Pergamon

SCIENCE DIRECT®

Bioorganic & Medicinal Chemistry Letters 13 (2003) 2185–2189

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

3-Azido-3-deoxy-glycopyranoside Derivatives as Scaffolds for the Synthesis of Carbohydrate-Based Universal Pharmacophore Mapping Libraries

Rakesh Jain,[†] Muthoni Kamau,[‡] Chunguang Wang, Robert Ippolito,[§] Huiming Wang, Richard Dulina, Jan Anderson, David Gange[¶] and Michael J. Sofia*

Intercardia Research Labs, 8 Cedar Brooke Drive, Cranbury, NJ 08512, USA[‖]

Received 21 January 2003; accepted 24 March 2003

Abstract—The regiospecific syntheses of six monosaccharide scaffolds, **1**–**6**, containing a carboxylic acid, an azido and a free hydroxyl group were accomplished through the utilization of a key intermediate, namely, methyl 3-azido-3-deoxy-β-D-glucopyranoside (**10**). Scaffold **2** was also used in generating combinatorial libraries using solid-phase methodologies.

One aspect of combinatorial chemistry focuses on generating chemically diverse libraries suitable for broad screening. Our search for novel combinatorial platforms for preparing broad screening libraries lead us to investigate monosaccharides as versitile scaffolds. Conceptually, the use of a monosaccharide nucleus as a scaffold to display substituents in a stereochemically defined way was first pioneered by Hirschmann and coworkers in their effort to identify a mimetic of a known biologically active agent.[1,2]

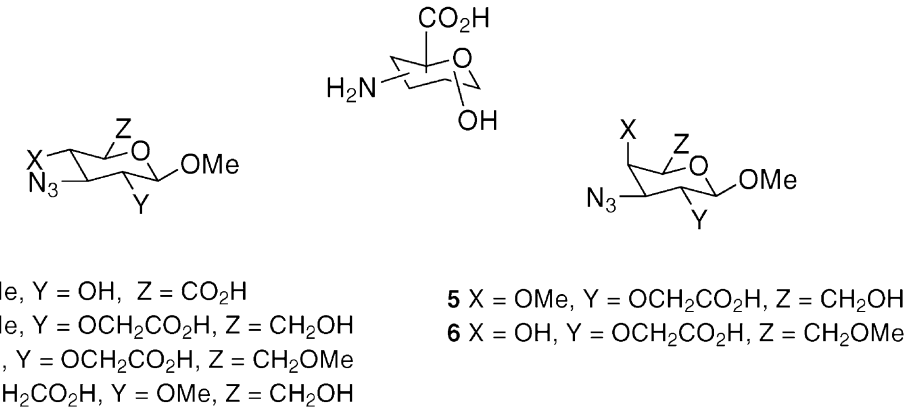
We were particularly attracted to monosaccharides as combinatorial scaffolds because of the unique set of characteristics they possess. The monosaccharide system provides a rigid ring system with a defined three dimensional spatial arrangement of substituents. The high functional density of this nucleus provides multiple sites for chemical modification. However, the difficulty faced in trying to chemically differentiate each of the hydroxyl groups on any natural sugar nucleus presents a problem if one wants to use a sugar nucleus as a combinatorial scaffold.[3]

In designing a practical monosaccharide scaffold system we focused on building a system that would provide a classical three-point pharmacophoric recognition motif (Fig. 1).[4,5] Then to circumvent the chemoselectivity problem, we used an easily differentiated functional group triad that included a carboxylic acid, a free hydroxyl group and a masked amino group. The two remaining hydroxyl groups on a hexose nucleus would be converted to their methyl ethers. Methyl ethers were chosen because they provide permanent protecting groups that are stable to a wide variety of chemical transformations and we hoped that they would minimize the contribution of these sites to molecular recognition events maximizing the effect of the three point motif. To accommodate a carboxylic acid at secondary carbon sites, we chose to use a carboxymethyl unit. Although not isosteric with a C-6 uronic acid, the carboxymethyl group could be easily introduced by direct alkylation of a sugar hydroxyl group.

In our earlier report, our scaffolds incorporated a fluorenylmethoxycarbonyl (Fmoc) protected amino group as the masked amino functionality.[4,5] However, it became apparent to us that the Fmoc group lacked the chemical stability we required for doing a wide variety of chemical transformations. To circumvent this problem, we chose to replace the NHFmoc group with an azido group.

*Corresponding author at current address: Bristol-Myers Squibb, 5 Research Parkway, Wallingford, CT 06492, USA. Fax: +1-203-677-6984; e-mail: michael.sofia@bms.com
[†]Current address: Versicor Inc., 34790 Ardentech Court, Fremont, CA 94555, USA.
[‡]Current address: Bristol-Myers Squibb, 206 and Provinceline Rd, Princeton, NJ 08543, USA.
[§]Current address: ChemRoutes Corp., 3912-116th St., Edmonton, Alberta, Canada T6J 1R4.
[¶]Current address: Row2 Technologies, Inc. 200 Lanidex Plaza, Parsippany, NJ 07054, USA.
[‖]Operations ceased September 2000.

0960-894X/03/$ - see front matter © 2003 Elsevier Science Ltd. All rights reserved.
doi:10.1016/S0960-894X(03)00384-6



1 X = OMe, Y = OH, Z = CO_2H
2 X = OMe, Y = OCH_2CO_2H, Z = CH_2OH
3 X = OH, Y = OCH_2CO_2H, Z = CH_2OMe
4 X = OCH_2CO_2H, Y = OMe, Z = CH_2OH

5 X = OMe, Y = OCH_2CO_2H, Z = CH_2OH
6 X = OH, Y = OCH_2CO_2H, Z = CH_2OMe

Figure 1.

The scaffold design described above provides the potential for accessing a large diverse set of scaffolds that could be used to explore a wide range of three-dimensional space. We envisioned that families of these scaffolds, would find utility in mapping pharmacophore recognition sites on target biomolecules and hence we refer to these libraries a carbohydrate-based universal pharmcophore mapping libraries. In this report we present the synthesis of a family of 3-azido containing monosaccharide scaffolds **1**, **2**, **3**, **4**, **5**, and **6** and demonstrate the construction of a combinatorial library using scaffold **2**.

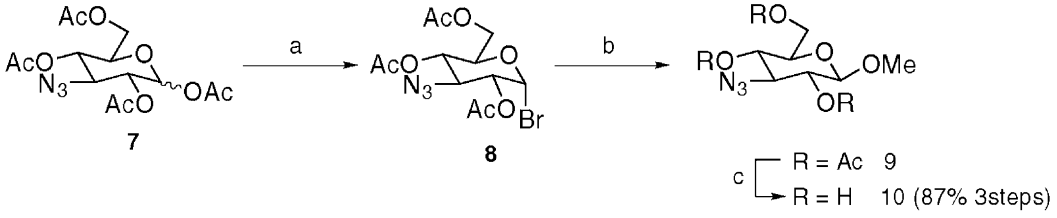
The syntheses of the 3-azido scaffolds **1**, **2**, **3**, **4**, **5**, and **6** began with construction of the common intermediate methyl 3-azido-3-deoxy-β-D-glucopyranoside **10**. Intermediate **10** would allow us to construct scaffold systems having a carboxylic acid unit or a free hydroxyl group in the 2-, 4- or 6-position of a glucose or galactose nucleus. For the synthesis of **10**, we employed 1,2,4,6-tetra-*O*-acetyl-3-azido-3-deoxy-D-glucopyranose (**7**) which was prepared from 1,2-5,6-di-*O*-isopropylidene-α-D-allofuranose using a previously described procedure for the preparation of 3-azido-3-deoxy-D-mannose.[6] Treatment of **7** with Me_3SiBr and $BiBr_3$[7] in CH_2Cl_2 gave crude bromide **8**. Treatment of **8** with MeOH–CH_2Cl_2 in the presence of Ag_2O followed by de-*O*-acetylation with methanolic sodium methoxide afforded **10** in 87% yield after chromatographic purification on silica gel. The ^1H NMR spectrum [δ 4.25 (d, $J = 7.5$ Hz, H-1)] as well as ^{13}C NMR spectrum [δ 105.23 (C-1)] confirmed the formation of β-linkage at C-1 (Scheme 1).

The synthesis of scaffold **1** from intermediate **10**, required that we oxidize the C-6 hydroxyl group to the corresponding carboxylic acid in the presence of the other functionality (Scheme 2). This was accomplished by first protecting the C-4 and C-6 hydroxyl groups as a 4,6-*O*-*p*-methoxybenzylidene cyclic acetal using anisaldehyde dimethyl acetal and *p*-toluenesulfonic acid monohydrate in DMF to give **11**. Acetylation of the free hydroxyl group followed by reductive ring opening[8] gave **13** in 96% yield.

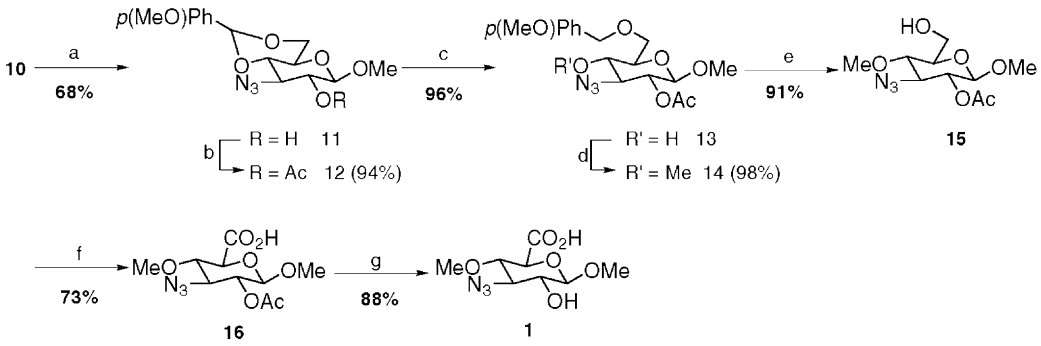
With **13** providing the needed azido sugar with each of the hydroxyl groups differentiated we were able to proceed with completing the synthesis of scaffold **1**. This was done by first methylating the C-4 hydroxyl group of **13** and removing the 6-*O*-*p*-methoxybenzyl group with DDQ.[9] Oxidation with Jones reagent[10] followed by de-*O*-acetylation produced compound **1** in 88% yield after silica gel column chromatography.

Scaffolds containing a carboxylate functionality attached to either the 2- or 4-position of the sugar nucleus required that we introduce a carboxymethyl substituent. In each case, we were able to accomplish the attachment of this carboxymethyl group by alkylation of a free hydroxyl group with methyl 2-bromoacetate and Ag_2O-KI. For the synthesis of scaffold **2** containing a carboxymethyl group at C-2, we began with compound **11** having a free hydroxyl at C-2. Alkylation with methyl 2-bromo-acetate gave **17** in 80% yield (Scheme 3). Conversion of the 4,6-benzylidene derivative **17** to the 4-*O*-methyl-6-hydroxy derivative **20** was accomplished as previously described for the preparation of compound **15**. Hydrolysis of methyl ester in compound **20** provide scaffold **2** in 94% yield.

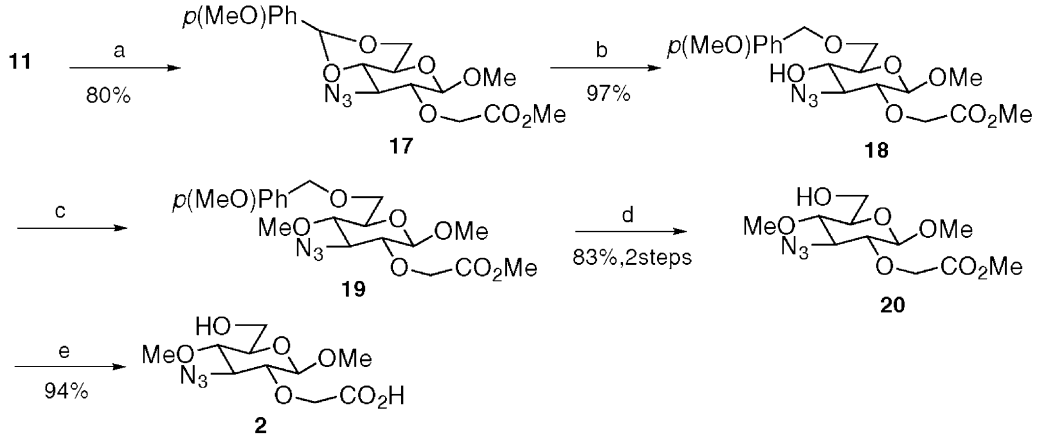
The synthesis of the 2,3,6-substituted scaffold **3** began with previously prepared intermediate **17** (Scheme 4); however, unlike in the preparation of scaffold **2**, here we



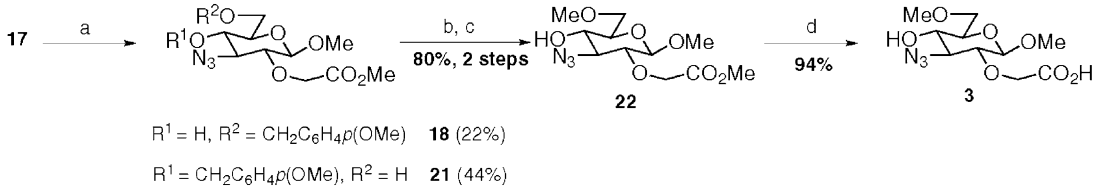
Scheme 1. Reagents and conditions: (a) Me_3SiBr, $BiBr_3$, CH_2Cl_2, 4 h; (b) Ag_2O, $CaSO_4$, CH_2Cl_2, MeOH, 1 h; (c) MeOH, MeONa, 4 h.



Scheme 2. Reagents and conditions: (a) MeO–C_6H_4–$CH(OMe)_2$, p-TSA, DMF, 4 h; (b) C_5H_5N, Ac_2O, 4 h; (c) $NaBH_3CN$, DMF, TFA, 3 Å molecular sieves, 48 h; (d) Ag_2O, CH_3I, DMF, 16 h; (e) DDQ, CH_2Cl_2 saturated with water, 2 h; (f) Jones reagent, acetone, 1.5 h; (g) MeOH–MeONa, 4 h.



Scheme 3. Reagents and conditions: (a) Ag_2O, $BrCH_2COOMe$, KI, DMF, 16 h; (b) $NaBH_3CN$, DMF, TFA, 3 Å molecular sieves, 48 h; (c) Ag_2O, CH_3I, DMF, 16 h; (d) DDQ, CH_2Cl_2 saturated with water, 2 h; (e) Dioxane, 2N NaOH, 1 h.

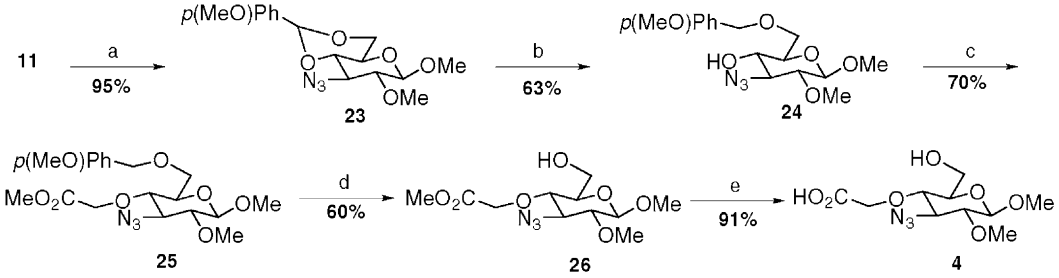


Scheme 4. Reagents and conditions: (a) $NaBH_3CN$, Me_3SiCl, 3 Å molecular sieves, CH_3CN, 4 h; (b) **21**, Ag_2O, CH_3I, DMF, 16 h; (c) DDQ, CH_2Cl_2 saturated with water, 2 h; (d) dioxane, 2N NaOH, 1 h.

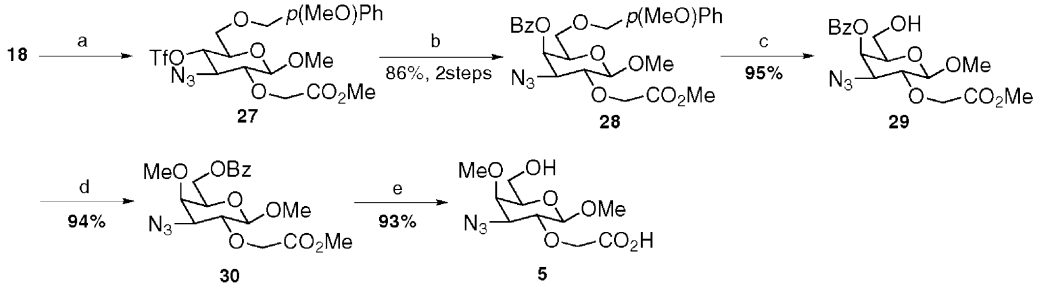
needed to achieve acetal ring opening that would favor *p*-methoxybenzyl protection on the C-4 hydroxyl. This was accomplished by treating compound **17** with Me_3SiCl–$NaBH_3CN$[11] in acetonitrile. These conditions provide the 6-*O*-*p*-methoxybenzyl derivative **18** and 4-*O*-*p*-methoxybenzyl derivative **21** in 1:2 ratio. The C-6 hydroxyl group was then methylated using Ag_2O and methyl iodide and the *p*-methoxybenzyl group was removed under oxidative conditions. Hydrolysis of the methyl ester gave scaffold **3** in 57% overall yield.

For the preparation of scaffold **4**, we needed to differentiate the C-2, C-4, and C-6 hydroxyl groups as we had done in the preparation of scaffold **1**. Therefore, we again used intermediate **11** and employed the regioselective *p*-methoxybenzylidene ring opening protocol (Scheme 5). Starting with **11**, treatment with CH_3I-NaH in DMF gave the C-2 methyl ether **23** in 95% yield. Acetal ring opening and attachment of the carboxymethyl group gave **25**. Removal of the C-6 hydroxyl protecting group and ester hydrolysis gave scaffold **4** in 38% overall yield.

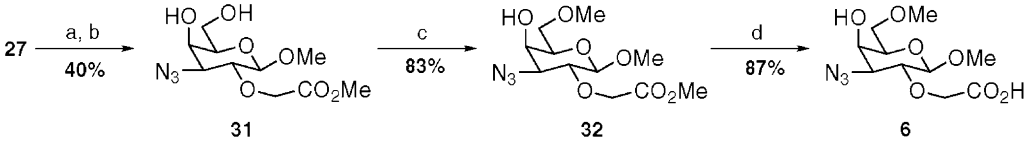
Synthesis of galactose scaffolds **5** and **6** from the glucopyranosyl intermediate **18** required inverting the stereochemistry at C-4. This inversion was accomplished through intermediacy of the C-4 triflate **27** which was obtained from **18** on treatment with triflic anhydride in dichloromethane (Scheme 6). Nucleophilic displacement of the C-4 triflate with tetrabutyl-ammonium benzoate in toluene gave the axial benzoate **28** in 86% yield. Removal of the *p*-methoxybenzyl protecting group gave **29**. We next wanted to methylate the C-6 hydroxyl group; however, simultaneous transfer of the benzoate group from the C-4–C-6 hydroxyl group and methylation of the C-4 hydroxyl occurred on exposure



Scheme 5. Reagents and conditions: (a) NaH, DMF, CH_3I, 1 h; (b) $NaBH_3CN$, DMF, TFA, 3 Å molecular sieves, 48 h; (c) Ag_2O, $BrCH_2COOMe$, KI, DMF, 4 h; (d) DDQ, CH_2Cl_2 saturated with water, 2 h; (e) Dioxane, 2N NaOH,1 h.



Scheme 6. Reagents and conditions: (a) Triflic anhydride, pyridine, CH_2Cl_2, 0 °C, 2 h; (b) Tetrabutylammonium benzoate, toluene, 1 h; (c) DDQ, CH_2Cl_2 saturated with water, 2 h; (d) Ag_2O, CH_3I, DMF, 16 h; (e) Dioxane, 2N NaOH, 1 h.

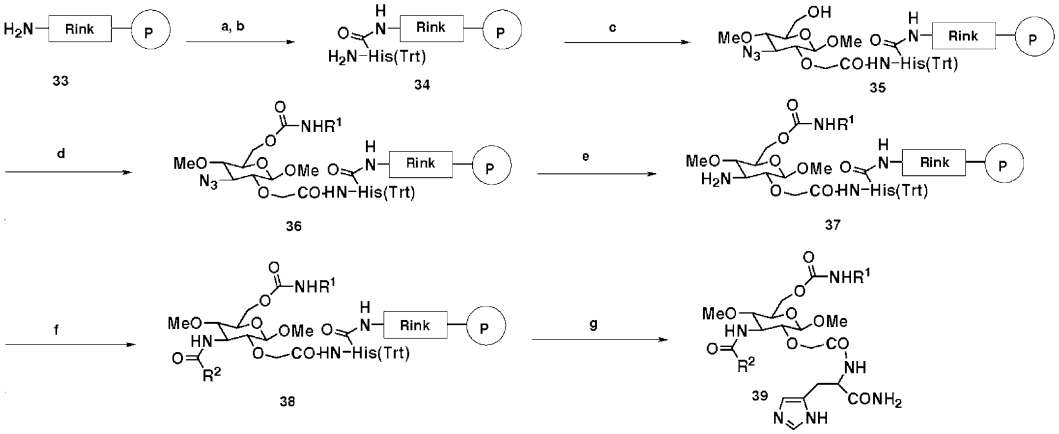


Scheme 7. Reagents and conditions: (a) Triflic anhydride, pyridine, CH_2Cl_2, 0 °C, 2 h; $NaNO_2$, DMF, 4 h; (b) 95% aq. TFA, CH_2Cl_2, 2 h; (c) Trimethyloxonium tetrafluoroborate, 2,6-di-*tert*-butyl-4-methyl-pyridine, CH_2Cl_2, 16 h; (d) Dioxane, 2 N NaOH, 1 h.

to Ag_2O/CH_3I in DMF providing compound **30** in 94% yield. With compound **30** in hand, base hydrolysis gave scaffold **5** in 93% yield (Scheme 6).

To access scaffold **6**, triflate **27** was treated with sodium nitrite[12] in DMF providing the desired C-4 axial hydroxyl group (Scheme 7). Subsequent removal of the C-6 *p*-methoxybenzyl group with TFA–CH_2Cl_2 provided diol **31** in 40% yield after silica gel column chromatography. Selective methylation of **31** with trimethyloxonium tetrafluoroborate-2,6-di*tert*butyl-4-methylpyridine[13] gave the 6-*O*-methyl derivative **32**. The hydrolysis of methyl ester provided **6** in 87% yield.

To demonstrate the use of a 3-azido scaffold in the construction of a library of substituted monosaccharides we chose scaffold **2**. Using Rink amide resin[14] as the solid support, a blocked amino acid,



Scheme 8. Reagents and conditions: (a) (i) 20% piperidine, DMF, 0.5 h; (ii) Fmoc histidine (trt), HATU, DIEPA, DMF, 16 h; (b) 20% piperidine, DMF, 0.5 h; (c) **2**, HATU, DIEPA, DMF, 16 h; (d) isocyanate, THF, Et_3N, 6–16 h or isocyanate, DMF, CuCl; (e) Me_3P, THF/EtOH/H_2O(4/4/1); (f) carboxylic acid, HATU, DIEPA, DMF, 16 h; (g) 20% TFA, CH_2Cl_2, 2% Et_3SiH.

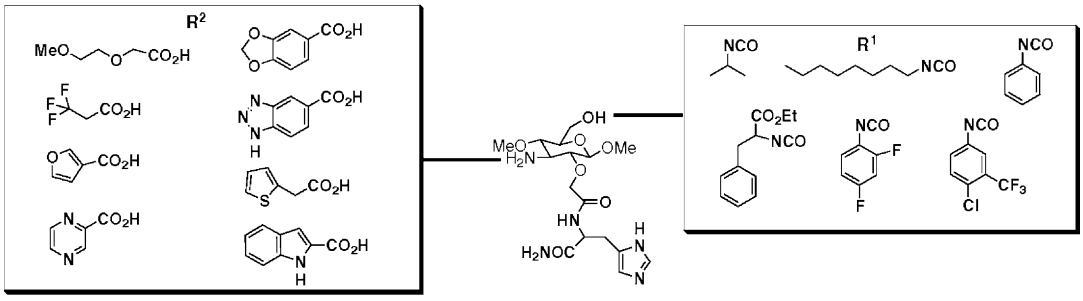
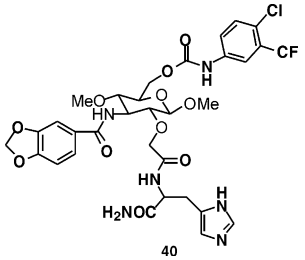


Figure 2.

Fmoc-histidine (Trt), was first coupled to the support (Scheme 8). Removal of the Fmoc protecting group was then followed by coupling of scaffold **2** to the free amino group by amide bond formation to the C-2 carboxymethyl group of **2**. The resin-bound scaffold was then treated with a series of isocyanates in the presence of CuCl in DMF to generate the corresponding C-6 carbamates. Reduction of the C-3 azido group with trimethylphosphine generated the desired amines which were then reacted with a series of carboxylic acids to provide the corresponding amides.

A library of 48 compounds was prepared using the directed sorting mix-and-split synthesis method[15] (Fig. 2).

All 48 compounds were analyzed by LC/MS using evaporative light-scattering detection (ELSD). The yield of each product was determined from the ELSD trace using standard curves and product identity was determined by electrospray mass spectal analysis. The average yield for the six-step sequence was 55%. One of the library products was further purified by preparative HPLC and its structure was confirmed by NMR spectroscopy and LC/MS analysis to be the desired compound **40**.[16]



40

In conclusion, we believe that carbohydrate scaffolds are biologically relevant molecular platforms that can be used in combinatorial library strategies to identify unique ligands for a wide variety of biomolecular drug targets. To exploit this potential, we have developed a series of 3-azido glycopyranosides.

References and Notes

1. Hirschmann, R.; Nicolaou, K. C.; Pietranico, S.; Leahy, E. M.; Salvino, J.; Arison, B.; Cichy, M. A.; Spoors, P. G.; Shakespear, W. C.; Sprengeler, P. A.; Hamley, P.; Smith, A. B., III; Reisine, T.; Raynor, K.; Maechler, L.; Donaldson, C.; Vale, W.; Freidinger, R. M.; Cascieri, M. R.; Strader, C. D. *J. Am. Chem. Soc.* **1993**, *115*, 12550.
2. Hirschmann, R; Yao, W.; Cascieri, M. A.; Strader, C. D.; Maechler, L.; Cichy-Knight, M. A.; Hynes, J., Jr; van Rijn, R. D.; Sprengeler, P. A.; Smith, A. B., III *J. Med. Chem.* **1996**, *39*, 2441.
3. Wunberg, T.; Kallus, C.; Opatz, T.; Henke, S.; Schmidt, W.; Kunz, H. *Angew. Chemie. Int. Ed. Engl.* **1998**, *37*, 2503.
4. Sofia, M. J.; Hunter, R.; Chan, T. Y.; Vaughan, A.; Dulina, R.; Wang, H.; Gange, D. *J. Org. Chem.* **1998**, *63*, 2802.
5. Sofia, M. *J. Med. Chem. Res.* **1998**, *8*, 362.
6. Holla, E. W.; Sinnwell, V.; Klaffke, W. *Synlett* **1992**, *5*, 413.
7. Montero, J.-L.; Winum, J.-Y.; Leydet, A.; Kamal, M.; Pavia, A. A.; Roque, J.-P. *Carbohydr. Res.* **1997**, *297*, 175.
8. Johansson, R.; Samuelsson, B. *J. Chem. Soc., Chem. Commun.* **1984**, 201.
9. Oikawa, Y.; Yshioka, T.; Yonemitsu, O. *Tetrahedron Lett.* **1982**, *23*, 885.
10. Allanson, N. M.; Liu, D.; Chi, F.; Jain, R. K.; Chen, A.; Ghosh, M.; Hong, L.; Sofia, M. J. *Tetrahedron Lett.* **1998**, *39*, 1889.
11. Johansson, R.; Samuelsson, B. *J. Chem. Soc., Perkin Trans. 1* **1994**, 2317.
12. El-Sokkary, R. I.; Silwanis, B. A.; Nashed, M. A. *Carbohydr. Res.* **1990**, *203*, 319.
13. Fugedi, P.; Kovac, J. XV International Carbohydrate Symposium, Aug. 12–17, 1990, p 77.
14. Purchased from Novabiochem.
15. Radiofrequency tagging system. Nicolaou, K. C.; Xiao, X. Y.; Parandoosh, Z.; Senyei, A.; Nova, M. P. *Angew. Chem. Int. Ed. Engl.* **1995**, *34*, 2289.
16. NMR diagnostic signals at δ 8.04 (d, 1H), 7.39 (s, 1H), 6.07 (s, 2H), 4.48 (d, 1H, $J = 7.5$ Hz), 3.73 (s, 3H), 3.70 (s, 3H).