), 3.46−3.48 (m, 2H), 2.73−2.80 (m, 1H), 2.61−2.66 (m, 1H), 1.25−1.36 (m, 14H), 0.87 (t, *J* = 7.1 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 138.2, 137.7, 128.5, 127.7, 127.6, 74.9, 73.4, 71.9, 71.6, 71.1, 64.8, 60.4, 50.1, 31.9, 29.6, 29.4, 27.5, 22.7, 14.1; HRMS (ESI) calcd for $[C_{28}H_{41}NO_3 + H]^+$ 440.3165, found 440.3168.

*((2S,4S)-1-Butyl-3-hydroxyazetidine-2,4-diyl)dimethanol (**26**).* Compound **26** was obtained as a pale brown oil in 68% yield: $[\alpha]^{23}_{D}$ +5.5 (*c* 0.20, MeOH). ^{1}H NMR (400 MHz, MeOD) δ 4.52 (t, *J* = 6.2 Hz, 1H), 4.06−4.23 (m, 3H), 3.80−3.91 (m, 3H), 3.15−3.23 (m, 2H), 1.51−1.64 (m, 2H), 1.30−1.35 (m, 2H), 0.88 (t, *J* = 7.3 Hz, 3H); ^{13}C NMR (100 MHz, MeOD) δ 77.1, 71.9, 64.3, 58.8, 57.5, 50.5, 28.3, 20.1, 13.9; HRMS (ESI) calcd for $[C_9H_{19}NO_3 + H]^+$ 190.1443, found 190.1446.

*((2S,4S)-3-Hydroxy-1-nonylazetidine-2,4-diyl)dimethanol (**27**).* Compound **27** was obtained as a pale brown oil in 74% yield: $[\alpha]^{23}_{D}$ +6.9 (*c* 0.60, MeOH); ^{1}H NMR (400 MHz, MeOD) δ 4.48 (t, *J* = 6.3, 1H), 4.06−4.18 (m, 3H), 3.75−3.88 (m, 3H), 3.24−3.27 (m, 2H), 1.56−1.63 (m, 2H), 1.21−1.25 (m, 12H), 0.78−0.81 (t, 7.21, 3H); ^{13}C NMR (100 MHz, MeOD) δ 77.1, 71.7, 64.5, 59.1, 57.7, 50.9, 33.1, 30.7, 30.4, 27.8, 27.3, 26.6, 23.7, 14.4; HRMS (ESI) calcd for $[C_{14}H_{29}NO_3 + H]^+$ 260.2226, found 260.2220.

Microsome Preparations from C57BL/6 Mouse and LE Rat Testes. Testes in 5 g batches were placed in a 50 mL culture tube containing 25 mL of Reagent B (0.5 M Tris, 2.0 M Sucrose) and reagent A [Reagent A: 20 μL antipain, 20 μL leupeptin, 200 μL aprotinin, 110 μL APMSF, 372 mg KCl and 18.5 mL Milli-Q water (all protease inhibitors were made as 1 mg/mL stock)]. The testes were minced with scissors and then blended by 10 s bursts repeated 2−3 times at a time on Power Gen 700 while on ice. The homogenate was centrifuged at 7500 rpm for 10 min at 4 °C using a SW28 rotor (5660*g*). The resulting supernatant was collected and centrifuged at 23500 rpm for 1 h at 4 °C in a SW40 rotor. The supernatant was discarded and the pellet containing the microsomes was suspended in 600 μL of reagent D (reagent C, 200 mM DTT, 0.1 M EDTA, 10 mM UDP-glucose and 10% CHAPSO) and dispersed by passage through a 25 gauge needle followed by an insulin needle. The microsome suspension was stored as 100 μL aliquots in microcentrifuge tubes, flash frozen in liquid nitrogen for 1−2 min, kept at −80 °C, and used as needed. (Reagent C contained 250 μL of 10% *N*-laurosarcosine, 6.25 mL of 0.2 M HEPES, 5 mL of glycerol, 250 μL of 2% NaN_3, 250 μL of 0.1 M EDTA, 2.25 mL of reagent A, and 250 μL of 200 mM DTT)

Ceramide-Specific Glucosyltransferase Assay. The following solutions were added to each tube: 295 μL of Assay mix (50 mM HEPES; pH 7.4, reagent A, 5 mM $MnCl_2$, 10 mM phosphatidylcholine, 50 μM CBE, 1 mM EDTA, and 10 mM UDP-Glucose), 145 μL of water, 50 μL of iminosugar, and 100 μg of testicular microsomes. Control tubes contained the same components except microsomes. Reactions were initiated by the addition of 3 μL of BSA-ceramide, incubated at 37 °C for 30 min, and then terminated by addition of 1 mL of 2:1 (v/v) chloroform/methanol, vortexed, and incubated at room temperature for 30−60 min to allow phase separation. The upper phase and the midlayer were removed and discarded, and 500 μL of chloroform/methanol/water (3:48:47) was added to the bottom layer, which was vortexed and allowed to sit for 15 min at room temperature. The resulting upper phase was again removed, 100 μL of chloroform/methanol (2:1) was added, and then the sample tubes were dried in a vortex evaporator overnight.

Thin-Layer Chromatography (TLC). TLC plates were pretreated (Whatman silica gel 60 A, 20 × 20 cm, layer thickness 250 μm) by immersion in chloroform/methanol/water (50:50:15) for 5 min, air-dried for 10 min, and then immersed in 5% sodium borate (prepared in methanol) for 1 min, dried, and heated at 120 °C for 1.5 h. The dried sample tubes were reconstituted with 100 μL of chloroform/methanol (2:1) and vortexed, and 20 μL was then spotted onto the plates at the origin. The spotted plates were air-dried and placed in a sealed TLC chamber saturated with of chloroform/methanol/water (60:30:5) and run for approximately 1 h until the solvent reached within 1 cm from the top of the plate.

Detection and Quantitation of Substrate/Product. The TLC plate was documented using UV transilluminator (302 nm) and analyzed using AlphaEase (Fluorchem SP) software. The IDV values were plotted against iminosugar concentration using Sigma Plot 10. Linear regression plot was used to determine IC_{50} values.

Testicular Glucosidase Assay. The assay was carried out in 96-well plates. A 50 μL portion of 4-methylumbelliferyl β-D-glucoside (MUG; 3 mg/mL concentration) was added to each well using a multichannel pipet, followed by 10 μL of iminosugar dilutions (0, 5, 10, 50, 100, 500, and 1000 μM) added from left to the right so as to have increasing concentration of the iminosugar from top to the bottom of the plate. A 50 μL portion of testicular microsome

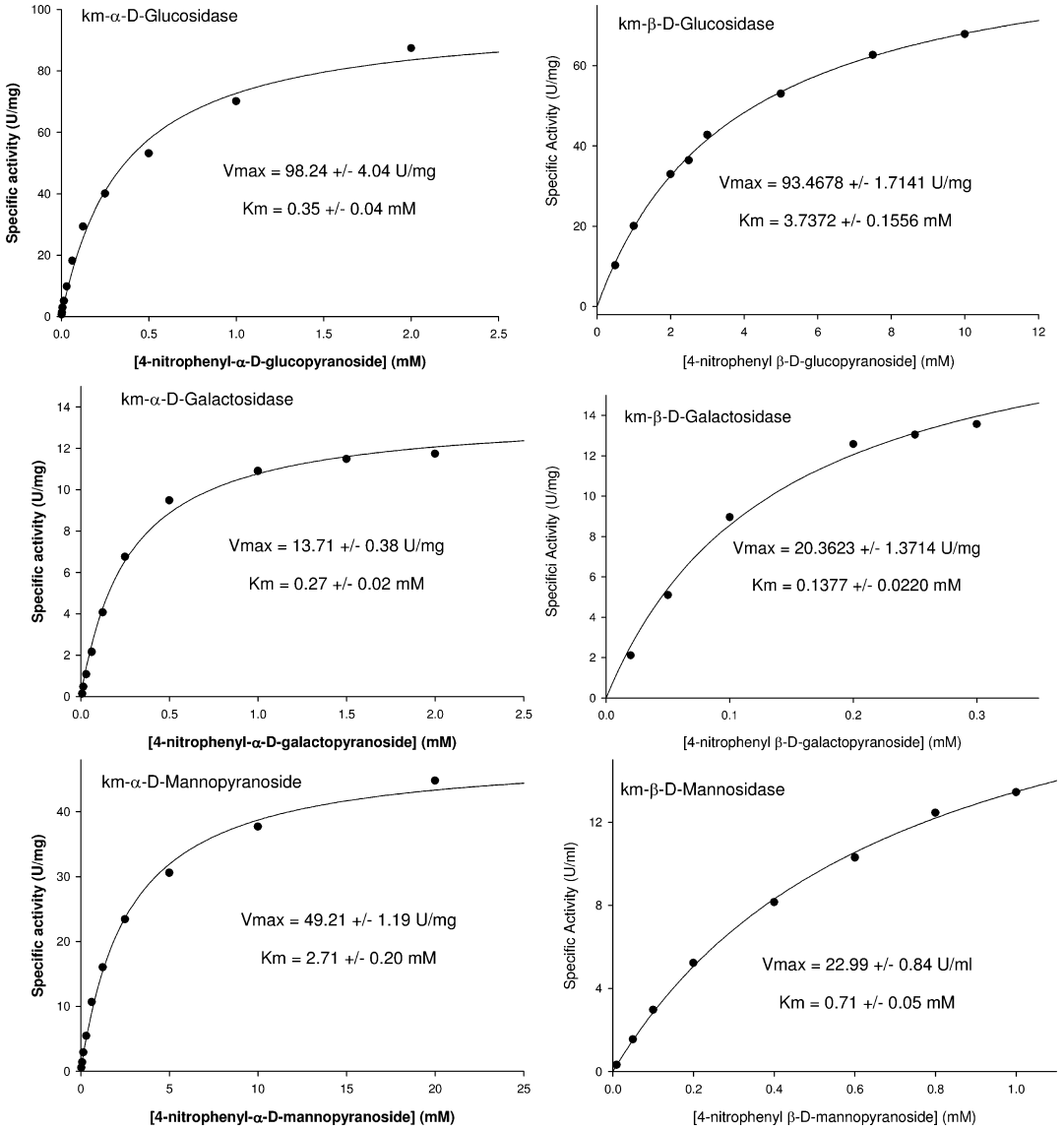


Figure 7. K_m determination for the enzyme/substrate combinations used for glycosidase inhibition assays (data were fit to the Michaelis–Menten equation).

(1 μg/μL) was then added to each well using a multichannel pipet in the first column. Another multichannel pipet was kept ready loaded with terminator solution (100 μL 1 M sodium carbonate, pH 10.7) and was added simultaneously to this row. Microsome was then added to the remaining rows after setting the timer to 1 min. Every 1 min, the terminator solution was added to each row until the 12th row. Absorbance was then detected at 360/460 nm using a Synergy HT multi-mode microplate reader. The absorbance values were subtracted from background (MUG only). Linear regression plot created using prism software (Graph Pad Prism 5) was used to determine the IC_{50} values.

Glycosidase Inhibition Assays. Chemicals and enzymes for the inhibition kinetics were purchased from a commercial supplier. Nonlinear regression analysis was performed using SigmaPlot (Systat Software, Inc., San Jose, CA). Assays were carried out in 96-well plate format, each well containing 2 μL of compound, 10 μL of substrate, and 78 μL of buffer. The reaction was started by addition of 10 μL of enzyme and incubated for 4–5 min at room temperature. The reaction was quenched by addition of 200 μL of 0.2 M sodium borate (pH 9.8). Buffer conditions, substrate, and enzyme concentrations were similar to those described.[37] The K_m values for each substrate/enzyme combination were determined experimentally (Figure 7), and for the inhibition assays the substrate concentration was equal to K_m. α-Glucosidase (*Saccharomyces cerevisiae*) was assayed at 0.49 μg/mL in sodium phosphate buffer (50 mM, pH 6.5) with 0.35 mM 4-nitrophenyl α-D-glucopyranoside. β-Glucosidase (almond) was assayed at 0.83 μg/mL in sodium acetate buffer (50 mM, pH 5.0) with 3.6 mM 4-nitrophenyl β-D-glucopyranoside. α-Galactosidase (green coffee beans) was assayed at 5.0 μg/mL in sodium phosphate buffer (50 mM, pH 6.5) with 0.27 mM 4-nitrophenyl α-D-galactopyranoside. β-Galactosidase (*Escherichia coli*) was assayed at 3.6 μg/mL in sodium phosphate buffer (50 mM, pH 7.3) with 0.13 mM 4-nitrophenyl β-D-galactopyranoside. α-mannosidase (jack bean) was assayed at 1.7 μg/mL in sodium citrate buffer (50 mM, pH 4.5) with 2.7 mM 4-nitrophenyl α-D-mannpyranoside. β-Mannosidase (Roman snail) was assayed at 4.1 μg/mL in acetate buffer (50 mM, pH 4.0) with 0.7 mM 4-nitrophenyl β-D-mannopyranoside. Absorbance was measured at 405 nm using a Spectra-Max 340PC plate reader (Molecular Devices, Sunnyvale, CA). IC_{50} values were determined by fitting data to eq 1, where A is the relative activity, [I] is the concentration of the compound, and n is the Hill slope coefficient.

$$A = \frac{1}{1 + \left(\frac{[\mathrm{I}]}{\mathrm{IC}_{50}}\right)^{n}} \quad (1)$$

dx.doi.org/10.1021/jo202054g | J. Org. Chem. 2012, 77, 3082–3098

■ ASSOCIATED CONTENT

Supporting Information

Copies of ^{1}H and ^{13}C NMR spectra of all new compounds and X-ray structural data. This material is available free of charge via the Internet at http://pubs.acs.org.

■ AUTHOR INFORMATION

Corresponding Author

*E-mail: georg@umn.edu.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We gratefully acknowledge financial support from the NIH (NICHD Reproductive Branch Contract HHSN275200503400C). We also thank Dr. Victor Young and Benjamin E. Kucera for obtaining X-ray crystallographic data, Dr. Ramappa Chakrasali for some initial efforts with the chemistry, and Brian Kern for help with the microsome preparations. We also thank Kwon Ho Hong for assistance with the modeling studies.

■ REFERENCES

(1) (a) Finer, L. B.; Henshaw, S. K. *Perspect. Sex. Repro. Health* **2006**, *38*, 90−96. (b) Jones, R. K.; Zolna, M. R. S.; Stanley, K.; Henshaw; Finer, L. B. *Perspect. Sex. Repro. Health* **2008**, *40*, 6−16. (c) Page, S. T.; Amory, J. K.; Bremner, W. J. *Endocr. Rev.* **2008**, *29*, 465−493. (d) Liu, P. Y.; Swerdloff, R. S.; Wang, C. *Contraception* **2010**, *82*, 471−475. (e) Wang, C.; Swerdloff, R. S. *Curr. Opin. Urol.* **2010**, *20*, 520−524.

(2) Mruk, D. D. *Trends Endocrinol. Metab.* **2008**, *19*, 57−64.

(3) Gu, Z. P.; Mao, B. Y.; Wang, Y. X.; Zhang, R. A.; Tan, Y. Z.; Chen, Z. X.; Cao, L.; You, G. D.; Segal, S. J. *Asian J. Androl.* **2000**, *2*, 283−287.

(4) Jones, A. R. *Aust. J. Biol. Sci.* **1983**, *36*, 333−350.

(5) (a) Tash, J. S.; Attardi, B.; Hild, S. A.; Chakrasali, R.; Jakkaraj, S. R.; Georg, G. I. *Biol. Reprod.* **2008**, *78*, 1127−1138. (b) Tash, J. S.; Chakrasali, R.; Jakkaraj, S. R.; Hughes, J.; Smith, S. K.; Hornbaker, K.; Heckert, L. L.; Ozturk, S. B.; Hadden, M. K.; Kinzy, T. G.; Blagg, B. S. J.; Georg, G. I. *Biol. Reprod.* **2008**, *78*, 1139−1152. (c) Su, L.; Cheng, C. Y.; Mruk, D. D. *Int. J. Biochem. Cell Biol.* **2010**, *42*, 1864−1875. (d) Chung, S. S.; Wang, X.; Roberts, S. S.; Griffey, S. M.; Reczek, P. R.; Wolgemuth, D. J. *Endocrinology* **2011**, *152*, 2492−502.

(6) van der Spoel, A. C.; Jeyakumar, M.; Butters, T. D.; Charlton, H. M.; Moore, H. D.; Dwek, R. A.; Platt, F. M. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 17173−17178.

(7) (a) Cox, T.; Lachmann, R.; Hollak, C.; Aerts, J.; van Weely, S.; Hrebicek, M.; Platt, F.; Butters, T.; Dwek, R.; Moyses, C.; Gow, I.; Elstein, D.; Zimran, A. *Lancet* **2000**, *355*, 1481−1485. (b) Elstein, D.; Hollak, C.; Aerts, J. M. F. G.; van Weely, S.; Maas, M.; Cox, T. M.; Lachmann, R. H.; Hrebicek, M.; Platt, F. M.; Butters, T. D.; Dwek, R. A.; Zimran, A. *J. Inherited Metab. Dis.* **2004**, *27*, 757−766. (c) Heitner, R.; Elstein, D.; Aerts, J.; van Weely, S.; Zimran, A. *Blood Cell Mol. Dis.* **2002**, *28*, 127−133. (d) Elstein, D.; Dweck, A.; Attias, D.; Hadas-Halpern, I.; Zevin, S.; Altarescu, G.; Aerts, J. F. M. G.; van Weely, S.; Zimran, A. *Blood* **2007**, *110*, 2296−2301. (e) Dulsat, C.; Mealy, N. *Drug Future* **2009**, *34*, 147−149.

(8) (a) Stutz, A. E. *Iminosugars as Glycosidase Inhibitors: Nojirimycin and Beyond*; Wiley-VCH: Weinheim, 1999. (b) Compain, P.; Martin, O. R. *Iminosugars: From Synthesis to Therapeutic Applications*; Wiley-VCH: Weinheim, 2007. (c) Horne, G.; Wilson, F. X.; Tinsley, J.; Williams, D. H.; Storer, R. *Drug Discov. Today* **2011**, *16*, 107−118. (d) Sorbera, L. A.; Castaner, J.; Bayes, M. *Drugs Future* **2003**, *28*, 229−236.

(9) Butters, T. D.; Dwek, R. A.; Platt, F. M. *Glycobiology* **2005**, *15*, R43−R52.

(10) Walden, C. M.; Sandhoff, R.; Chuang, C. C.; Yildiz, Y.; Butters, T. D.; Dwek, R. A.; Platt, F. M.; van der Spoel, A. C. *J. Biol. Chem.* **2007**, *282*, 32655−32664.

(11) Bone, W.; Walden, C. M.; Fritsch, M.; Voigtmann, U.; Leifke, E.; Gottwald, U.; Boomkamp, S.; Platt, F. M.; van der Spoel, A. C. *Reprod. Biol. Endocrinol.* **2007**, *5*, 1.

(12) Amory, J. K.; Muller, C. H.; Page, S. T.; Leifke, E.; Pagel, E. R.; Bhandari, A.; Subramanyam, B.; Bone, W.; Radlmaier, A.; Bremner, W. J. *Hum. Reprod.* **2007**, *22*, 702−707.

(13) (a) Butters, T. D.; van den Broek, L. A. G. M.; Fleet, G. W. J.; Krulle, T. M.; Wormald, M. R.; Dwek, R. A.; Platt, F. M. *Tetrahedron: Asymmetry* **2000**, *11*, 113−124. (b) Faugeroux, V.; Genisson, Y.; Andrieu-Abadie, N.; Colie, S.; Levade, T.; Baltas, M. *Org. Biomol. Chem.* **2006**, *4*, 4437−4439. (c) Butters, T. D.; Dwek, R. A.; Platt, F. M. *Chem. Rev.* **2000**, *100*, 4683−4696. (d) Butters, T. D.; Dwek, R. A.; Platt, F. M. *Curr. Top. Med. Chem.* **2003**, *3*, 561−574. (e) Mellor, H. R.; Platt, F. M.; Dwek, R. A.; Butters, T. D. *Biochem. J.* **2003**, *374*, 307−314. (f) van der Spoel, A. C. J., M.; Butters, T. D.; Dwek, R. A. U.S. Patent 20040019082, 2004. (g) Boucheron, C.; Desvergnes, V.; Compain, P.; Martin, O. R.; Lavi, A.; Mackeen, M.; Wormald, M.; Dwek, R.; Butters, T. D. *Tetrahedron: Asymmetry* **2005**, *16*, 1747−1756. (h) Li, H. Q.; Liu, T.; Zhang, Y. M.; Favre, S.; Bello, C.; Vogel, P.; Butters, T. D.; Oikonomakos, N. G.; Marrot, J.; Bleriot, Y. *ChemBioChem* **2008**, *9*, 253−260. (i) Deschamp, J.; Mondon, M.; Nakagawa, S.; Kato, A.; Alonzi, D. S.; Butters, T. D.; Zhang, Y.; Sollogoub, M.; Bleriot, Y. *Biorg. Med. Chem.* **2012**, *20*, 641−649.

(14) Ghisaidoobe, A.; Bikker, P.; de Bruijn, A. C. J.; Godschalk, F. D.; Rogaar, E.; Guijt, M. C.; Hagens, P.; Halma, J. M.; van't Hart, S. M.; Luitjens, S. B.; van Rixel, V. H. S.; Wijzenbroek, M.; Zweegers, T.; Donker-Koopman, W. E.; Strijland, A.; Boot, R.; van der Marel, G.; Overkleeft, H. S.; Aerts, J. M. F. G.; van den Berg, R. J. B. H. N. *ACS Med. Chem. Lett.* **2011**, *2*, 119−123.

(15) (a) Wennekes, T.; van den Berg, R. J. B. H. N.; Donker, W.; van der Marel, G. A.; Donker, W.; van der Marel, G. A.; Strijland, A.; Aerts, J. M. F. G.; Overkleeft, H. S. *J. Org. Chem.* **2007**, *72*, 1088−1097. (b) Ardes-Guisot, N.; Alonzi, D. S.; Reinkensmeier, G.; Butters, T. D.; Norez, C.; Becq, F.; Shimada, Y.; Nakagawa, S.; Kato, A.; Bleriot, Y.; Sollogoub, M.; Vauzeilles, B. *Org. Biomol. Chem.* **2011**, *9*, 5373−5388. (c) Wennekes, T.; Meijer, A. J.; Groen, A. K.; Boot, R. G.; Groener, J. E.; van Eijk, M.; Ottenhoff, R.; Bijl, N.; Ghauharali, K.; Song, H.; O'Shea, T. J.; Liu, H. L.; Yew, N.; Copeland, D.; van den Berg, R. J.; van der Marel, G. A.; Overkleeft, H. S.; Aerts, J. M. *J. Med. Chem.* **2010**, *53*, 689−698.

(16) (a) Diot, J. D.; Moreno, I. G.; Twigg, G.; Mellet, C. O.; Haupt, K.; Butters, T. D.; Kovensky, J.; Gouin, S. G. *J. Org. Chem.* **2011**, *76*, 7757−7768. (b) Orwig, S. D.; Tan, Y. L.; Grimster, N. P.; Yu, Z.; Powers, E. T.; Kelly, J. W.; Lieberman, R. L. *Biochemistry* **2011**, *50*, 10647−10657.

(17) Lenagh-Snow, G. M. J.; Araujo, N.; Jenkinson, S. F.; Rutherford, C.; Nakagawa, S.; Kato, A.; Yu, C. Y.; Weymouth-Wilson, A. C.; Fleet, G. W. J. *Org. Lett.* **2011**, *13*, 5834−5837.

(18) Pascher, I.; Sundell, S. *Chem. Phys. Lipids* **1977**, *20*, 175−191.

(19) Boyer, F. D.; Hanna, I.; Nolan, S. P. *J. Org. Chem.* **2001**, *66*, 4094−4096.

(20) Bernet, B.; Vasella, A. *Helv. Chim. Acta* **1979**, *62*, 1990−2016.

(21) Skaanderup, P. R.; Hyldtoft, L.; Madsen, R. *Monatsh. Chem.* **2002**, *133*, 467−472.

(22) AbdelMagid, A. F.; Carson, K. G.; Harris, B. D.; Maryanoff, C. A.; Shah, R. D. *J. Org. Chem.* **1996**, *61*, 3849−3862.

(23) (a) Michaut, A.; Rodriguez, J. *Angew. Chem., Int. Ed.* **2006**, *45*, 5740−5750. (b) Godin, G.; Garnier, E.; Compain, P.; Martin, O. R.; Ikeda, K.; Asano, N. *Tetrahedron Lett.* **2004**, *45*, 579−581. (c) Chang, M. Y.; Kung, Y. H.; Ma, C. C.; Chen, S. T. *Tetrahedron* **2007**, *63*, 1339−1344.

(24) Bergmeier, S. C.; Seth, P. P. *Tetrahedron Lett.* **1999**, *40*, 6181−6184.

(25) Liu, H. J.; Yip, J.; Shia, K. S. *Tetrahedron Lett.* **1997**, *38*, 2253−2256.

(26) Sharpless, K. B.; Amberg, W.; Bennani, Y. L.; Crispino, G. A.; Hartung, J.; Jeong, K. S.; Kwong, H. L.; Morikawa, K.; Wang, Z. M.; Xu, D. Q.; Zhang, X. L. *J. Org. Chem.* **1992**, *57*, 2768−2771.
(27) (a) Nash, R. J.; Kato, A.; Yu, C. Y.; Fleet, G. W. J. *Future Med. Chem.* **2011**, *3*, 1513−1521. (b) Tajima, Y.; Saito, S.; Ohno, K.; Tsukimura, T.; Tsujino, S.; Sakuraba, H. *J. Hum. Genet.* **2011**, *56*, 440−446. (c) Parenti, G.; Zuppaldi, A.; Pittis, M. G.; Tuzzi, M. R.; Annunziata, I.; Meroni, G.; Porto, C.; Donaudy, F.; Rossi, B.; Rossi, M.; Filocamo, M.; Donati, A.; Bembi, B.; Ballabio, A.; Andria, G. *Mol. Ther.* **2007**, *15*, 508−514. (d) Sugawara, K.; Tajima, Y.; Kawashima, I.; Tsukimura, T.; Saito, S.; Ohno, K.; Iwamoto, K.; Kobayashi, T.; Itoh, K.; Sakuraba, H. *Mol. Genet. Metab.* **2009**, *96*, 233−238. (e) Horne, G.; Wilson, F. X. *Prog. Med. Chem.* **2011**, *50*, 135−176.
(28) (a) Clinch, K.; Evans, G. B.; Frohlich, R. F. G.; Furneaux, R. H.; Kelly, P. M.; Legentil, L.; Murkin, A. S.; Li, L.; Schramm, V. L.; Tyler, P. C.; Woolhouse, A. D. *J. Med. Chem.* **2009**, *52*, 1126−1143. (b) Guan, R.; Ho, M. C.; Almo, S. C.; Schramm, V. L. *Biochemistry* **2011**, *50*, 1247−1254. (c) Basso, L. A.; Santos, D. S.; Shi, W.; Furneaux, R. H.; Tyler, P. C.; Schramm, V. L.; Blanchard, J. S. *Biochemistry* **2001**, *40*, 8196−8203.
(29) (a) Risseeuw, M. D. P.; van den Berg, R. J. B. H. N.; Donker-Koopman, W. E.; van der Marel, G. A.; Aerts, J. M. F. G.; Overhand, M.; Overkleeft, H. S. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 6600−6603. (b) Kato, A.; Kato, N.; Kano, E.; Adachi, I.; Ikeda, K.; Yu, L.; Okamoto, T.; Banba, Y.; Ouchi, H.; Takahata, H.; Asano, N. *J. Med. Chem.* **2005**, *48*, 2036−2044. (c) Kuriyama, C.; Kamiyama, O.; Ikeda, K.; Sanae, F.; Kato, A.; Adachi, I.; Imahori, T.; Takahata, H.; Okamoto, T.; Asano, N. *Bioorg. Med. Chem.* **2008**, *16*, 7330−7336. (d) Shibano, M.; Tsukamoto, D.; Fujimoto, R.; Masui, Y.; Sugimoto, H.; Kusano, G. *Chem. Pharm. Bull. (Tokyo)* **2000**, *48*, 1281−1285. (e) Schitter, G.; Scheucher, E.; Steiner, A. J.; Stutz, A. E.; Thonhofer, M.; Tarling, C. A.; Withers, S. G.; Wicki, J.; Fantur, K.; Paschke, E.; Mahuran, D. J.; Rigat, B. A.; Tropak, M.; Wrodnigg, T. M. *Beilstein J. Org. Chem.* **2010**, 6. (f) Paquette, L. A.; Moura-Letts, G.; Wang, G. P. *J. Org. Chem.* **2009**, *74*, 2099−2107.
(30) Bartoszewicz, A.; Kalek, M.; Stawinski, J. *Tetrahedron* **2008**, *64*, 8843−8850.
(31) Wang, W.; Li, X. L.; Zhang, P. Z.; Chen, H. *Chin. J. Chem.* **2008**, *26*, 208−212.
(32) Ko, K. S.; Zea, C. J.; Pohl, N. L. *J. Am. Chem. Soc.* **2004**, *126*, 13188−13189.
(33) Kleban, M.; Kautz, U.; Greul, J.; Hilgers, P.; Kugler, R.; Dong, H. Q.; Jäger, V. *Synthesis* **2000**, 1027−1033.
(34) Hubschwerlen, C. *Synthesis* **1986**, 962−964.
(35) Hubschwerlen, C.; Specklin, J.; Higelin, J. *Org. Synth.* **1998**, *9*, 454−456.
(36) (a) Takano, S.; Kurotaki, A.; Takahashi, M.; Ogasawara, K. *Synthesis* **1986**, 403−406. (b) Zhong, Y. L.; Shing, T. K. M. *J. Org. Chem.* **1997**, *62*, 2622−2624. (c) Kagawa, N.; Ihara, M.; Toyota, M. *Org. Lett.* **2006**, *8*, 875−878.
(37) Pandey, G.; Dumbre, S. G.; Khan, M. I.; Shabab, M. *J. Org. Chem.* **2006**, *71*, 8481−8488.